

COMPILATION OF THE LITERATURE REPORTS FOR THE SCREENING OF VASCULAR PLANTS, ALGAE, FUNGI AND NON-ARTHROPOD INVERTEBRATES FOR THE PRESENCE OF ECDYSTEROIDS

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★NEW! This version contains not only the latest relevant references, but also two new developments:

- Plant species are now also allocated to Families according to the molecular plant classification (APGIV, PPG1 etc.)
- Where possible, Abstracts have been added to the reference lists.

Important notice:

This database has been designed as a tool to help the scientific community in research on ecdysteroids. The authors wish it to be an evolving system and would encourage other researchers to submit new data, additional publications, proposals for modifications or comments to the authors for inclusion. All new material will be referenced to its contributor.

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Please notify Laurie Dinan (dinanlaurence@gmail.com) of any errors or additional literature sources.

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1. VASCULAR PLANTS

1a. Table of the occurrence of phytoecdysteroids according to species

Notes

Species are listed as designated in the original publications.

Vascular plant species are allocated to families according to Brummitt R.K. "Vascular plant families and genera" Royal Botanic Gardens, Kew, U.K. (1992) and (in square brackets) according to the new molecular classification (Angiosperms: APGIV [2016]; Lycophytes and Ferns: Smith et al. [2006], PPG1 (2016); Gymnosperms: Christenhusz et al. [2010]).

APGIV (2016) Botanical Journal of the Linnean Society 181, 1-20.

Christenhusz M.J.M. et al. (2010) Phytotaxa 19, 55-70.

Smith A.R. et al. (2006) Taxon 55(3), 705-731.

PPG1 (2016) Journal of Systematics and Evolution 54(6), 563-603.

The presence (+) or absence (-) of ecdysteroids are indicated in the +/- column. (+) indicates weakly positive. Where numbers appear in this column, data derive from the paper of Wong *et al.* (1979) and refer to the shortening (in hours) of the 5th (last) larval instar of *Bombyx mori* after application of ethanolic extracts of plants to the mulberry leaves fed to the insects. According to the authors, a shortening of the 5th instar by >5 hrs is regarded as positive.

Abbreviations

FA = Fern Ally; F = Fern; G = Gymnosperm; D = Dicotyledon; M = Monocotyledon; E = Eudicot; Mag = Magnolids; C = Ceratophyllales; Cyc = Cycadophyta; Equ – Equisetidae; Gin = Ginkgoales; Nym = Nymphaeales; Chl = Chloranthales; Lyc = Lycophyte

The family Leguminosae is subdivided into 3 sub-families; C: Caesalpinoideae, M: Mimosoideae and P: Papilionoideae.

SPECIES	FAMILY	+/-	REFERENCE
A			
<i>Abacopteris triphylla</i> (<i>Cyclosorus triphyllus</i>) (syn. <i>Pronephrium triphyllum</i>)	Thelypteridaceae (F) [Thelypteridaceae (F)]	+	Yen <i>et al.</i> (1974)
<i>Abelmoschus esculentum</i>	Malvaceae (D) [Malvaceae (E)]	- -	Dinan <i>et al.</i> (2020a) Dinan <i>et al.</i> (2020b)
<i>Abies firma</i> <i>A. sibirica</i>	Pinaceae (G) [Pinaceae (G)]	+ -	Takemoto <i>et al.</i> (1967c) Volodin <i>et al.</i> (2002)
<i>Abuta velutina</i>	Menispermaceae (D) [Menispermaceae (E)]	+	Pinheiro <i>et al.</i> (1983)
<i>Acacia melanoxylon</i>	Leguminosae-M. (D) [Fabaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Acaena fissistipula</i> <i>A. saccaticupula</i>	Rosaceae (D) [Rosaceae (E)]	- -	Dinan <i>et al.</i> (2001d) Dinan <i>et al.</i> (2020b)
<i>Acanthopanax</i> (<i>Eleutherococcus</i>) <i>gracilistylus</i> <i>A. sieboldiana</i>	Araliaceae (D) [Araliaceae (E)]	1.9 -	Wong <i>et al.</i> (1979) Takemoto <i>et al.</i> (1967c)
<i>Acanthophyllum gypsophylloides</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	+ + + +	Almagambetov <i>et al.</i> (2015) Tuleuov (2016) Tuleuov <i>et al.</i> (2018) Temirgaziev <i>et al.</i> (2019)
<i>Acer mono</i>	Aceraceae (D)	1.0	Wong <i>et al.</i> (1979)

<i>A. palmatum</i>	[Sapindaceae (E)]-		Dinan <i>et al.</i> (2020b)
<i>Achillea millefolium</i>	Compositae (D)	(+)	Volodin <i>et al.</i> (1993)
	[Asteraceae (E)]	-	Blackford & Dinan (1997a)
		(+)	Volodin <i>et al.</i> (2002)
<i>Achyranthes aspera</i>	Amaranthaceae (D)	+	Banerji <i>et al.</i> (1971)
	[Amaranthaceae (E)]	+	Banerji & Chadha (1971)
		+	Ikan <i>et al.</i> (1971)
		+	Ogawa <i>et al.</i> (1971)
		+	Sihra (1974)
		+	Chou & Lu (1980)
		+	Kunert <i>et al.</i> (2000)
		+	Laddha & Ghosh (2005)
		+	Sreejit (2014)
		+	Ezhava <i>et al.</i> (2016)
		+	Jaiswal <i>et al.</i> (2018)
		+	John <i>et al.</i> (2017)
		+	Lakshimi <i>et al.</i> (2017)
		+	John <i>et al.</i> (2018)
		+	Sreejit <i>et al.</i> (2018)
		+	Taralkar & Chattopadhyay (2018)
		+	Kamble (2018)
		+	Ou <i>et al.</i> (2018)
		+	Ouyang <i>et al.</i> (2020)
		+	Hu <i>et al.</i> (2020)
<i>A. bidentata</i>		+	Takemoto <i>et al.</i> (1967d)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
		+	Ogawa <i>et al.</i> (1971)
		16	Wong <i>et al.</i> (1979)
		+	Chou & Lu (1980)
		+	Yao & Hu (1989)
		+	Gao <i>et al.</i> (2000)
		+	Ma <i>et al.</i> (2000)
		+	Wang <i>et al.</i> (2003)
		+	Wang <i>et al.</i> (2004)
		+	Zhu <i>et al.</i> (2004)
		+	Meng <i>et al.</i> (2005)
		+	Wang <i>et al.</i> (2005)
		+	Bai <i>et al.</i> (2005)
		+	Lin <i>et al.</i> (2006)
		+	Li <i>et al.</i> (2007a)
		+	Li <i>et al.</i> (2007b)
		+	Li <i>et al.</i> (2007c)
		+	Zhao <i>et al.</i> (2007)
		+	Zheng <i>et al.</i> (2008)
		+	Li <i>et al.</i> (2010)
		+	Dong <i>et al.</i> (2010)
		+	Wang <i>et al.</i> (2011)
		+	Wu <i>et al.</i> (2011)
		+	Zhao <i>et al.</i> (2011)
		+	Zhang <i>et al.</i> (2012)
		+	Dat <i>et al.</i> (2012)
		+	Zhao <i>et al.</i> (2012)
		+	Ruan <i>et al.</i> (2012)
		+	Wang <i>et al.</i> (2012a)
		+	Wang <i>et al.</i> (2012b)
		+	Wang <i>et al.</i> (2013)
		+	Wei <i>et al.</i> (2013)

	+	Zhang <i>et al.</i> (2013a)
	+	Zhang <i>et al.</i> (2013b)
	+	Sreejit (2014)
	+	Liang <i>et al.</i> (2014)
	+	Luo <i>et al.</i> (2014)
	+	Li <i>et al.</i> (2014)
	+	Mu <i>et al.</i> (2014)
	+	Zhang <i>et al.</i> (2015)
	+	Zhang <i>et al.</i> (2015)
	+	Li <i>et al.</i> (2015)
	+	Pan <i>et al.</i> (2015)
	+	Yang <i>et al.</i> (2016)
	+	Du <i>et al.</i> (2016)
	+	Li <i>et al.</i> (2016)
	+	Zhang <i>et al.</i> (2016)
	+	Sun <i>et al.</i> (2017)
	+	Jiang <i>et al.</i> (2017)
	+	Jaiswal <i>et al.</i> (2017)
	+	Zhao <i>et al.</i> (2017)
	+	Tao <i>et al.</i> (2017)
	+	Sreejit <i>et al.</i> (2018)
	+	Li <i>et al.</i> (2018)
	+	Volodin <i>et al.</i> (2018)
	+	Hu <i>et al.</i> (2018)
	+	Zhang <i>et al.</i> (2018)
	+	Tao <i>et al.</i> (2019a)
	+	Tao <i>et al.</i> (2019b)
	+	Tang <i>et al.</i> (2019)
	+	Liang <i>et al.</i> (2019)
	+	Yang <i>et al.</i> (2019)
	+	Tu <i>et al.</i> (2019)
	+	Ju <i>et al.</i> (2020)
	+	Ouyang <i>et al.</i> (2020)
	+	Chen & Feng (2021)
<i>A. fauriei</i>	+	Hikino <i>et al.</i> (1975a)
	+	Takemoto <i>et al.</i> (1967c)
	+	Takemoto <i>et al.</i> (1967f)
	+	Takemoto <i>et al.</i> (1967g)
	+	Takemoto <i>et al.</i> (1967h)
	+	Takemoto <i>et al.</i> (1967k)
	+	Hikino & Hikino (1970)
	+	Hikino <i>et al.</i> (1971d)
	+	Ogawa <i>et al.</i> (1971)
	+	Tomita & Sakurai (1974)
	+	Hikino <i>et al.</i> (1975c)
	+	Jin <i>et al.</i> (1975)
	+	Hikino <i>et al.</i> (1976b)
<i>A. japonica</i>	+	Hikino <i>et al.</i> (1975a)
	+	Takemoto <i>et al.</i> (1967c)
	+	Imai <i>et al.</i> (1969d)
	+	Hikino <i>et al.</i> (1971d)
	+	Ogawa <i>et al.</i> (1971)
	+	Takemoto <i>et al.</i> (1967d)
	+	Matsuoka <i>et al.</i> (1969)
	+	Chae <i>et al.</i> (2001)
	+	Boo <i>et al.</i> (2010)
	+	Boo <i>et al.</i> (2013)
	+	Kim <i>et al.</i> (2015)
<i>A. japonica</i> var. <i>hachijoensis</i>	+	Ogawa <i>et al.</i> (1971)
	+	Hikino <i>et al.</i> (1971d)

<i>A. longifolia</i> (= <i>A. bidentata</i>)		+	Takemoto <i>et al.</i> (1967d)
		+	Takemoto <i>et al.</i> (1967j)
		+	Hikino <i>et al.</i> (1975a)
		+	Takemoto <i>et al.</i> (1967c)
		+	Ogawa <i>et al.</i> (1971)
		+	Ouyang <i>et al.</i> (2020)
<i>A. mollicula</i> (= <i>A. bidentata</i>)		+	Ogawa <i>et al.</i> (1971)
<i>A. obtusifolia</i> (= <i>A. aspera</i> var <i>indica</i>)		+	Hikino <i>et al.</i> (1975a)
		+	Takemoto <i>et al.</i> (1967i)
		+	Takemoto <i>et al.</i> (1967j)
		+	Hikino & Hikino (1970)
		+	Hikino <i>et al.</i> (1971d)
		+	Ogawa <i>et al.</i> (1971)
<i>A. ogatai</i> (= <i>A. bidentata</i>)		+	Ogawa <i>et al.</i> (1971)
<i>Achyranthes</i> Radix (pharm. prep)		+	Ogawa <i>et al.</i> (1977)
<i>A. rubrofusca</i> (= <i>A. aspera</i> var <i>rubrofusca</i>)		+	Hikino <i>et al.</i> (1975a)
		+	Takemoto <i>et al.</i> (1967c)
		+	Takemoto <i>et al.</i> (1967j)
		+	Takemoto <i>et al.</i> (1969b)
		+	Hikino <i>et al.</i> (1971d)
		+	Ogawa <i>et al.</i> (1971)
<i>Acnistus</i> (<i>Dunalia</i>) <i>australis</i> (syn. <i>Eriolarynx australis</i> , <i>Iochroma australe</i>) [Solanaceae (E)]	Solanaceae (D)	-	Savchenko <i>et al.</i> (2000)
<i>Aconitum anthora</i>	Ranunculaceae (D)	-	Dinan <i>et al.</i> (2002a)
<i>A. carmichaelii</i>	[Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2002a)
<i>A. columbianum</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. eulophum</i>		-	Volodin <i>et al.</i> (2002)
<i>A. loczyanum</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. napellus</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. napellus</i> var. <i>carneum</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. orientale</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. septentrionale</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. volubile</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. vulparia</i>		-	Dinan <i>et al.</i> (2020b)
<i>Acrocarpus fraxinifolius</i>	Leguminosae-C. (D) [Fabaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Acroglochis chenopodioides</i>	Chenopodiaceae (D)	-	Dinan <i>et al.</i> (1998)
<i>A. persicarioides</i>	[Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Acrophorus stipellatus</i>	Dryopteridaceae (F) [Dryopteridaceae (F)]	+	Yen <i>et al.</i> (1974)
		+	Hikino <i>et al.</i> (1973)
<i>Acroptilon repens</i> (syn. <i>Rhaponticum repens</i>)	Compositae (D) [Asteraceae (E)]	+	Volodin <i>et al.</i> (2002)
<i>Acrostichum aureum</i>	Pteridaceae (F)	+	Bergamasco & Horn (1983)
<i>A. speciosum</i>	[Pteridaceae (F)]	+	Bergamasco & Horn (1983)
<i>Actaea alba</i>	Ranunculaceae (D)	-	Dinan <i>et al.</i> (2002a)
<i>A. arguta</i>	[Ranunculaceae (E)]	(+)	Dinan <i>et al.</i> (2002a)
<i>A. asiatica</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. erythrocarpa</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. pachypoda</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. rubra</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. spicata</i>		-	Dinan <i>et al.</i> (2002a)

		-	Volodin <i>et al.</i> (2002)
<i>Actinidia arguta</i>	Actinidiaceae (D)	-	Takemoto <i>et al.</i> (1967c)
	[Actinidiaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>A. deliciosa</i>		-	Dinan <i>et al.</i> (2020a)
<i>A. polygama</i>		-	Takemoto <i>et al.</i> (1967c)
<i>Adiantum aethiopicum</i>	Adiantaceae (F)	+	Russell & Fenemore (1971)
<i>A. capillus-veneris</i>	[Pteridaceae (F)]	+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
		-	Yen <i>et al.</i> (1974)
		+	Hikino <i>et al.</i> (1973)
<i>A. palmiformis</i>		-	Hikino <i>et al.</i> (1973)
<i>A. caudatum</i>		-	Yen <i>et al.</i> (1974)
<i>A. cunninghamii</i>		+	Russell & Fenemore (1971)
<i>A. diaphanum</i>		-	Yen <i>et al.</i> (1974)
<i>A. flabellulatum</i>		+	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
<i>A. formosum</i>		-	Russell & Fenemore (1971)
<i>A. hispidulum</i>		+	Russell & Fenemore (1971)
		+	Yen <i>et al.</i> (1974)
<i>A. monochlamys</i>		+/-	Hikino <i>et al.</i> (1973)
<i>A. pedatum</i>		+/-	Hikino <i>et al.</i> (1973)
		-	Dreier (1987)
<i>A. philippense</i>		+	Yen <i>et al.</i> (1974)
<i>Adoxa moschatellina</i>	Adoxaceae (D)	+	Matsuoka <i>et al.</i> (1969)
	[Adoxaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Aegopodium podagraria</i>	Umbelliferae (D)	-	Volodin <i>et al.</i> (2002)
	[Apiaceae (E)]		
<i>Aerva lanata</i>	Amaranthaceae (D)	+	Baltaev <i>et al.</i> (1992)
	[Amaranthaceae (E)]	-	Sreejit (2014)
<i>Aerva japonica</i>		+	Saleem <i>et al.</i> (2013)
<i>A. tomentosa</i>		+	Hardman & Mahar (1978)
<i>Aesculus chinensis</i>	Hippocastanaceae (D)	3.0	Wong <i>et al.</i> (1979)
	[Sapindaceae (E)]		
<i>Agapanthus africanus</i>	Alliaceae (M)	+	Savchenko <i>et al.</i> (1997)
<i>A. campanulatus</i>	[Amaryllidaceae (M)]	+	Savchenko <i>et al.</i> (1997)
<i>A. caulescens</i>		(+)	Savchenko <i>et al.</i> (1997)
<i>A. coddii</i>		+	Savchenko <i>et al.</i> (1997)
<i>A. comptonii</i>		+	Savchenko <i>et al.</i> (1997)
<i>A. inapertus</i>		+	Savchenko <i>et al.</i> (1997)
<i>A. minor</i>		-	Savchenko <i>et al.</i> (1997)
<i>A. praecox</i>		+/-	Savchenko <i>et al.</i> (1997)
		+	Dinan <i>et al.</i> (2020b)
<i>Agastache foeniculum</i>	Labiatae (D)	(+)	Dinan <i>et al.</i> (2001d)
	[Lamiaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Agathis australis</i>	Araucariaceae (G)	-	Russell & Fenemore (1970)
<i>A. loranthifolia</i>	[Araucariaceae (G)]	-	Hoffmeister <i>et al.</i> (1967)
<i>Agave</i> sp.	Agavaceae (M)	+	Saeng-ngam <i>et al.</i> (on-line)
	[Asparagaceae (M)]		
<i>Aglaonema modestum</i>	Araceae (M)	-	Volodin <i>et al.</i> (2018)

	[Araceae (M)]		
<i>Agrimonia striata</i>	Rosaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>A. gyrosepala</i>	[Rosaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Agriophyllum minus</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Agrostemma githago</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>A. gracile</i>		-	Zibareva <i>et al.</i> (2003)
		+	Bespayeva <i>et al.</i> (2012)
		+	Bespayeva <i>et al.</i> (2012)
<i>Agrostis nebulosa</i>	Gramineae (M)	-	Dinan (1995a)
<i>A. stolonifera</i>	[Poaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Ailanthus altissima</i>	Simaroubaceae (D) [Simaroubaceae (E)]	5.7 +	Wong <i>et al.</i> (1979) Chou & Lu (1980)
<i>Ajuga</i> spp.	Labiatae (D)	+	Coll Toledano (1998)
<i>Ajuga australis</i>	[Lamiaceae (E)]	+	Darvas (1991)
		+	Bergamasco & Horn (1983)
		+	Saatov <i>et al.</i> (1994)
		+	Ramazanov (2005)
<i>A. bracteosa</i>		+	Matsuoka <i>et al.</i> (1969)
		+	Darvas (1991)
		+	Fekete <i>et al.</i> (2004)
		+	Ramazanov (2005)
		+	Kayani <i>et al.</i> (2014)
		+	Kayani <i>et al.</i> (2016a)
		+	Kayani <i>et al.</i> (2016b)
		+	Kayani <i>et al.</i> (2017)
<i>A. chamaecistus</i>		+	Kubo <i>et al.</i> (1983)
		+	Darvas (1991)
<i>A. chamaecistus</i> ssp. <i>tomentella</i>		+	Sadati <i>et al.</i> (2012)
		+	Najafi <i>et al.</i> (2015)
		+	Sadati <i>et al.</i> (2016)
		+	Khanavi <i>et al.</i> (2017)
<i>A. chamaepitys</i> (syn. <i>A. chia</i>)		+	Camps <i>et al.</i> (1984)
		+	Darvas (1991)
		+	Saatov <i>et al.</i> (1994)
		+	Fekete <i>et al.</i> (2004)
		+	Ramazanov (2005)
		+	Taha-Salaime <i>et al.</i> (2019)
<i>A. chia</i> (syn. <i>A. chamaepitys</i>)		+	Darvas (1991)
		+	Ikan & Ravid (1971)
		+	Usmanov <i>et al.</i> (1973b)
		+	Gorovits <i>et al.</i> (1974)
		+	Abubakirov (1982)
		+	Abubakirov (1984)
		+	Saatov <i>et al.</i> (1994)
		+	Ramazanov (2005)
<i>A. ciliata</i>		+	Matsuoka <i>et al.</i> (1969)
		+	Saatov <i>et al.</i> (1994)
		+	Ramazanov (2005)
<i>A. decumbens</i>		+	Imai <i>et al.</i> (1969b)
		+	Imai <i>et al.</i> (1969c)
		+	Imai <i>et al.</i> (1969d)
		+	Darvas (1991)

	+	Matsuoka <i>et al.</i> (1969)
	+	Saatov <i>et al.</i> (1994)
	+	Takasaki <i>et al.</i> (1999)
	+	Ramazanov (2005)
	+	Chen <i>et al.</i> (2018)
<i>A. forrestii</i>	+	Chen <i>et al.</i> (2017)
<i>A. genevensis</i>	+	Fekete <i>et al.</i> (2004)
	+	Ramazanov (2005)
<i>A. incisa</i>	+	Matsuoka <i>et al.</i> (1969)
	+	Imai <i>et al.</i> (1969a)
	+	Imai <i>et al.</i> (1969c)
	+	Imai <i>et al.</i> (1969d)
	+	Darvas (1991)
	+	Ramazanov (2005)
<i>A. iva</i>	+	Sabri <i>et al.</i> (1981)
	+	Ikan and Ravid (1970)
	+	Wessner <i>et al.</i> (1992)
	+	Ghedira <i>et al.</i> (1991)
	+	Darvas (1991)
	+	Saatov <i>et al.</i> (1994)
	+	Ramazanov (2005)
	+	Coll (2007)
	+	Bakrim <i>et al.</i> (2014)
<i>A. japonica</i>	+	Taha-Salaime <i>et al.</i> (2019)
	+	Imai <i>et al.</i> (1969b)
	+	Darvas (1991)
	+	Matsuoka <i>et al.</i> (1969)
	+	Saatov <i>et al.</i> (1994)
	+	Ramazanov (2005)
<i>A. laxmanii</i>	+	Malakov <i>et al.</i> (1998)
	+	Ramazanov (2005)
<i>A. linearifolia</i>	+	Ramazanov (2005)
<i>A. lobata</i>	+	Li <i>et al.</i> (2013)
	+	Sun <i>et al.</i> (2015)
	+	Qian <i>et al.</i> (2015)
	+	Qian <i>et al.</i> (2016)
	+	Yany <i>et al.</i> (2017)
	+	Wang <i>et al.</i> (2018)
<i>A. macrosperma</i> var. <i>breviflora</i>	+	Castro <i>et al.</i> (2008)
<i>A. multiflora</i>	+	Darvas (1991)
	11	Wong <i>et al.</i> (1979)
	+	Chou & Lu (1980)
	+	Saatov <i>et al.</i> (1994)
	+	Kim <i>et al.</i> (2005)
	+	Ramazanov (2005)
	+	Chi <i>et al.</i> (2002)
	+	Sun <i>et al.</i> (2015)
	+	Wang <i>et al.</i> (2018)
<i>A. nipponensis</i>	+	Imai <i>et al.</i> (1969c)
	+	Imai <i>et al.</i> (1969d)
	+	Matsuoka <i>et al.</i> (1969)
	+	Chou & Lu (1980)
	+	Anon (1981)
	+	Darvas (1991)
	+	Saatov <i>et al.</i> (1994)
	+	Zeng <i>et al.</i> (2000)
	+	Ramazanov (2005)
	+	Coll <i>et al.</i> (2007)
	+	He <i>et al.</i> (2013)
	+	Liang <i>et al.</i> (2014)

			+	Zakirova <i>et al.</i> (1998)
			+	Saatov <i>et al.</i> (1999)
			+	Zakirova & Malikova (2000)
			+	Zakirova & Malikova (2001)
			+	Ramazanov (2005)
			+	Abdukadirov <i>et al.</i> (2005)
			+	Cheng <i>et al.</i> (2008)
			+	Vanyolos (2012)
			+	Mamadaliyeva <i>et al.</i> (2014)
			+	Guibout <i>et al.</i> (2015)
<i>Akebia quintata</i>	Lardizabalaceae (D) [Lardizabalaceae (E)]		+	Takemoto <i>et al.</i> (1967c)
		5.2		Wong <i>et al.</i> (1979)
<i>Alangium chinense</i>	Alangiaceae (D)		1.0	Wong <i>et al.</i> (1979)
<i>A. platanifolium</i>	[Cornaceae (E)]		-	Takemoto <i>et al.</i> (1967c)
<i>Alcea pallida</i>	Malvaceae (D) [Malvaceae (E)]		(+)	Dinan <i>et al.</i> (2001d)
<i>Alchemilla murbeckiana</i>	Rosaceae (D) [Rosaceae (E)]		-	Volodin <i>et al.</i> (2002)
<i>Aleuritopteris (Cheilanthes) argentea</i>	Adiantaceae (F) [Pteridaceae (F)]		+	Hikino <i>et al.</i> (1973)
<i>Alisma plantago-aquatica</i>	Alismataceae (M) [Alismataceae (M)]		-	Volodin <i>et al.</i> (2002)
<i>Allenrolfia occidentalis</i>	Chenopodiaceae (D) [Amaranthaceae (E)]		-	Dinan <i>et al.</i> (1998)
<i>Alliaria petiolata</i>	Cruciferae (D) [Brassicaceae (E)]		-	Dinan <i>et al.</i> (2020b)
<i>Allium albidum</i>	Alliaceae (M)		-	Dinan <i>et al.</i> (2001d)
<i>A. ampeloprasum</i>	[Amaryllidaceae (M)]		-	Dinan <i>et al.</i> (2020b)
<i>A. canadense</i>			-	Dinan <i>et al.</i> (2020b)
<i>A. carinatum</i> ssp. <i>pulchellum</i>			-	Dinan <i>et al.</i> (2001d)
<i>A. cepa</i>			-	Dinan <i>et al.</i> (2020a)
<i>A. cepa ascalonicum</i>			-	Dinan <i>et al.</i> (2020a)
<i>A. fistulosum</i>			-	Blackford <i>et al.</i> (1996)
<i>A. montanum</i>			-	Dinan <i>et al.</i> (2001d)
<i>A. porrum</i>			-	Dinan <i>et al.</i> (2020a)
<i>A. sativum</i>			+	Matsuoka <i>et al.</i> (1969)
			-	Dinan <i>et al.</i> (2020a)
<i>A. schoenoprasum</i>			-	Volodin <i>et al.</i> (2002)
<i>Allmania nodiflora</i>	Amaranthaceae (D) [Amaranthaceae (E)]		-	Sreejit (2014)
<i>Alnus hirsuta</i>	Betulaceae (D)		+	Matsuoka <i>et al.</i> (1969)
<i>A. incana</i>	[Betulaceae (E)]		-	Volodin <i>et al.</i> (2002)
<i>A. serrulatooides</i>			+	Matsuoka <i>et al.</i> (1969)
<i>Alocasia cf. macrorrhiza</i>	Araceae (M) [Araceae (M)]		-	Volodin <i>et al.</i> (2018)
<i>Alopecurus aequalis</i>	Gramineae (M) [Poaceae (M)]		-	Volodin <i>et al.</i> (2002)

<i>Alstroemeria ligtu</i>	Alstroemeriaceae (M) [Alstroemeriaceae (M)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Alternanthera brasiliana</i>	Amaranthaceae (D)	-	Sreejit (2014)
<i>A. philoxcroides</i>	[Amaranthaceae (E)]	3.4	Wong <i>et al.</i> (1979)
		-	Sreejit (2014)
<i>A. sessilis</i>		+	Takemoto <i>et al.</i> (1967c)
		-	Sreejit (2014)
<i>A. tenella</i>		-	Sreejit (2014)
<i>Althaea officinalis</i>	Malvaceae (D) [Malvaceae (E)]	-	Blackford & Dinan (1997a)
<i>Amaranthus blitum</i>	Amaranthaceae (D)	+	Bespayeva <i>et al.</i> (2012)
<i>A. caudatus</i>	[Amaranthaceae (E)]	+	Bespayeva <i>et al.</i> (2012)
		-	Dinan <i>et al.</i> (2020b)
<i>A. cruentus</i>		+	Bespayeva <i>et al.</i> (2012)
<i>A. gangeticus</i>		+	Bespayeva <i>et al.</i> (2012)
		-	Dinan <i>et al.</i> (2020b)
<i>A. hybridus</i>		+	Bespayeva <i>et al.</i> (2012)
<i>A. hypochondriacus</i>		+	Kiran <i>et al.</i> (2012)
<i>A. indica</i>		+	Bratoeff <i>et al.</i> (1996)
<i>A. lividus</i>		-	Volodin <i>et al.</i> (2018)
<i>A. magnostanus</i>		1.0	Wong <i>et al.</i> (1979)
		+	Takemoto <i>et al.</i> (1967c)
<i>A. paniculatus</i>		+	Bespayeva <i>et al.</i> (2012)
<i>A. powellii</i>		+	Bespayeva <i>et al.</i> (2012)
<i>A. spinosus</i>		+	Takemoto <i>et al.</i> (1967c)
		1.7	Wong <i>et al.</i> (1979)
		-	Sreejit (2014)
<i>A. patulus</i>		+	Takemoto <i>et al.</i> (1967c)
<i>A. viridis</i>		+	Takemoto <i>et al.</i> (1967c)
		+	Saeng-ngam <i>et al.</i> (on-line)
<i>Ambrosia sp.</i>	Compositae (D) [Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Amischotolype mollissima</i>	Commelinaceae (M) [Commelinaceae (M)]	+	Crouzet <i>et al.</i> (2009)
<i>Amelanchier alnifolia</i>	Rosaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>A. florida</i>	[Rosaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Amorpha repens</i>	Leguminosae-P. (D) [Fabaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Ampelodesmos mauritanica</i>	Gramineae (M) [Poaceae (M)]	-	Dinan (1995a)
<i>Ampelopsis brevipedunculata</i>	Vitaceae (D) [Vitaceae (E)]	-	Takemoto <i>et al.</i> (1967c)
<i>Anabasis setifera</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	+	Dinan <i>et al.</i> (1998)
<i>Anagallis foemina</i>	Primulaceae (D) [Primulaceae (E)]	-	Dinan <i>et al.</i> (2001d)

<i>Anamirta cocculus</i>	Menispermaceae (D) [Menispermaceae (E)]	-	Sreejit (2014)
<i>Ananas comosus</i>	Bromeliaceae (M) [Bromeliaceae (M)]	-	Dinan <i>et al.</i> (2020a)
<i>Andrographis paniculata</i>	Acanthaceae (D) [Acanthaceae (E)]	-	Sreejit & Nelshi (2019)
<i>Andromeda polifolia</i>	Ericaceae (D) [Ericaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Andropogon scoparius</i>	Gramineae (M) [Poaceae (M)]	-	Dinan (1995a)
<i>Androsace filiformis</i>	Primulaceae (D)	+	Volodin <i>et al.</i> (2002)
<i>A. sepestralis</i>	[Primulaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Aneilema aequinoctiale</i>	Commelinaceae (M)	(+)	Crouzet <i>et al.</i> (2009)
<i>A. beninense</i>	[Commelinaceae (M)]	(+)	Crouzet <i>et al.</i> (2009)
<i>A. papuarum</i> (syn. <i>A. acuminatum</i>)		+	Crouzet <i>et al.</i> (2009)
<i>A. spiratum</i> (syn. <i>Murdannia spirata</i>)		(+)	Crouzet <i>et al.</i> (2009)
<i>A. zebrinum</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>Anemia phylliditis</i>	Schizaeaceae (F) [Anemiaceae (F)]	+	Bürcky (1977)
<i>Anemone altaica</i>	Ranunculaceae (D)	+	Savchenko <i>et al.</i> (1998a)
<i>A. apennina</i>	[Ranunculaceae (E)]	-	Savchenko <i>et al.</i> (1998a)
<i>A. baldensis</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. biarmiensis</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. blanda</i>		(+)	Savchenko <i>et al.</i> (1998a)
<i>A. canadensis</i>		(+)	Savchenko <i>et al.</i> (1998a)
<i>A. caroliniana</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. coronaria</i>		(+)	Savchenko <i>et al.</i> (1998a)
<i>A. cylindrica</i>		(+)	Savchenko <i>et al.</i> (1998a)
<i>A. decapetala</i>		(+)	Savchenko <i>et al.</i> (1998a)
<i>A. drummondii</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. hortensis</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. hupehensis</i> var. <i>japonica</i>		-	Savchenko <i>et al.</i> (1998a)
<i>A. japonica</i>		-	Savchenko <i>et al.</i> (1998a)
<i>A. leveillei</i>		-	Savchenko <i>et al.</i> (1998a)
<i>A. magellanica</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. multifida</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. narcissiflora</i>		-	Savchenko <i>et al.</i> (1998a)
<i>A. nemerosa</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. plamata</i>		-	Savchenko <i>et al.</i> (1998a)
<i>A. pavonina</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. polyanthes</i>		(+)	Savchenko <i>et al.</i> (1998a)
<i>A. ranunculoides</i>		-(+)	Savchenko <i>et al.</i> (1998a)
<i>A. rivularis</i>		-	Savchenko <i>et al.</i> (1998a)
<i>A. sylvestris</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. tetrasepala</i>		-	Savchenko <i>et al.</i> (1998a)
<i>A. tomentosa</i>		-	Savchenko <i>et al.</i> (1998a)
<i>A. tuberosa</i>		+	Savchenko <i>et al.</i> (1998a)
<i>A. virginiana</i>		(+)	Savchenko <i>et al.</i> (1998a)
<i>A. vitifolia</i>		-	Savchenko <i>et al.</i> (1998a)
<i>Anemonopsis macrophylla</i>	Ranunculaceae (D) [Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2020b)

<i>Anethum graveolens</i>	Umbelliferae (D) [Apiaceae (E)]	-	Blackford & Dinan (1997b)
<i>Angelica acutiloba</i> var. <i>acutiloba</i>	Umbelliferae (D)	-	Takemoto <i>et al.</i> (1967c)
<i>A. archangelica</i>	[Apiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>A. sylvestris</i>		-	Volodin <i>et al.</i> (2002)
<i>Angiopteris lygodiifolia</i>	Marattiaceae (F) [Marattiaceae (F)]	-	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)
<i>Annona cherimola</i>	Annonaceae (D)	-	Dinan <i>et al.</i> (2020a)
<i>A. muricata</i>	[Annonaceae (Mag)]	-	Dinan <i>et al.</i> (2020b)
<i>Antennaria dioica</i>	Compositae (D) [Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Anthemis punctata</i>	Compositae (D) [Asteraceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Anthocercis gentioides</i>	Solanaceae (D)	-	Savchenko <i>et al.</i> (2000)
<i>A. intricata</i>	[Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
<i>A. littorea</i>		-	Savchenko <i>et al.</i> (2000)
<i>A. viscosa</i>		-	Savchenko <i>et al.</i> (2000)
<i>Anthochlamys turcomania</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	+	Dinan <i>et al.</i> (1998)
<i>Anthoxanthum odoratum</i>	Gramineae (M) [Poaceae (M)]	-	Dinan (1995a)
		-	Volodin <i>et al.</i> (2002)
<i>Anthriscus cerefolium</i>	Umbelliferae (D)	-	Dinan <i>et al.</i> (2020b)
<i>A. sylvestris</i>	[Apiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Anthyllis montana rubra</i>	Leguminosae-P. (D)	-	Dinan <i>et al.</i> (2020b)
<i>A. vulneraria</i>	[Fabaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Apium graveolens</i>	Umbelliferae (D) [Apiaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Aquilegia alpina</i>	Ranunculaceae (D)	-	Dinan <i>et al.</i> (2002a)
<i>A. atrata</i>	[Ranunculaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
		-	Dinan <i>et al.</i> (2002a)
<i>A. bertolonii</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. canadensis</i>		(+)	Dinan <i>et al.</i> (2001d)
		(+)	Dinan <i>et al.</i> (2002a)
<i>A. chrysantha</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. einseleana</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. fabellata</i> var. <i>nana alba</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. flavescens</i>		-	Dinan <i>et al.</i> (2001d)
		-	Dinan <i>et al.</i> (2002a)
<i>A. formosa</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. nevadensis</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. skinneri</i>		-	Dinan <i>et al.</i> (2020b)
<i>A. viridifolia</i>		-	Dinan <i>et al.</i> (2002a)
<i>A. vulgaris</i>		-	Dinan <i>et al.</i> (2002a)
<i>Arabidopsis thaliana</i>	Cruciferae (D) [Brassicaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)

<i>Arabis (Cardaminopsis) arenosa</i>	Cruciferae (D)	-	Dinan <i>et al.</i> (2001d)
<i>A. collina rosea</i>	[Brassicaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>A. cypria</i>		-	Dinan <i>et al.</i> (2020b)
<i>A. drumondii</i>		-	Dinan <i>et al.</i> (2001d)
<i>A. pumila</i>		-	Dinan <i>et al.</i> (2020b)
<i>A. soyeri</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>Arachis hypogaea</i>	Leguminosae-P. (D)	(+)	Blackford <i>et al.</i> (1996)
	[Fabaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Araiostegia parvipinnula</i>	Davalliaceae (F)	+	Yen <i>et al.</i> (1974)
<i>A. perdurans</i>	[Davalliaceae (F)]	+	Yen <i>et al.</i> (1974)
<i>Aralia cachemirica</i>	Araliaceae (D)	-	Dinan <i>et al.</i> (2020b)
	[Araliaceae (E)]		
<i>Araucaria bidwillii</i>	Araucariaceae (G)	-	Hoffmeister <i>et al.</i> (1967)
	[Araucariaceae (G)]		
<i>Arcangelisia flava</i>	Menispermaceae (D)	+	Subeki <i>et al.</i> (2005)
	[Menispermaceae (E)]		
<i>Arctanthemum arcticum</i>	Compositae (D)	-	Volodin <i>et al.</i> (2002)
	[Asteraceae (E)]		
<i>Arctium lappa</i>	Compositae (D)	-	Takemoto <i>et al.</i> (1967c)
	[Asteraceae (E)]	-	Blackford & Dinan (1997a)
		-	Blackford & Dinan (1997b)
<i>A. minus</i>		-	Blackford & Dinan (1997a)
<i>A. tomentosum</i>		-	Volodin <i>et al.</i> (2002)
<i>Arctophila fulva</i>	Gramineae (M)	-	Volodin <i>et al.</i> (2002)
	[Poaceae (M)]		
<i>Arctostaphylos uva-ursi</i>	Ericaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Ericaceae (E)]		
<i>Arctous alpina</i>	Ericaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Ericaceae (E)]		
<i>Ardisia corymbifera</i>	Myrsinaceae (D)	-	Volodin <i>et al.</i> (2018)
<i>A. crispa</i>	[Primulaceae (E)]	-	Volodin <i>et al.</i> (2018)
<i>Ardisia japonica</i>		+	Matsuoka <i>et al.</i> (1969)
<i>Arenaria balearica</i>	Caryophyllaceae (D)	-	Zibareva <i>et al.</i> (2003)
<i>A. erinacea</i>	[Caryophyllaceae (E)]	-	Zibareva <i>et al.</i> (2003)
<i>A. montana</i>		-	Zibareva <i>et al.</i> (2003)
<i>A. obtusiloba</i>		-	Zibareva <i>et al.</i> (2003)
<i>A. stenophylla</i>		+	Bespayeveva <i>et al.</i> (2012)
<i>Argemone mexicana</i>	Papaveraceae (D)	-	Dinan <i>et al.</i> (2001d)
	[Papaveraceae (E)]		
<i>Arisaema ambiguum</i>	Araceae (M)	6.0	Wong <i>et al.</i> (1979)
	[Araceae (M)]	+	Chou & Lu (1980)
<i>A. balansae</i>		-	Volodin <i>et al.</i> (2018)
<i>Armeria scabra</i>	Plumbaginaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Plumbaginaceae (E)]		

<i>Artemisia annua</i>	Compositae (D)	0	Wong <i>et al.</i> (1979)
<i>A. campestris</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>A. capillaris</i>		0.3	Wong <i>et al.</i> (1979)
<i>A. dracuncululus</i>		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
		-	Dinan <i>et al.</i> (2020b)
<i>A. sacrorum</i>		3.7	Wong <i>et al.</i> (1979)
<i>A. tilesii</i>		-	Volodin <i>et al.</i> (2002)
<i>A. vulgaris</i>		(+)	Volodin <i>et al.</i> (1993)
<i>Arthropteris tenella</i>	Oleandraceae (F) [Tectariaceae (F)]	+	Russell & Fenemore (1971)
<i>Arum maculatum</i>	Araceae (M) [Araceae (M)]	- -	Clément & Dinan (1991) Dinan <i>et al.</i> (2001d)
<i>Asarina barclaiana</i>	Scrophulariaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>A. erubescens</i>	[Plataginaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Asclepias speciosa</i>	Asclepiadaceae (D) [Apocynaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Asparagus acutifolius</i>	Asparagaceae (M)	-	Dinan <i>et al.</i> (2020b)
<i>A. asparagoides</i>	[Asparagaceae (M)]	-	Dinan <i>et al.</i> (2001a)
<i>A. densiflorus</i> var. <i>myersii</i>		-	Dinan <i>et al.</i> (2001a)
<i>A. dumosus</i>		+	Ahmad <i>et al.</i> (1996)
		+	Khaliq-uz-Zaman <i>et al.</i> (2000)
<i>A. falcatus</i>		+	Dinan <i>et al.</i> (2001a)
<i>A. filicinus</i>		+	Wu <i>et al.</i> (2006)
		+	Wu <i>et al.</i> (2008)
		+	Wu <i>et al.</i> (2010)
		+	Zhou <i>et al.</i> (2012)
		+	Tao <i>et al.</i> (2012)
		+	Li <i>et al.</i> (2015)
<i>A. hybridus</i>		(+)	Dinan <i>et al.</i> (2001a)
<i>A. laricinus</i>		+	Dinan <i>et al.</i> (2001a)
<i>A. myriocladus</i>		-	Dinan <i>et al.</i> (2020b)
<i>A. officinalis</i>		(+)	Dinan <i>et al.</i> (2001a)
		+	Saeng-ngam <i>et al.</i> (on-line)
<i>A. plumosus nanus</i>		-	Dinan <i>et al.</i> (2001a)
<i>A. pseudoscaber</i>		-	Dinan <i>et al.</i> (2001a)
<i>A. racemosus</i>		(+)	Dinan <i>et al.</i> (2001a)
<i>A. ramosissimus</i>		+	Dinan <i>et al.</i> (2001a)
<i>A. sarmentosus</i>		(+)	Dinan <i>et al.</i> (2001a)
<i>A. scandens</i>		+	Dinan <i>et al.</i> (2001a)
<i>A. setaceus</i>		-	Dinan <i>et al.</i> (2001a)
<i>A. sprengeri</i>		-	Dinan <i>et al.</i> (2001a)
<i>A. subulatus</i>		-	Dinan <i>et al.</i> (2001a)
<i>A. verticillatus</i>		-	Dinan <i>et al.</i> (2001a)
		-	Dinan <i>et al.</i> (2020b)
<i>A. virgatus</i>		-	Dinan <i>et al.</i> (2001a)
<i>Aspidotis densa</i>	Sinopteridaceae (F) [Pteridaceae (F)]	+	Dreier (1987)
<i>Asplenium antiquum</i>	Aspleniaceae (F)	+	Hikino <i>et al.</i> (1973)
<i>A. bulbiferum</i>	[Aspleniaceae (F)]	-	Russell & Fenemore (1971)
<i>A. cheilosorum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. ensiforme</i>		+	Hikino <i>et al.</i> (1973)
<i>A. falcatum</i>		-	Russell & Fenemore (1971)

<i>A. flaccidum</i>		-	Russell & Fenemore (1971)
<i>A. griffithianum</i>		-	Hikino <i>et al.</i> (1973)
<i>A. incisum</i>		-	Hikino <i>et al.</i> (1973)
		-	Takemoto <i>et al.</i> (1967c)
<i>A. lamprophyllum</i>		-	Russell & Fenemore (1971)
<i>A. laserpitiiifolium</i>		+	Yen <i>et al.</i> (1974)
<i>A. loriceum</i>		-	Yen <i>et al.</i> (1974)
<i>A. lucidum</i>		-	Russell & Fenemore (1971)
<i>A. nidus</i>		+	Yen <i>et al.</i> (1974)
<i>A. normale</i>		-	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)
<i>A. obliquiissimum</i>		-	Hikino <i>et al.</i> (1973)
<i>A. prolongatum</i>		+/-	Hikino <i>et al.</i> (1973)
<i>A. pseudo-wilfordii</i>		+/-	Hikino <i>et al.</i> (1973)
<i>A. ritoense</i>		+	Hikino <i>et al.</i> (1973)
<i>A. ruta-muraria</i>		-	Hikino <i>et al.</i> (1973)
<i>A. sarelii</i>		-	Hikino <i>et al.</i> (1973)
<i>A. scolopendrium</i>		+	Hikino <i>et al.</i> (1973)
<i>A. trichmanes</i>		-	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
<i>A. unilaterale</i>		-	Hikino <i>et al.</i> (1973)
<i>A. wilfordii</i>		-	Hikino <i>et al.</i> (1973)
<i>A. wrightii</i>		-	Hikino <i>et al.</i> (1973)
<i>A. yakumontanum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. yoshinagae</i>		-	Hikino <i>et al.</i> (1973)
<i>Aster scaber</i>	Compositae (D)	+	Chou & Lu (1980)
<i>A. tataricus</i>	[Asteraceae (E)]	-?	Klein (2007)
<i>Astragalus sinicus</i>	Leguminosae-P. (D)	-	Blackford & Dinan (1997a)
<i>A. subpolaris</i>	[Fabaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Athyrium</i> sp.	Woodsiaceae (F)	5.0	Wong <i>et al.</i> (1979)
<i>A. anisopterum</i>	[Athyriaceae (F)]	+	Yen <i>et al.</i> (1974)
<i>A. aphanoneuron</i>		+	Hikino <i>et al.</i> (1973)
<i>A. arisanense</i>		+	Yen <i>et al.</i> (1974)
<i>A. atkinsonii</i>		+	Yen <i>et al.</i> (1974)
<i>A. crenuloserratum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. decurrenti-alatum</i>		+	Matsuoka <i>et al.</i> (1969)
		+/-	Hikino <i>et al.</i> (1973)
<i>A. distentifolium</i>		+/-	Revina & Gureeva (1985)
<i>A. doederleinii</i>		+	Hikino <i>et al.</i> (1973)
<i>A. dubium</i>		+	Hikino <i>et al.</i> (1973)
<i>A. filix-femina</i>		+/-	Revina & Gureeva (1985)
		-	Dreier (1987)
		(+)	Olennikov & Kashchenko (2018)
<i>A. fluviale</i>		+	Hikino <i>et al.</i> (1973)
<i>A. grammitoides</i>		-	Hikino <i>et al.</i> (1973)
<i>A. grammitoides</i> var. <i>simplicifolium</i>		-	Hikino <i>et al.</i> (1973)
<i>A. hachijoense</i>		+	Hikino <i>et al.</i> (1973)
<i>A. henryi</i>		+/-	Hikino <i>et al.</i> (1973)
<i>A. iseanum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. iseanum</i> var. <i>angustisectum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. japonicum</i>		-	Takemoto <i>et al.</i> (1967c)
		+	Russell & Fenemore (1971)
		-	Hikino <i>et al.</i> (1973)
<i>A. japonicum</i> var. <i>dimorphophyllum</i>		-	Hikino <i>et al.</i> (1973)
<i>A. japonicum</i> var. <i>grammitoides</i>		+	Hikino <i>et al.</i> (1973)
<i>A. maximum</i>		+/-	Hikino <i>et al.</i> (1973)

<i>A. melanolepis</i>		+	Hikino <i>et al.</i> (1973)
<i>A. mesosorum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. mettenianum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. mettenianum</i> var. <i>fauriei</i>		+	Hikino <i>et al.</i> (1973)
<i>A. multifidum</i>		-	Takemoto <i>et al.</i> (1967c)
		-	Hikino <i>et al.</i> (1973)
<i>A. multifidum</i> var. <i>acutissima</i>		+/-	Hikino <i>et al.</i> (1973)
<i>A. naganumanum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. nakanoi</i>		+	Hikino <i>et al.</i> (1973)
<i>A. nipponicola</i>		+	Hikino <i>et al.</i> (1973)
<i>A. niponicum</i>		+	Imai <i>et al.</i> (1969d)
		+	Takemoto <i>et al.</i> (1967c)
		+	Matsuoka <i>et al.</i> (1969)
		+	Hikino & Hikino (1970)
		+	Hikino <i>et al.</i> (1973)
		+	Takemoto <i>et al.</i> (1973)
<i>A. okudairai</i>		-	Hikino <i>et al.</i> (1973)
<i>A. otophorum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. petri</i>		+	Hikino <i>et al.</i> (1973)
<i>A. procerum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. pycnosorum</i>		+/-	Hikino <i>et al.</i> (1973)
<i>A. spinulosum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. squamigerum</i>		+	Takemoto <i>et al.</i> (1967c)
		+	Hikino <i>et al.</i> (1973)
<i>A. subrigescens</i>		+	Hikino <i>et al.</i> (1973)
<i>A. unifurcatum</i>		+	Hikino <i>et al.</i> (1973)
<i>A. vidalii</i>		+	Takemoto <i>et al.</i> (1967c)
		+	Matsuoka <i>et al.</i> (1969)
		+	Hikino <i>et al.</i> (1973)
<i>A. virescens</i>		-/+	Hikino <i>et al.</i> (1973)
<i>A. wardii</i>		+	Hikino <i>et al.</i> (1973)
<i>A. wardii</i> var. <i>majus</i>		+	Hikino <i>et al.</i> (1973)
<i>A. wichurae</i>		-	Hikino <i>et al.</i> (1973)
<i>A. yakushimense</i>		+	Hikino <i>et al.</i> (1973)
<i>A. yokoscense</i>		+	Matsuoka <i>et al.</i> (1969)
		+	Imai <i>et al.</i> (1969c)
		+	Imai <i>et al.</i> (1969d)
		+	Hikino & Hikino (1970)
		+	Hikino <i>et al.</i> (1973)
		+	Takemoto <i>et al.</i> (1973)
		+	Ohta <i>et al.</i> (1996)
<i>Atractylodes lancea</i>	Compositae (D) [Asteraceae (E)]	1.0	Wong <i>et al.</i> (1979)
<i>Atragene sibirica</i>	Ranunculaceae (D) [Ranunculaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Atriplex acuminata</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Báthory <i>et al.</i> (1984)
<i>A. acuminata</i>		-	Dinan <i>et al.</i> (1998)
<i>A. amnicola</i>		+	Dinan <i>et al.</i> (1998)
<i>A. babingtonii</i>		-	Dinan <i>et al.</i> (1998)
<i>A. bunburyana</i>		+	Dinan <i>et al.</i> (1998)
<i>A. calotheca</i>		-	Báthory <i>et al.</i> (1984)
		-	Dinan <i>et al.</i> (1998)
<i>A. cana</i>		+	Agabekova <i>et al.</i> (2010)
<i>A. canescens</i>		+	Dinan <i>et al.</i> (1998)
<i>A. chenopodioides</i>		-	Dinan <i>et al.</i> (1998)
<i>A. cinerea</i>		+	Dinan <i>et al.</i> (1998)
<i>A. codonocarpa</i>		+	Dinan <i>et al.</i> (1998)

<i>A. eardleyae</i>	-	Dinan <i>et al.</i> (1998)
<i>A. glabriuscula</i>	-	Dinan <i>et al.</i> (1991)
	-	Dinan <i>et al.</i> (1998)
<i>A. glauca</i>	-	Dinan <i>et al.</i> (1998)
<i>A. halimus</i>	-	Báthory <i>et al.</i> (1984)
	+	Blackford & Dinan (1997c)
	+	Dinan <i>et al.</i> (1998)
<i>A. hastata</i>	+	Báthory <i>et al.</i> (1984)
	+	Dinan <i>et al.</i> (1991)
	+/-	Dinan <i>et al.</i> (1998)
<i>A. holocarpa</i>	-	Dinan <i>et al.</i> (1998)
<i>A. hortensis</i>	-	Báthory <i>et al.</i> (1984)
	+/-	Dinan <i>et al.</i> (1998)
<i>A. hortensis rubra</i>	-	Clément & Dinan (1991)
	-	Dinan <i>et al.</i> (1991)
	-	Dinan <i>et al.</i> (2020b)
<i>A. inflata</i>	+	Ateya <i>et al.</i> (2005)
	+	Ben Nejma <i>et al.</i> (2017)
<i>A. isatidea</i>	+	Dinan <i>et al.</i> (1998)
<i>A. laciniata</i>	+	Dinan <i>et al.</i> (1998)
<i>A. lentiformis</i>	-	Dinan <i>et al.</i> (1998)
<i>A. leucoclada</i>	+	El-Sakhawy <i>et al.</i> (2012)
<i>A. lindleyi</i>	+	Mohammed <i>et al.</i> (2012)
<i>A. lindleyi</i> ssp. <i>inflata</i>	-	Dinan <i>et al.</i> (1998)
	+	El-Sakhawy <i>et al.</i> (2012)
<i>A. littoralis</i>	-	Dinan <i>et al.</i> (1991)
	-/+	Dinan <i>et al.</i> (1998)
<i>A. longipes</i>	-	Dinan <i>et al.</i> (1998)
<i>A. mulleri</i>	+	Dinan <i>et al.</i> (1998)
<i>A. nitens</i>	+/-	Báthory <i>et al.</i> (1984)
	-	Dinan <i>et al.</i> (1998)
<i>A. nummularia</i>	+	Dinan <i>et al.</i> (1998)
	+	Keckeis <i>et al.</i> (2000)
<i>A. nutallii</i>	+	Dinan <i>et al.</i> (1998)
<i>A. oblongifolia</i>	+	Dinan (1995b)
	+	Dinan <i>et al.</i> (1998)
<i>A. paludosa</i>	+	Dinan <i>et al.</i> (1998)
<i>A. patula</i>	(+)	Clément & Dinan (1991)
	+	Dinan <i>et al.</i> (1991)
	-/+	Bathory <i>et al.</i> (1984)
	+	Blackford & Dinan (1997b)
	+/-	Dinan <i>et al.</i> (1998)
<i>A. parvifolia</i>	(+)	Dinan <i>et al.</i> (1998)
<i>A. polycarpa</i>	+	Dinan <i>et al.</i> (1998)
<i>A. portulacoides</i> (syn. <i>Obione portulacoides</i>)	+	Nejma <i>et al.</i> (2015)
<i>A. prostrata</i>	-	Dinan <i>et al.</i> (1998)
<i>A. pumilio</i>	+	Dinan <i>et al.</i> (1998)
<i>A. quinii</i>	(+)	Dinan <i>et al.</i> (1998)
<i>A. rhagodioides</i>	+	Dinan <i>et al.</i> (1998)
<i>A. rosea</i>	-	Dinan <i>et al.</i> (1998)
<i>A. semibaccata</i>	(+)	Dinan <i>et al.</i> (1998)
<i>A. semilunaris</i>	(+)	Dinan <i>et al.</i> (1998)
<i>A. sibirica</i>	-	Dinan <i>et al.</i> (1998)
<i>A. sosea</i> (<i>rosea</i> ?)	-	Báthory <i>et al.</i> (1984)
<i>A. spongiosa</i>	-	Dinan <i>et al.</i> (1998)
<i>A. suberecta</i>	-	Dinan <i>et al.</i> (1998)
<i>A. tatarica</i>	-	Báthory <i>et al.</i> (1984)
<i>A. undulata</i>	+	Dinan <i>et al.</i> (1998)
<i>A. verricifera</i>	+	Agabekova <i>et al.</i> (2010)
<i>A. vesicaria</i>	+	Dinan <i>et al.</i> (1998)

<i>Atropa baetica</i>	Solanaceae (D)	-/(+)	Savchenko <i>et al.</i> (2000)
<i>A. belladonna</i>	[Solanaceae (E)]	-	Blackford & Dinan (1997c)
		-/(+)	Savchenko <i>et al.</i> (2000)
<i>Aucuba japonica</i>	Aucubaceae (D)	-	Takemoto <i>et al.</i> (1967c)
<i>A. japonica</i>	[Garryaceae (E)]	+	Matsuoka <i>et al.</i> (1969)
<i>Avena sativa</i>	Gramineae (M)	(+)	Dinan (1995a)
	[Poaceae (M)]	-	Dinan <i>et al.</i> (2020a)
<i>Arvenella flexuosa</i>	Gramineae (M)	-	Volodin <i>et al.</i> (2002)
	[Poaceae (M)]		
<i>Averrhoa carambola</i>	Oxalidaceae (D)	(+)	Volodin <i>et al.</i> (2018)
	[Oxalidaceae (E)]	-	Dinan <i>et al.</i> (2020a)
		-	Dinan <i>et al.</i> (2020b)
<i>Axyris amaranthoides</i>	Chenopodiaceae (D)	+	Dinan (1995b)
	[Amaranthaceae (E)]	+	Dinan <i>et al.</i> (1998)
		+	Sarker <i>et al.</i> (1998c)
<i>Ayapana triplinervius</i>	Compositae (D)	-	Sreejit & Nelshi (2019)
	[Asteraceae (E)]		
<i>Azadirachta indica</i>	Meliaceae (D)	-	Sreejit & Nelshi (2019)
	[Meliaceae (E)]		
<i>Azolla imbricata</i>	Azollaceae (F)	+	Hikino <i>et al.</i> (1973)
	[Salviniaceae (F)]		
B			
<i>Bambusa arundinacea</i>	Gramineae (M)	-	Dinan (1995a)
	[Poaceae (M)]		
<i>Barbarea verna</i>	Cruciferae (D)	-	Dinan <i>et al.</i> (2020b)
	[Brassicaceae (E)]		
<i>Bartsia alpina</i>	Scrophulariaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Orabanchaceae (E)]		
<i>Bassia arabica</i>	Chenopodiaceae (D)	-	Dinan <i>et al.</i> (1998)
<i>B. eriophora</i>	[Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>B. hirsuta</i>		-	Dinan <i>et al.</i> (1991)
		-	Dinan <i>et al.</i> (1998)
<i>B. hyssopifolia</i>		-	Dinan <i>et al.</i> (1998)
<i>B. laniflora</i>		-	Dinan <i>et al.</i> (1998)
<i>B. quinquecupis</i>		+	Bergamasco & Horn (1983)
<i>B. scoparia</i>		-/+	Dinan <i>et al.</i> (1998)
<i>Batrachium</i> sp.	Ranunculaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Ranunculaceae (E)]		
<i>Bellevalia romana</i>	Hyacinthaceae (M)	(+)	Dinan <i>et al.</i> (2001d)
	[Asparagaceae (M)]		
<i>Belosynapsis</i> sp.	Commelinaceae (M)	(+)	Crouzet <i>et al.</i> (2009)
	[Commelinaceae (M)]		
<i>Berchemia racemosa</i>	Rhamnaceae (D)	-	Takemoto <i>et al.</i> (1967c)

	[Rhamnaceae (E)]		
<i>Berteroa</i> sp.	Cruciferae (D) [Brassicaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Beta lomatomogona</i>	Chenopodiaceae (D)	-	Dinan <i>et al.</i> (1998)
<i>B. macrocarpa</i>	[Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>B. patellaris</i>		+	Dinan (1995b)
		+	Dinan <i>et al.</i> (1998)
<i>B. procumbens</i>		-	Dinan (1995b)
		-	Dinan <i>et al.</i> (1998)
<i>B. trigyna</i>		-	Dinan <i>et al.</i> (1998)
<i>B. vulgaris</i>		-	Clément & Dinan (1991)
		-	Dinan <i>et al.</i> (1998)
<i>B. vulgaris</i> ssp. <i>maritima</i>		-	Dinan <i>et al.</i> (1991)
		-	Blackford & Dinan (1997b)
		-	Blackford & Dinan (1997c)
		-	Dinan <i>et al.</i> (1998)
<i>B. vulgaris</i> ssp. <i>rapacea</i> convar. <i>altissima</i>		+	Báthory <i>et al.</i> (1984)
<i>B. vulgaris</i> ssp. <i>rapacea</i> convar. <i>conditiva</i>		+	Báthory <i>et al.</i> (1984)
<i>B. vulgaris</i> ssp. <i>rapacea</i> convar. <i>crassa</i>		+	Báthory <i>et al.</i> (1984)
<i>B. vulgaris vulgaris</i>		-	Dinan <i>et al.</i> (2020a)
<i>B. webbiana</i>		+	Dinan (1995b)
		+	Dinan <i>et al.</i> (1998)
<i>Betula globispica</i>	Betulaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>B. nana</i>	[Betulaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>B. tortuosa</i>		-	Volodin <i>et al.</i> (2002)
<i>Bidens bipinnata</i>	Compositae (D)	2.5	Wong <i>et al.</i> (1979)
<i>B. cernua</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>B. tripartita</i>		-	Volodin <i>et al.</i> (2002)
<i>Bienertia cycloptera</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Biota orientalis</i> (syn. <i>Platycladus orientalis</i>)	Cupressaceae (G) [Cupressaceae (G)]	1.4	Wong <i>et al.</i> (1979)
<i>Bischofia javanica</i>	Euphorbiaceae (D) [Phyllanthaceae (E)]	5.3	Wong <i>et al.</i> (1979)
<i>Bistorta major</i>	Polygonaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>B. vivipara</i>	[Polygonaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Blandfordia grandiflora</i>	Blandfordiaceae (M)	+	Sarker <i>et al.</i> (1996b)
<i>B. punicea</i>	[Blandfordiaceae (M)]	+	Sarker <i>et al.</i> (1996b)
<i>Blechnum amabile</i>	Blechnaceae (F) [Blechnaceae (F)]	+	Takemoto <i>et al.</i> (1969)
		+	Takemoto <i>et al.</i> (1973)
		+	Hikino <i>et al.</i> (1973)
<i>B. castaneum</i>		+	Hikino <i>et al.</i> (1973)
		+	Matsuoka <i>et al.</i> (1969)
<i>B. chilense</i>		-	Dinan <i>et al.</i> (2020b)
<i>B. fraseri</i>		-	Russell & Fenemore (1971)
<i>B. capense</i>		-	Russell & Fenemore (1971)
<i>B. discolor</i>		-	Russell & Fenemore (1971)
<i>B. filiforme</i>		-	Russell & Fenemore (1971)
<i>B. fluviatile</i>		+	Russell & Fenemore (1971)
<i>B. lanceolatum</i>		+	Russell & Fenemore (1971)

<i>B. minus</i>		+	Suksamrarn <i>et al.</i> (1986)
		+	Bergamasco & Horn (1983)
<i>B. nigrum</i>		+	Russell & Fenemore (1971)
<i>B. niponicum</i>		+	Hikino <i>et al.</i> (1975b)
		+	Takemoto <i>et al.</i> (1967c)
		+	Takemoto <i>et al.</i> (1968h)
		+	Takemoto <i>et al.</i> (1969)
		+	Takemoto <i>et al.</i> (1973)
		+	Matsuoka <i>et al.</i> (1969)
		+	Yen <i>et al.</i> (1974)
		+	Hikino <i>et al.</i> (1973)
<i>B. orientale</i>		-	Hikino <i>et al.</i> (1973)
		-/+	Yen <i>et al.</i> (1974)
		1.0	Wong <i>et al.</i> (1979)
<i>B. patersonii</i>		-	Russell & Fenemore (1971)
<i>B. spicant</i>		+	Dreier (1987)
		+	Vaisar & Pis (1993)
<i>B. vulcanicum</i>		+	Russell & Fenemore (1971)
		+	Russell <i>et al.</i> (1981)
<i>Blitum capitatum</i>	Chenopodiaceae (D)	-	Dinan <i>et al.</i> (1998)
<i>B. virgatum</i>	[Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Bloomeria crocea</i>	Alliaceae (M) [Asparagaceae (M)]	-	Dinan <i>et al.</i> (2020b)
<i>Boerhaavia diffusa</i>	Nyctaginaceae (D) [Nyctaginaceae (E)]	+	Suri <i>et al.</i> (1982)
		-	Sreejit (2014)
<i>Bolbitis subcordata</i>	Lomariopsidaceae (F) [Dryopteridaceae (F)]	+/-	Hikino <i>et al.</i> (1973)
<i>Bolbostemma paniculatum</i>	Cucurbitaceae (D) [Cucurbitaceae (E)]	+?	Zeng <i>et al.</i> (2018)
<i>Borago officinalis</i>	Boraginaceae (D) [Boraginaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Bosea yervamora</i>	Amaranthaceae (D) [Amaranthaceae (E)]	+	Takemoto <i>et al.</i> (1967d)
<i>Botrychium danucefolium</i>	Ophioglossaceae (F)	-	Hikino <i>et al.</i> (1973)
<i>B. japonicum</i>	[Ophioglossaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>B. ternatum</i>		-	Hikino <i>et al.</i> (1973)
<i>B. virginianum</i>		-	Hikino <i>et al.</i> (1973)
<i>Bouteloua curtipendula</i>	Gramineae (M)	-	Dinan (1995a)
<i>B. gracilis</i>	[Poaceae (M)]	-	Dinan (1995a)
<i>Boweia volubilis</i>	Hyacinthaceae (M) [Asparagaceae (M)]	-	Dinan <i>et al.</i> (2020b)
<i>Boykinia jamesii</i>	Saxifragaceae (D) [Saxifragaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Brainea insignis</i>	Blechnaceae (F) [Blechnaceae (F)]	+	Wu <i>et al.</i> (2010)
<i>Brassica kaber</i>	Cruciferae (D) [Brassicaceae (E)]	-	Blackford <i>et al.</i> (1996)
		-	Blackford & Dinan (1997b)

<i>B. napus</i>		-	Blackford <i>et al.</i> (1996)
<i>B. oleracea</i> var. <i>capitata</i>		+	Matsuoka <i>et al.</i> (1969)
		-	Blackford & Dinan (1997b)
		-	Dinan <i>et al.</i> (2020a)
<i>B. oleracea</i> cv <i>alboglabra</i>		-	Blackford <i>et al.</i> (1996)
		-	Blackford & Dinan (1997b)
<i>B. oleracea</i> cv <i>botrytis</i>		-	Blackford & Dinan (1997b)
		-	Dinan <i>et al.</i> (2020a)
<i>B. oleracea</i> cv <i>gemmaifera</i>		-	Blackford & Dinan (1997b)
<i>B. oleracea</i> cv <i>gongylodes</i>		-	Blackford <i>et al.</i> (1996)
		-	Blackford & Dinan (1997b)
<i>B. oleracea</i> cv <i>italica</i>		-	Blackford & Dinan (1997b)
		-	Dinan <i>et al.</i> (2020a)
<i>B. pekinensis</i>		-	Blackford <i>et al.</i> (1996)
<i>B. rapa rapa</i>		-	Dinan <i>et al.</i> (2020a)
<i>Briedelia stipularis</i>	Euphorbiaceae (D) [Phyllanthaceae (E)]	-	Sreejit (2014)
<i>Briza elatior</i>	Gramineae (M)	+	Savchenko <i>et al.</i> (1998c)
<i>B. erecta</i>	[Poaceae (M)]	-	Savchenko <i>et al.</i> (1998c)
<i>B. humilis</i>		-	Savchenko <i>et al.</i> (1998c)
<i>B. lamarkianum</i>		-	Savchenko <i>et al.</i> (1998c)
<i>B. lindmanii</i>		-	Savchenko <i>et al.</i> (1998c)
<i>B. maxima</i>		+	Dinan (1995a)
		+	Savchenko <i>et al.</i> (1998c)
<i>B. media</i>		+	Dinan (1995a)
		+	Savchenko <i>et al.</i> (1998c)
<i>B. minor</i>		+	Dinan (1995a)
		+	Savchenko <i>et al.</i> (1998c)
<i>B. poaemorpha</i>		-	Savchenko <i>et al.</i> (1998c)
<i>B. subaristata</i>		-	Savchenko <i>et al.</i> (1998c)
<i>B. triloba</i>		-	Savchenko <i>et al.</i> (1998c)
<i>B. uniolae</i>		-	Savchenko <i>et al.</i> (1998c)
<i>Bromopsis inermis</i>	Gramineae (M) [Poaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Bromus interruptis</i>	Gramineae (M)	-	Dinan <i>et al.</i> (2001d)
<i>B. macrostachys</i>	[Poaceae (M)]	-	Dinan (1995a)
<i>Broussonetia kazinoki</i>	Moraceae (D)	-	Takemoto <i>et al.</i> (1967c)
<i>B. papyrifera</i>	[Moraceae (E)]	1.5	Wong <i>et al.</i> (1979)
<i>B. papyrifera</i>		+	Matsuoka <i>et al.</i> (1969)
<i>Browallia speciosa</i>	Solanaceae (D)	+	Savchenko <i>et al.</i> (2000)
<i>B. viscosa</i>	[Solanaceae (E)]	+	Savchenko <i>et al.</i> (2000)
<i>Brugmansia suaveolens</i>	Solanaceae (D) [Solanaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Brunfelsia americana</i>	Solanaceae (D)	-	Savchenko <i>et al.</i> (2000)
<i>B. australis</i>	[Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
<i>B. calycina</i> var. <i>floribunda</i>		-	Savchenko <i>et al.</i> (2000)
<i>B. grandiflora</i>		-	Savchenko <i>et al.</i> (2000)
<i>B. jamaicensis</i>		-	Savchenko <i>et al.</i> (2000)
<i>B. lactea</i>		-	Savchenko <i>et al.</i> (2000)
<i>B. latifolia</i>		-	Savchenko <i>et al.</i> (2000)
<i>B. nitida</i>		-	Savchenko <i>et al.</i> (2000)
<i>B. plicata</i>		-	Savchenko <i>et al.</i> (2000)

<i>B. undulata</i>		-	Savchenko <i>et al. et al.</i> (2000)
<i>B. uniflora</i>		-	Savchenko <i>et al.</i> (2000)
<i>Buddleja davidii</i>	Buddlejaceae (D)	(+)	Blackford & Dinan (1997c)
<i>B. globosa</i>	[Scrophulariaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Buglossoides arvensis</i>	Boraginaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
	[Boraginaceae (E)]		
<i>Bunias orientalis</i>	Cruciferae (D)	-	Volodin <i>et al.</i> (2002)
	[Brassicaceae (E)]		
<i>Butomus umbellata</i>	Butomaceae (M)	+	Matsuoka <i>et al.</i> (1969)
	[Butomaceae (M)]	+	Volodin <i>et al.</i> (2002)
C			
<i>Cacalia hastata</i>	Compositae (D)	-	Volodin <i>et al.</i> (2002)
	[Asteraceae (E)]		
<i>Caladrina grandiflora</i>	Portulaccaceae (D)	-	Dinan <i>et al.</i> (2001d)
	[Montiaceae (E)]		
<i>Calamagrostis arundinacea</i>	Gramineae (M)	-	Volodin <i>et al.</i> (2002)
<i>C. epigeios</i>	[Poaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>C. lapponica</i>		-	Volodin <i>et al.</i> (2002)
<i>C. neglecta</i>		-	Volodin <i>et al.</i> (2002)
<i>C. purpurea</i>		-	Volodin <i>et al.</i> (2002)
<i>Calamintha confinis</i>	Labiatae (D)	3.0	Wong <i>et al.</i> (1979)
	[Lamiaceae (E)]		
<i>Calla palustris</i>	Araceae (M)	-	Volodin <i>et al.</i> (2002)
	[Araceae (M)]		
<i>Callicarpa japonica</i>	Verbenaceae (D)	-	Takemoto <i>et al.</i> (1967c)
<i>Callicarpa</i> sp.	[Lamiaceae (E)]	-	Volodin <i>et al.</i> (2018)
<i>Callisia elegans</i> (syn. <i>C. gentelei</i> var <i>elegans</i>)	Commelinaceae (M)	(+)	Crouzet <i>et al.</i> (2009)
<i>C. fragrans</i>	[Commelinaceae (M)]	(+)	Crouzet <i>et al.</i> (2009)
		+	Hang <i>et al.</i> (2014)
<i>C. guerriensis</i> (syn. <i>C. socunuscensis</i>)		(+)	Crouzet <i>et al.</i> (2009)
<i>C. insignis</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>C. macedorigalli</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>C. multiflora</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>C. navicularis</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>C. repens</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>C. warscewicziana</i>		+	Crouzet <i>et al.</i> (2009)
<i>Callistemon piyoides</i>	Myrtaceae (D)	-	Dinan <i>et al.</i> (2001d)
	[Myrtaceae (E)]		
<i>Callitriche hermaphroditica</i>	Callitrichaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>C. palustris</i>	[Plantaginaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Calluna vulgaris</i>	Ericaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Ericaceae (E)]		
<i>Calochortus nitidus</i>	Liliaceae (M)	-	Dinan <i>et al.</i> (2001c)
	[Liliaceae (M)]		

<i>Caltha palustris</i>	Ranunculaceae (D) [Ranunculaceae (E)]	- -	Volodin <i>et al.</i> (2002) Dinan <i>et al.</i> (2020b)
<i>Calystegia sepium</i>	Convolvulaceae (D) [Convolvulaceae (E)]	-	Blackford & Dinan (1997b)
<i>Camassia quamash</i>	Hyacinthaceae (M) [Asparagaceae (M)]	-	Dinan <i>et al.</i> (2020b)
<i>Campanula carpatica</i>	Campanulaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>C. makaschvilii</i>	[Campanulaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>C. rotundifolia</i>		-	Volodin <i>et al.</i> (2002)
<i>Campanumoea (Codonopsis) javanica</i>	Campanulaceae (D) [Campanulaceae (E)]	+	Yang <i>et al.</i> (2015)
<i>Camphorosma monspeliaceum</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Agabekova <i>et al.</i> (2010)
<i>Camptosorus sibiricus</i> (syn. <i>Asplenium ruprechtii</i>)	Aspleniaceae (F) [Aspleniaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>Camptotheca acuminata</i>	Cornaceae (D) [Nyssaceae (E)]	2.9	Wong <i>et al.</i> (1979)
<i>Capsicum sp.</i>	Solanaceae (D)	-	Blackford <i>et al.</i> (1996)
<i>Capsicum anuum</i>	[Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
		-	Dinan <i>et al.</i> (2020a)
<i>C. frutescens</i>		-	Savchenko <i>et al.</i> (2000)
<i>C. grossum</i>		(+)	Savchenko <i>et al.</i> (2000)
<i>Cardamine pratensis</i>	Cruciferae (D)	-	Volodin <i>et al.</i> (2002)
<i>Cardamine sp.</i>	[Brassicaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Cardiocrinum cordata</i>	Liliaceae (M)	-	Dinan <i>et al.</i> (2001c)
<i>C. giganteum</i>	[Liliaceae (M)]	-	Dinan <i>et al.</i> (2001c)
<i>C. yunnanensis</i>		-	Dinan <i>et al.</i> (2001c)
<i>Cardiospermum helicacabrum</i>	Sapindaceae (D) [Sapindaceae (E)]	-	Sreejit & Nelshi (2019)
<i>Carduus crispus</i>	Compositae (D)	-	Volodin <i>et al.</i> (2002)
<i>C. nutans</i>	[Asteraceae (E)]	-	Blackford & Dinan (1997a)
		-	Volodin <i>et al.</i> (2002)
<i>Carex aquatilis</i>	Cyperaceae (M)	-	Volodin <i>et al.</i> (2002)
<i>C. baccans</i>	[Cyperaceae (M)]	(+)	Dinan <i>et al.</i> (2001d)
		-	Meng <i>et al.</i> (2001b)
<i>C. berggrenii</i>		(+)	Dinan <i>et al.</i> (2001d)
		-	Meng <i>et al.</i> (2001b)
<i>C. bohemica</i>		-	Meng <i>et al.</i> (2001b)
<i>C. buchananii</i>		-	Meng <i>et al.</i> (2001b)
<i>C. comans</i>		-	Meng <i>et al.</i> (2001b)
<i>C. dallii</i>		-	Meng <i>et al.</i> (2001b)
<i>C. demissa</i>		-	Meng <i>et al.</i> (2001b)
<i>C. duriuscula</i>		-	Meng <i>et al.</i> (2001b)
<i>C. flagellifera</i>		-	Dinan <i>et al.</i> (2001d)
		-	Meng <i>et al.</i> (2001b)
<i>C. globularis</i>		-	Volodin <i>et al.</i> (2002)
<i>C. grayi</i>		-	Meng <i>et al.</i> (2001b)

<i>C. hartmanii</i>		-	Volodin <i>et al.</i> (2002)
<i>C. hirta</i>		-	Meng <i>et al.</i> (2001b)
<i>C. hordeistichos</i>		-	Meng <i>et al.</i> (2001b)
<i>C. lasiocarpa</i>		-	Meng <i>et al.</i> (2001b)
<i>C. licherica</i>		-	Meng <i>et al.</i> (2001b)
<i>C. limosa</i>		-	Meng <i>et al.</i> (2001b)
<i>C. muricata</i>		-	Meng <i>et al.</i> (2001b)
<i>C. muskingumensis</i>		-	Meng <i>et al.</i> (2001b)
<i>C. nigra</i>		-	Meng <i>et al.</i> (2001b)
		-	Volodin <i>et al.</i> (2002)
<i>C. otrubae</i>		-	Meng <i>et al.</i> (2001b)
<i>C. pallescens</i>		-	Meng <i>et al.</i> (2001b)
		-	Volodin <i>et al.</i> (2002)
<i>C. pendula</i>		-	Meng <i>et al.</i> (2001b)
<i>C. plantaginea</i>		-	Meng <i>et al.</i> (2001b)
<i>C. pseudocyperus</i>		-	Meng <i>et al.</i> (2001b)
<i>C. rariflora</i>		-	Volodin <i>et al.</i> (2002)
<i>C. rostrata</i>		-	Meng <i>et al.</i> (2001b)
<i>C. secta</i>		-	Dinan <i>et al.</i> (2001d)
		-	Meng <i>et al.</i> (2001b)
<i>C. sylvatica</i>		-	Meng <i>et al.</i> (2001b)
<i>C. trifida</i>		-	Meng <i>et al.</i> (2001b)
		-	Dinan <i>et al.</i> (2020b)
<i>C. vesicaria</i>		-	Volodin <i>et al.</i> (2002)
<i>Carlina vulgaris</i>	Compositae (D)	-	Blackford & Dinan (1997a)
<i>Carlina</i> sp.	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Carica papaya</i>	Caricaceae (D) [Caricaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Carpesium abrotanoides</i>	Compositae (D) [Asteraceae (E)]	0	Wong <i>et al.</i> (1979)
<i>Carthamus tinctoria</i>	Compositae (D)	-	Blackford & Dinan (1997a)
<i>C. tinctorius</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Carum carvi</i>	Umbelliferae (D) [Apiaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Carya illinoensis</i>	Juglandaceae (D) [Juglandaceae (E)]	-	Dinan <i>et al.</i> (2020a)
		-	Dinan <i>et al.</i> (2020b)
<i>Caryopteris nepetifolia</i> (syn. <i>Schnabella nepetifolia</i>)	Verbenaceae (D) [Lamiaceae (E)]	4.0	Wong <i>et al.</i> (1979)
<i>Cassia fistula</i>	Leguminosae-C. (D)	-	Sreejit (2014)
<i>C. tora</i>	[Fabaceae (E)]	5.0	Wong <i>et al.</i> (1979)
		+	Chou & Lu (1980)
		+	Shivakumar <i>et al.</i> (1995)
(syn. <i>Senna tora</i>)		-	Sreejit (2014)
<i>Castanea sativa</i>	Fagaceae (D) [Fagaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Castilleja miniata</i>	Scrophulariaceae(D) [Orobanchaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Casuarina equisetifolia</i>	Casuarinaceae (D) [Casuarinaceae (E)]	0	Wong <i>et al.</i> (1979)

<i>Cayratia japonica</i>	Vitaceae (D) [Vitaceae (E)]	0	Wong <i>et al.</i> (1979)
<i>Cedrus deodara</i>	Pinaceae (G) [Pinaceae (G)]	-	Takemoto <i>et al.</i> (1967c)
<i>Celastrus orbiculatus</i>	Celastraceae (D) [Celastraceae (E)]	+	Yang <i>et al.</i> (2020)
<i>Centaurea americana</i>	Compositae (D) [Asteraceae (E)]	+	Sarker <i>et al.</i> (1997a)
<i>C. arenaria</i>		+	Schoeb <i>et al.</i> (2006)
<i>C. collina</i>		-	Sarker <i>et al.</i> (1997a)
<i>C. cyanus</i>		-	Sarker <i>et al.</i> (1997a)
<i>C. dealbata</i>		-	Dinan <i>et al.</i> (2020b)
		(+)	Sarker <i>et al.</i> (1997a)
		(+)	Blackford & Dinan (1997a)
<i>C. fischeri</i>		-	Volodin <i>et al.</i> (2002)
<i>C. imperialis</i>		+	Blackford & Dinan (1997a)
<i>C. jacea</i>		-	Sarker <i>et al.</i> (1997a)
		-	Volodin <i>et al.</i> (2002)
<i>C. macrocephala</i>		-	Sarker <i>et al.</i> (1997a)
		(+)	Blackford & Dinan (1997a)
		-	Dinan <i>et al.</i> (2020b)
<i>C. micranthos</i>		-	Sarker <i>et al.</i> (1997a)
<i>C. montana</i>		-	Sarker <i>et al.</i> (1997a)
<i>C. moschata</i>		+	Sarker <i>et al.</i> (1997a)
<i>C. nervosa</i>		-	Sarker <i>et al.</i> (1997a)
<i>C. nigra</i>		-	Sarker <i>et al.</i> (1997a)
		(+)	Blackford & Dinan (1997a)
<i>C. orientalis</i>		-	Sarker <i>et al.</i> (1997a)
<i>C. paniculata</i> ssp. <i>henryi</i>		-	Sarker <i>et al.</i> (1997a)
<i>C. pannonica</i>		(+)	Sarker <i>et al.</i> (1997a)
<i>C. phrygia</i>		-	Sarker <i>et al.</i> (1997a)
		+	Volodin <i>et al.</i> (1993)
		-	Volodin <i>et al.</i> (2002)
<i>C. pulcherrima</i>		-	Sarker <i>et al.</i> (1997a)
<i>C. rothrockii</i>		+	Sarker <i>et al.</i> (1997a)
<i>C. rupestris</i>		(+)	Sarker <i>et al.</i> (1997a)
<i>C. ruthenica</i>		-	Sarker <i>et al.</i> (1997a)
		-	Dinan <i>et al.</i> (2020b)
<i>C. sadlerana</i>		-	Sarker <i>et al.</i> (1997a)
<i>C. scabiosa</i>		-	Sarker <i>et al.</i> (1997a)
		-	Volodin <i>et al.</i> (2002)
<i>C. uniflora</i>		-	Sarker <i>et al.</i> (1997a)
<i>C. uralensis</i>		-	Dinan <i>et al.</i> (2020b)
<i>Centaureum scilloides</i>	Gentianaceae (D) [Gentianaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Centella asiatica</i>	Umbelliferae (D) [Apiaceae (E)]	-	Sreejit & Nelshi (2019)
<i>Cephalotaxus fastigiata</i>	Cephalotaxaceae (G)	-	Imai <i>et al.</i> (1967)
<i>C. fortunei</i>	[Taxaceae (G)]	+	Feng <i>et al.</i> (2018)
		-	Dinan <i>et al.</i> (2020b)
<i>C. harringtonia</i>		-	Imai <i>et al.</i> (1967)
		+	Imai <i>et al.</i> (1969d)
		-	Takemoto <i>et al.</i> (1967c)

<i>Cerastium alpinum</i>	Caryophyllaceae (D)	-	Zibareva <i>et al.</i> (2003)
<i>C. arvense</i>	[Caryophyllaceae (E)]	-	Revina <i>et al.</i> (1988)
<i>C. biebersteinii</i>		-	Zibareva <i>et al.</i> (2003)
<i>C. boissieri</i>		-	Dinan <i>et al.</i> (2001d)
		-	Zibareva <i>et al.</i> (2003)
<i>C. bungii</i>		-	Agabekova <i>et al.</i> (2010)
<i>C. davuricum</i>		-	Revina <i>et al.</i> (1988)
<i>C. holosteoides</i>		-	Revina <i>et al.</i> (1988)
		-	Volodin <i>et al.</i> (2002)
<i>C. pauciflorum</i>		-	Revina <i>et al.</i> (1988)
<i>C. tomentosum</i>		-	Zibareva <i>et al.</i> (2003)
<i>Ceratocarpus arenaries</i>	Chenopodiaceae (D)	+	Agabekova <i>et al.</i> (2010)
	[Amaranthaceae (E)]		
<i>Ceratophyllum demersum</i>	Ceratophyllaceae (D)	+	Volodin <i>et al.</i> (2002)
	[Ceratophyllaceae (C)]		
<i>Cercidiphyllum japonicum</i>	Cercidiphyllaceae (D)	+	Matsuoka <i>et al.</i> (1969)
	[Cercidiphyllaceae (E)]		
<i>Cestrum nocturnum</i>	Solanaceae (D)	-	Savchenko <i>et al.</i> (2000)
<i>C. parqui</i>	[Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
<i>C. purpureum</i>		(+)	Savchenko <i>et al.</i> (2000)
<i>Chaenorhinum minus</i>	Scrophulariaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
	[Plantaginaceae (E)]		
<i>Chaerophyllum bulbosum</i>	Umbelliferae(D)	-	Dinan <i>et al.</i> (2020b)
<i>C. prescottii</i>	[Apiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Chamaenerion (Epilobium) angustifolium</i>	Onagraceae (D)	-	Volodin <i>et al.</i> (2002)
	[Onagraceae (E)]		
<i>Chamaepericlymenum (Cornus) suecicum</i>	Cornaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Cornaceae (E)]		
<i>Cheilanthes (Aleuritopteris) argentea</i>	Adiantaceae (F)	-	Yen <i>et al.</i> (1974)
<i>C. chusana</i>	[Pteridaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>C. farinosa</i>		+	Josephraj Kumar <i>et al.</i> (2000)
<i>C. hirsuta</i>		+	Yen <i>et al.</i> (1974)
		+	Iyer <i>et al.</i> (1973)
<i>C. seiberi</i>		+	Bergamasco & Horn (1983)
<i>C. tenuifolia</i>		+	Faux <i>et al.</i> (1970)
<i>Cheiropleuria bicuspis</i>	Cheiropleuriaceae (F)	-	Hikino <i>et al.</i> (1973)
	[Dipteridaceae (F)]		
<i>Chenopodium album</i>	Chenopodiaceae (D)	5.0	Wong <i>et al.</i> (1979)
	[Amaranthaceae (E)]	+	Báthori <i>et al.</i> (1986b)
		+	Tóth <i>et al.</i> (1981)
		+	Báthori <i>et al.</i> (1987)
		+	Dinan <i>et al.</i> (1990)
		+	Dinan <i>et al.</i> (1991)
		+	Dinan (1992a)
		+	Dinan (1992b)
		+	Dinan (1992c)
		+	Corio-Costet <i>et al.</i> (1993b)
		+	Bergamasco & Horn (1983)

	+	Bathory <i>et al.</i> (1984)
	+	Blackford & Dinan (1997a)
	+	Blackford & Dinan (1997b)
	+	Zibareva (1997)
	+	Corio-Costet <i>et al.</i> (1998)
	+	Dinan <i>et al.</i> (1998)
	+	DellaGreca <i>et al.</i> (2005)
<i>C. album</i> var. <i>album</i>	+	Báthory <i>et al.</i> (1984)
<i>C. album</i> var. <i>corymbosopaniculatum</i>	+	Báthory <i>et al.</i> (1984)
<i>C. album</i> ssp. <i>borbassii</i>	+	Báthory <i>et al.</i> (1984)
<i>C. album</i> ssp. <i>pseudoopulifolium</i>	+	Báthory <i>et al.</i> (1984)
<i>C. album</i> ssp. <i>microphyllum</i>	+	Báthory <i>et al.</i> (1984)
<i>C. amaranticolor</i>	+	Dinan (1995b)
	+	Dinan <i>et al.</i> (1998)
<i>C. ambrosioides</i>	2.3	Wong <i>et al.</i> (1979)
	+/-	Báthory <i>et al.</i> (1984)
	+	Zibareva (1997)
	-	Dinan <i>et al.</i> (1998)
	-	Sreejit (2014)
<i>C. ambrosioides</i> var. <i>anthelminthicum</i>	-	Báthory <i>et al.</i> (1984)
<i>C. antarcticum</i>	-	Dinan <i>et al.</i> (1998)
<i>C. anthelminticum</i>	-	Dinan (1992a)
	-	Dinan <i>et al.</i> (1991)
	-	Clément & Dinan (1991)
	-	Dinan <i>et al.</i> (1998)
<i>C. aristatum</i>	+	Báthory <i>et al.</i> (1984)
	-	Dinan <i>et al.</i> (1998)
<i>C. atro-virens</i>	-	Dinan <i>et al.</i> (1998)
<i>C. auricoforme</i>	+	Dinan <i>et al.</i> (1998)
<i>C. auricomum</i>	+	Dinan <i>et al.</i> (1998)
<i>C. berlandieri</i>	+	Dinan <i>et al.</i> (1998)
<i>C. bonus-henicus</i>	+	Báthory <i>et al.</i> (1986b)
	+	Báthory <i>et al.</i> (1984)
	+	Dinan <i>et al.</i> (1991)
	+	Báthory <i>et al.</i> (1987)
	+	Báthory <i>et al.</i> (1982)
	+	Clément & Dinan (1991)
	+	Blackford & Dinan (1997a)
	+	Blackford & Dinan (1997c)
	+	Zibareva (1997)
	+	Dinan <i>et al.</i> (1998)
<i>C. botrys</i>	-	Báthory <i>et al.</i> (1984)
	-	Zibareva (1997)
	-	Dinan <i>et al.</i> (1998)
<i>C. burkartii</i>	-	Dinan <i>et al.</i> (1998)
<i>C. bushianum</i>	+	Dinan <i>et al.</i> (1998)
<i>C. capitatum</i>	-	Dinan (1992a)
	-	Dinan <i>et al.</i> (1991)
	-	Clément & Dinan (1991)
	-	Zibareva (1997)
<i>C. carinatum</i>	-	Dinan <i>et al.</i> (1998)
<i>C. chenopodioides</i>	-	Báthory <i>et al.</i> (1984)
<i>C. congolanum</i>	-	Dinan <i>et al.</i> (1998)
<i>C. cristatum</i>	-	Dinan <i>et al.</i> (1998)
<i>C. desertorum</i>	+	Dinan <i>et al.</i> (1998)
<i>C. fasciculosum</i>	-	Dinan <i>et al.</i> (1998)
<i>C. ficifolium</i>	+	Dinan (1995b)
	+	Báthory <i>et al.</i> (1984)
	+	Dinan <i>et al.</i> (1998)
<i>C. foliosum</i>	-	Dinan (1992a)

	-	Dinan <i>et al.</i> (1991)
	-	Báthory <i>et al.</i> (1984)
	-	Zibareva (1997)
<i>C. giganteum</i>	+	Dinan (1992a)
	+	Báthory <i>et al.</i> (1987)
	+	Clément & Dinan (1991)
	+	Dinan <i>et al.</i> (1991)
	+	Báthory <i>et al.</i> (1984)
	+	Dinan <i>et al.</i> (1998)
	+	Bespayeva <i>et al.</i> (2012)
	+	Dinan <i>et al.</i> (2020b)
<i>C. glaucum</i>	-	Báthory <i>et al.</i> (1984)
	-	Dinan <i>et al.</i> (1991)
	-	Zibareva (1997)
	-	Dinan <i>et al.</i> (1998)
<i>C. graveolens</i>	-	Dinan <i>et al.</i> (1998)
<i>C. hircinum</i>	+	Dinan <i>et al.</i> (1998)
<i>C. hybridum</i>	-	Báthory <i>et al.</i> (1984)
	-/+	Dinan <i>et al.</i> (1998)
	+	Bespayeva <i>et al.</i> (2012)
<i>C. incisum</i>	-	Dinan <i>et al.</i> (1998)
<i>C. macrospermum</i>	-	Dinan <i>et al.</i> (1998)
<i>C. multifidum</i>	-	Dinan <i>et al.</i> (1998)
<i>C. murale</i>	+	Báthory <i>et al.</i> (1984)
	+	Dinan <i>et al.</i> (1998)
<i>C. oahuense</i>	+	Dinan (1995b)
	+	Dinan <i>et al.</i> (1998)
<i>C. opulifolium</i>	+	Báthory <i>et al.</i> (1984)
	-	Zibareva (1997)
	-	Dinan <i>et al.</i> (1998)
<i>C. pallidicaule</i>	+	Rastrelli <i>et al.</i> (1996)
<i>C. petiolare</i>	+	Dinan <i>et al.</i> (1998)
<i>C. polygonoides</i>	+	Dinan <i>et al.</i> (1998)
<i>C. polyspermum</i>	+	Dinan (1992a)
	+	Clément & Dinan (1991)
	+	Dinan <i>et al.</i> (1991)
	+	Báthory <i>et al.</i> (1984)
	+	Zibareva (1997)
	+	Dinan <i>et al.</i> (1998)
<i>C. procerum</i>	-	Dinan <i>et al.</i> (1998)
<i>C. pumilio</i>	-	Dinan <i>et al.</i> (1998)
<i>C. quinoa</i>	+	Báthory <i>et al.</i> (1984)
	+	Dinan (1992a)
	+	Báthory <i>et al.</i> (1987)
	+	Clément & Dinan (1991)
	+	Dinan <i>et al.</i> (1991)
	+	Dinan <i>et al.</i> (1998)
	+	Zhu <i>et al.</i> (2001)
	+	Dini <i>et al.</i> (2005)
	+	Nsimba <i>et al.</i> (2008)
	+	Kumpun <i>et al.</i> (2011)
	+	Bespayeva <i>et al.</i> (2012)
	+	Graf <i>et al.</i> (2014)
	+	Wang <i>et al.</i> (2019)
	+	Zeng <i>et al.</i> (2019)
	+	Dinan <i>et al.</i> (2020a)
	+	Dinan <i>et al.</i> (2020b)
	+	Chen & Feng (2021)
<i>C. rubrum</i>	-	Báthory <i>et al.</i> (1984)
	-	Dinan <i>et al.</i> (1991)

		-/+	Dinan <i>et al.</i> (1998)
<i>C. sancti-ambrosii</i>		+	Dinan <i>et al.</i> (1998)
<i>C. saxatile</i>		-	Dinan <i>et al.</i> (1998)
<i>C. schraderanum</i>		-	Dinan <i>et al.</i> (1998)
<i>C. serotinum</i>		0	Wong <i>et al.</i> (1979)
<i>C. strictum</i>		+	Báthory <i>et al.</i> (1984)
		+	Dinan <i>et al.</i> (1998)
<i>C. suecicum</i>		+	Dinan <i>et al.</i> (1998)
<i>C. trigonon</i>		+	Bergamasco & Horn (1983)
<i>C. urbicum</i>		+	Báthory <i>et al.</i> (1984)
		(+)	Dinan <i>et al.</i> (1991)
		-	Dinan <i>et al.</i> (1998)
<i>C. viride</i>		-	Báthory <i>et al.</i> (1984)
		+	Dinan <i>et al.</i> (1998)
<i>C. virgatum</i>		-	Báthory <i>et al.</i> (1984)
<i>C. vulvaria</i>		+	Báthory <i>et al.</i> (1984)
		+	Blackford & Dinan (1997a)
		+	Blackford & Dinan (1997b)
		+	Dinan <i>et al.</i> (1998)
<i>Chionochloa conspicua</i>	Gramineae (M)	-	Dinan (1995a)
<i>C. rubra</i>	[Poaceae (M)]	-	Dinan <i>et al.</i> (2020b)
<i>Chloranthus multistachys</i>	Chloranthaceae (D) [Chloranthaceae (Unplaced)]	+	Liu <i>et al.</i> (2013)
<i>Choerospondias axillaris</i> var. <i>japonica</i>	Anacardiaceae (D) [Anacardiaceae (E)]	+	Matsuoka <i>et al.</i> (1969)
<i>Chrysanthemum coronarium</i>	Compositae (D)	-	Dinan <i>et al.</i> (2001d)
<i>C. indicum</i>	[Asteraceae (E)]	+	Imai <i>et al.</i> (1969d)
		5.3	Wong <i>et al.</i> (1979)
<i>C. indicum</i> var. <i>hibernum</i>		+	Matsuoka <i>et al.</i> (1969)
<i>C. leucanthemum</i>		-	Blackford <i>et al.</i> (1996)
		-	Blackford & Dinan (1997a)
		-	Blackford & Dinan (1997b)
<i>C. makinoi</i>		+	Matsuoka <i>et al.</i> (1969)
		+	Imai <i>et al.</i> (1969d)
<i>C. morifolium</i>		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
<i>Chrysosplenium alternifolium</i>	Saxifragaceae (D) [Saxifragaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Cibotium barometz</i>	Dicksoniaceae (F) [Cibotiaceae (F)]	-	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)
<i>Cicer arietinum</i>	Leguminosae-P. (D) [Fabaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Cicuta virosa</i>	Umbelliferae (D) [Apiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Cimifuga americana</i>	Ranunculaceae (D)	-	Dinan <i>et al.</i> (2002a)
<i>C. dahurica</i>	[Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2002a)
<i>C. europaea</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. japonica</i>		(+)	Dinan <i>et al.</i> (2002a)
<i>C. racemosa</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. simplex</i>		-	Dinan <i>et al.</i> (2002a)

<i>Circaea alpina</i>	Onagraceae (D) [Onagraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Cirsium acaulon</i>	Compositae (D)	-	Blackford & Dinan (1997a)
<i>C. arvense</i>	[Asteraceae (E)]	(+)	Volodin <i>et al.</i> (1993)
		-	Blackford & Dinan (1997a)
<i>C. heterophyllum</i>		-	Volodin <i>et al.</i> (2002)
<i>C. oleraceum</i>		-	Volodin <i>et al.</i> (1993)
		-	Blackford & Dinan (1997a)
		-	Volodin <i>et al.</i> (2002)
<i>C. palustre</i>		-	Blackford & Dinan (1997a)
		-	Volodin <i>et al.</i> (2002)
<i>C. vulgare</i>		-	Blackford & Dinan (1997a)
		-	Volodin <i>et al.</i> (2002)
<i>Cissampelos pareira</i> var. <i>hirsuta</i>	Menispermaceae (D) [Menispermaceae (E)]	-	Sreejit (2014)
<i>Cistus albidus</i>	Cistaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>C. hirsutus</i>	[Cistaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>C. ladanifer</i>		-	Dinan <i>et al.</i> (2001d)
<i>C. populifolius</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>C. psilosepalus</i>		-	Dinan <i>et al.</i> (2001d)
<i>Citrus x clementina</i>	Rutaceae (D)	-	Dinan <i>et al.</i> (2020a)
<i>C. hystrix</i>	[Rutaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>C. japonica</i>		-	Dinan <i>et al.</i> (2020a)
<i>C. limon</i>		(+)	Dinan <i>et al.</i> (2020a)
<i>C. maxima</i> (<i>C. grandis</i>)		-	Dinan <i>et al.</i> (2020a)
<i>C. medica</i>		+	Yin <i>et al.</i> (2015)
<i>C. medica</i> var. <i>sarcodactylis</i>		-	Dinan <i>et al.</i> (2020a)
<i>Clematis armandii</i>	Ranunculaceae (D)	-	Dinan <i>et al.</i> (2002a)
<i>C. buchananiana</i>	[Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2002a)
<i>C. campaniflora</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. chinensis</i>		4.4	Wong <i>et al.</i> (1979)
<i>C. chrysochoma</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. connata</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. forsteri</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. heracleifolia</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. hirsutissima</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. hookeriana</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. integrifolia</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. ligusticifolia</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. maximowicziana</i> var. <i>robusta</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. microphylla</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. montana</i> var. <i>rubens</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. orientalis</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. peterae</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. recta</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. stanleyi</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. stans</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. tangutica</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. vitalba</i>		-	Dinan <i>et al.</i> (2002a)
<i>C. viticella</i>		-	Dinan <i>et al.</i> (2002a)
<i>Cleome burmannii</i>	Capparidaceae (D)	-	Sreejit (2014)
<i>C. viscosa</i>	[Cleomaceae (E)]	-	Sreejit (2014)
<i>Clerodendrum bungei</i>	Verbenaceae (D)	5.0	Wong <i>et al.</i> (1979)

<i>C. canescens</i>	[Lamiaceae (E)]	-	Volodin <i>et al.</i> (2018)
<i>C. trichotomum</i>		-	Takemoto <i>et al.</i> (1967c)
		4.0	Wong <i>et al.</i> (1979)
<i>Cnicus benedictus</i>	Compositae (D) [Asteraceae (E)]	-	Blackford & Dinan (1997a)
<i>Cocculus hirsutus</i>	Menispermaceae (D) [Menispermaceae (E)]	-	Sreejit (2014)
<i>Cochliostema odorantissimum</i>	Commelinaceae (M) [Commelinaceae (M)]	+	Crouzet <i>et al.</i> (2009)
<i>Coccyganthe flos-cuculi</i>	Caryophyllaceae (M) [Caryophyllaceae (E)]	+	Volodin <i>et al.</i> (2002)
<i>Cocos nucifera</i>	Arecaceae (M) [Arecaceae (M)]	-	Sreejit (2014)
		-	Dinan <i>et al.</i> (2020a)
<i>Coeloglossum viride</i>	Orchidaceae (M) [Orchidaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Coix lachryma-jobi</i>	Gramineae (M) [Poaceae (M)]	-	Dinan (1995a)
		-	Volodin <i>et al.</i> (2018)
<i>C. lachryma-jobi</i> var. Ma-Yuen		+	Matsuoka <i>et al.</i> (1969)
<i>Colchicum neapolitanum</i>	Colchicaceae (M) [Colchicaceae (M)]	-	Dinan <i>et al.</i> (2020b)
<i>Coleotrype goudotii</i>	Commelinaceae (M)	+	Crouzet <i>et al.</i> (2009)
<i>C. natalensis</i>	[Commelinaceae (M)]	+	Crouzet <i>et al.</i> (2009)
<i>Coleus (Plectranthus) frederici</i>	Labiatae (D) [Lamiaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Colysis (Leptochilus) elliptica</i>	Polypodiaceae (F) [Polypodiaceae (F)]	+	Yen <i>et al.</i> (1974)
		+	Hikino <i>et al.</i> (1973)
<i>C. hemionitidea</i>		+	Hikino <i>et al.</i> (1973)
<i>C. pothifolia</i>		+	Hikino <i>et al.</i> (1973)
<i>C. shintenensis</i>		+	Hikino <i>et al.</i> (1973)
<i>C. wrightii</i>		+	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
<i>C. wrightii</i> var. <i>henryi</i>		+	Hikino <i>et al.</i> (1973)
<i>Comarum palustre</i>	Rosaceae (D) [Rosaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Comastoma tenellum</i>	Gentianaceae (D) [Gentianaceae (E)]	+	Berkenov & Datkayev (2017)
<i>Commelina africana</i>	Commelinaceae (M)	(+)	Crouzet <i>et al.</i> (2009)
<i>C. benghalensis</i>	[Commelinaceae (M)]	(+)	Crouzet <i>et al.</i> (2009)
<i>C. coelestris</i>		+	Crouzet <i>et al.</i> (2009)
		-	Dinan <i>et al.</i> (2020b)
<i>C. communis</i>		-	Volodin <i>et al.</i> (2002)
<i>C. dianthifolia</i>		-	Dinan <i>et al.</i> (2020b)
<i>C. erecta</i>		-	Crouzet <i>et al.</i> (2009)
<i>C. leiocarpa</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>C. ramulosa</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>C. rupicola</i>		(+)	Crouzet <i>et al.</i> (2009)

<i>C. virginica</i>		-	Dinan <i>et al.</i> (2020b)
<i>Coniogramme fraxinea</i>	Adiantaceae (F)	-	Yen <i>et al.</i> (1974)
<i>C. fraxinea</i> var. <i>intermedia</i>	[Pteridaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>C. intermedia</i>		+	Yen <i>et al.</i> (1974)
<i>C. japonica</i>		-	Takemoto <i>et al.</i> (1967c)
		-	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)
<i>Conioselinum tataricum</i>	Umbelliferae (D) [Apiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Conium maculatum</i>	Umbelliferae (D) [Apiaceae (E)]	(+)	Blackford & Dinan (1997b)
<i>Coniza canadensis</i>	Compositae (D) [Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Consolida ambigua</i>	Ranunculaceae (D) [Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2001t)
		-	Dinan <i>et al.</i> (2002a)
<i>Convolvulus arvensis</i>	Convolvulaceae (D) [Convolvulaceae (E)]	-	Blackford & Dinan (1997b)
<i>Coptis japonica</i>	Ranunculaceae (D)	-	Dinan <i>et al.</i> (2002a)
<i>C. quinquefolia</i>	[Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2002a)
<i>Coreopsis major</i>	Compositae (D) [Asteraceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Coriandrum sativum</i>	Umbelliferae (D) [Apiaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Coriaria microphylla</i>	Coriariaceae (D) [Coriariaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Corispermum hyssopifolium</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	+	Clément & Dinan (1991)
		-	Dinan <i>et al.</i> (1991)
		-/+	Dinan <i>et al.</i> (1998)
<i>C. leptopterum</i>		+	Dinan <i>et al.</i> (1998)
<i>C. nitidum</i>		-	Báthory <i>et al.</i> (1984)
		+	Dinan <i>et al.</i> (1998)
<i>Cornopteris (Athyrium) decurrenti-alata</i>	Woodsiaceae (F) [Athyriaceae (F)]	+	Imai <i>et al.</i> (1969d)
<i>Cornus kousa</i>	Cornaceae (D)	-	Blackford & Dinan (1997c)
<i>C. mas</i>	[Cornaceae (E)]	-	Blackford & Dinan (1997c)
<i>Coronaria (Lychnis) flos-cuculi</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	+	Revina <i>et al.</i> (1988)
		+	Mamadalieva <i>et al.</i> (2008)
<i>Cortaderia argentea</i>	Gramineae (M) [Poaceae (M)]	-	Dinan (1995a)
<i>Cortusa matthioli</i>	Primulaceae (D) [Primulaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Corylus avellana</i>	Betulaceae (D) [Betulaceae (E)]	-	Dinan <i>et al.</i> (2020a)

<i>Corynephorus canescens</i>	Gramineae (M) [Poaceae (M)]	-	Dinan (1995a)
<i>Coscinum fenestratum</i>	Menispermaceae (D) [Menispermaceae (E)]	+ + + + +	Madhavan <i>et al.</i> (2014) Sreejit (2014) Sreejit <i>et al.</i> (2018) Sreejit <i>et al.</i> (2019) Sreejit & Nelshi (2019)
<i>Cotoneaster amoenus</i>	Rosaceae (D)	-	Dinan <i>et al.</i> (2001d)
<i>C. frigidus</i>	[Rosaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>C. marguandii</i>		-	Dinan <i>et al.</i> (2001d)
<i>Crateva nurvala</i>	Capparaceae (D)	(+)	Volodin <i>et al.</i> (2018)
<i>C. religiosa</i>	[Capparaceae (E)]	+	Takemoto <i>et al.</i> (1967c)
<i>Crepidomanes latealatum</i>	Hymenophyllaceae (F) [Hymenophyllaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>Crepis paludosa</i>	Compositae (D)	+	Volodin <i>et al.</i> (2002)
<i>C. praemorsa</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>C. sibirica</i>		-	Volodin <i>et al.</i> (2002)
<i>C. tectorum</i>		-	Volodin <i>et al.</i> (2002)
<i>Crypsinus (Selligaea) hastatus</i>	Polypodiaceae (F) [Polypodiaceae (F)]	+ + + + + +	Yen <i>et al.</i> (1974) Hikino & Hikino (1970) Takemoto <i>et al.</i> (1973) Hikino <i>et al.</i> (1973) Hikino <i>et al.</i> (1973) Yen <i>et al.</i> (1974) Hikino <i>et al.</i> (1973)
<i>C. engleri</i>		+	Hikino <i>et al.</i> (1973)
<i>C. hastatus</i>		+	Hikino <i>et al.</i> (1973)
<i>C. quasidivaricatus</i>		+	Yen <i>et al.</i> (1974)
<i>C. yakushimensis</i>		+	Hikino <i>et al.</i> (1973)
<i>Cryptogramma crispa</i>	Adiantaceae (F) [Pteridaceae (F)]	- +	Hikino <i>et al.</i> (1973) Dreier (1987)
<i>Cryptomeria japonica</i>	Taxodiaceae (G) [Cupressaceae (G)]	-	Hoffmeister <i>et al.</i> (1967)
<i>Cryptostegia grandiflora</i>	Asclepiadaceae (D) [Apocynaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Ctenitis eatoni</i>	Dryopteridaceae (F)	+	Hikino <i>et al.</i> (1973)
<i>C. glabella</i>	[Dryopteridaceae (F)]	-	Russell & Fenemore (1971)
<i>C. hendersoni</i>		-	Hikino <i>et al.</i> (1973)
<i>C. maximowicziana</i>		+	Hikino <i>et al.</i> (1973)
<i>C. sikokiana</i>		-	Hikino <i>et al.</i> (1973)
<i>C. sinii</i>		+	Hikino <i>et al.</i> (1973)
<i>C. subglandulosa</i>		-/+	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)
<i>C. velutina</i>		-	Russell & Fenemore (1971)
<i>Cucubalus baccifer</i> (syn. <i>Silene baccifera</i>)	Caryophyllaceae (D) [Caryophyllaceae (E)]	+ + +	Cheng <i>et al.</i> (2001) Bespayeva <i>et al.</i> (2012) Dinan <i>et al.</i> (2020b)
<i>Cucumis metuliferus</i>	Cucurbitaceae (D) [Cucurbitaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Cucurbita moschata</i>	Cucurbitaceae (D)	-	Dinan <i>et al.</i> (2020a)

<i>C. pepo</i>	[Cucurbitaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Cudrania (Maclura) fricuspida</i>	Moraceae (D) [Moraceae (E)]	0.9	Wong <i>et al.</i> (1979)
<i>Cuminum cyminum</i>	Umbelliferae (D) [Apiaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Cupressus cashmiriana</i>	Cupressaceae (G)	-	Hoffmeister <i>et al.</i> (1967)
<i>C. funebris</i>	[Cupressaceae (G)]	+	Imai <i>et al.</i> (1969d)
<i>C. macrocarpa</i>		-	Hoffmeister <i>et al.</i> (1967)
<i>C. tularosa</i>		+	Rufaie <i>et al.</i> (2011)
<i>Cyanotis sp.</i>	Commelinaceae (M)	+	Li <i>et al.</i> (2016)
<i>Cyanotis arachnoidea</i>	[Commelinaceae (M)]	+	Chou & Lu (1980)
		+	Nien <i>et al.</i> (1978)
		+	Nie & Yue (1983)
		+	Nie & Qiu (1987)
		+	Wang <i>et al.</i> (1996)
		+	Zhou <i>et al.</i> (1996)
		+	Tan <i>et al.</i> (2001)
		+	Tan (2002)
		+	Tan <i>et al.</i> (2002a)
		+	Tan <i>et al.</i> (2002b)
		+	Tan <i>et al.</i> (2003a)
		+	Tan <i>et al.</i> (2003b)
		+	Zhu & An. (2003)
		+	Tan <i>et al.</i> (2005)
		+	Zuo (2006)
		+	Tan <i>et al.</i> (2011)
		+	Mu <i>et al.</i> (2011)
		+	Zhu <i>et al.</i> (2011)
		+	Cao & Zhou (2013)
		-	Sreejit (2014)
		+	Liu <i>et al.</i> (2014)
		+	Wang <i>et al.</i> (2014)
		+	Hunyadi <i>et al.</i> (2016)
		+	Issaadi <i>et al.</i> (2017)
		+	Lei <i>et al.</i> (2018)
		+	Tóth <i>et al.</i> (2021)
		+	Chen & Feng (2021)
<i>C. cristata</i>		-	Sreejit (2014)
<i>C. hirsuta</i>		+	Crouzet <i>et al.</i> (2009)
<i>C. kewensis</i> (syn. <i>C. beddomei</i>)		+	Crouzet <i>et al.</i> (2009)
<i>C. longifolia</i>		+	Crouzet <i>et al.</i> (2009)
<i>C. somaliensis</i>		+	Crouzet <i>et al.</i> (2009)
<i>C. speciosa</i>		+	Crouzet <i>et al.</i> (2009)
<i>C. vaga</i>		+	Santos <i>et al.</i> (1970)
		+	Santos <i>et al.</i> (1972)
		+	Gagalac-Nicolas & Sylianco (1981)
		+	Chua <i>et al.</i> (1982)
		+	Xu (2010)
<i>Cyathea boninsimensis</i>	Cyatheaceae (F)	+	Hikino <i>et al.</i> (1973)
<i>C. cooperi</i>	[Cyathaceae (F)]	+	Bergamasco & Horn (1983)
<i>C. dealbata</i>		+	Russell & Fenemore (1971)
<i>C. hancockii</i> (<i>Gymnosphaera denticulata</i>)		+	Yen <i>et al.</i> (1974)
<i>C. lepifera</i>		+	Yen <i>et al.</i> (1974)
<i>C. medullaris</i>		-	Russell & Fenemore (1971)
<i>C. mentteniana</i>		+	Yen <i>et al.</i> (1974)

<i>C. podophylla</i>		+	Hikino <i>et al.</i> (1973)
<i>C. spinulosa</i>		+	Yen <i>et al.</i> (1974)
		+	Yen <i>et al.</i> (1974)
<i>Cyathula capitata</i>	Amaranthaceae (D) [Amaranthaceae (E)]	+	Takemoto <i>et al.</i> (1967b)
		+	Takemoto <i>et al.</i> (1967d)
		+	Hikino <i>et al.</i> (1968)
		+	Takemoto <i>et al.</i> (1968d)
		+	Takemoto <i>et al.</i> (1968i)
		+	Hikino <i>et al.</i> (1969a)
		+	Hikino <i>et al.</i> (1970c)
		+	Hikino <i>et al.</i> (1970d)
		+	Hikino <i>et al.</i> (1971a)
		+	Hikino <i>et al.</i> (1971b)
		+	Hikino <i>et al.</i> (1971c)
		15	Wong <i>et al.</i> (1979)
		+	Hikino <i>et al.</i> (1970b)
		+	Chou & Lu (1980)
<i>C. officinalis</i>		+	Zhou <i>et al.</i> (2005)
		+	Okuzumi <i>et al.</i> (2005)
		+	Cao <i>et al.</i> (2017)
		+	Jiang <i>et al.</i> (2017)
<i>C. polycephala</i>		+	Sihra (1974)
<i>C. prostrata</i>		+	Shah & de Souza (1971)
		+	Sreejit (2014)
		+	Sreejit <i>et al.</i> (2018)
		+	Volodin <i>et al.</i> (2018)
		+	Sreejit <i>et al.</i> (2019)
<i>Cyathula sp.</i>		+	Wu & Zhang (2017)
<i>Cycas revoluta</i>	Cycadaceae (G) [Cycadaceae (Cyc)]	-	Takemoto <i>et al.</i> (1967c)
<i>Cyclamen hederifolium</i>	Primulaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>C. persicum</i>	[Primulaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Cyclea barbata</i>	Menispermaceae (D)	+	Wang <i>et al.</i> (2018)
<i>C. hypoglauca</i>	[Menispermaceae (E)]	+	Zhang <i>et al.</i> (2017)
<i>C. peltata</i>		-	Sreejit (2014)
<i>Cycloloma atriplicifolium</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Cyclosorus acuminatus</i>	Thelypteridaceae (F) [Thelypteridaceae (F)]	+	Imai <i>et al.</i> (1969c)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
		-	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
<i>C. dentatus</i>		-	Hikino <i>et al.</i> (1973)
<i>C. goggilodus</i>		-	Hikino <i>et al.</i> (1973)
<i>C. parasiticus</i>		-	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
<i>C. subpubescens</i>		-	Hikino <i>et al.</i> (1973)
<i>C. taiwanensis</i>		+	Yen <i>et al.</i> (1974)
<i>C. triphyllus</i>		+	Hikino <i>et al.</i> (1973)
<i>Cydonia oblonga</i>	Rosaceae (D) [Rosaceae (E)]	-	Dinan <i>et al.</i> (2020a)

<i>Cymbopogon exaltatus</i>	Gramineae (M) [Poaceae (M)]	-	Dinan (1995a)
<i>Cynanchum forrestii</i>	Asclepiadaceae (D) [Apocynaceae (E)]	+	Liu <i>et al.</i> (2007)
<i>Cynara scolymus</i> cv green globe	Compositae (D) [Asteraceae (E)]	-	Blackford <i>et al.</i> (1996)
<i>C. scolymus</i> cv large green		- (+) (+)	Blackford & Dinan (1997a) Blackford <i>et al.</i> (1996) Blackford & Dinan (1997a)
<i>Cynoglossum hungaricum</i>	Boraginaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>C. officinale</i>	[Boraginaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Cyphomandra betacea</i>	Solanaceae (D) [Solanaceae (E)]	- -	Savchenko <i>et al.</i> (2000) Dinan <i>et al.</i> (2020a)
<i>Cypripedium calceolus</i>	Orchidaceae (M)	-	Volodin <i>et al.</i> (2002)
<i>C. parviflorum pubescens</i>	[Orchidaceae (M)]	-	Dinan <i>et al.</i> (2020b)
<i>Cystopteris fragilis</i>	Woodsiaceae (F) [Cryopteridaceae (F)]	- -	Hikino <i>et al.</i> (1973) Dreier (1987)
D			
<i>Dacrycarpus imbricatus (Podocarpus kawaii)</i>	Podocarpaceae (G) [Podocarpaceae (G)]	+ +	Thuy <i>et al.</i> (2017) Quan <i>et al.</i> (2018)
<i>Dacrydium bidwillii</i>	Podocarpaceae (G)	-	Russell & Fenemore (1970)
<i>D. biforme</i>	[Podocarpaceae (G)]	-	Russell & Fenemore (1970)
<i>D. colensoi</i>		-	Russell & Fenemore (1970)
<i>D. cupressinum</i>		+	Russell & Fenemore (1970)
<i>D. intermedium</i>		+	Russell & Fenemore (1970)
		+	Russell <i>et al.</i> (1971)
		+	Russell <i>et al.</i> (1972)
		+	Russell & Fraser (1973)
		+	Blunt <i>et al.</i> (1979)
<i>D. kirkii</i>		-	Russell & Fenemore (1970)
<i>D. laxifolium</i>		+	Russell & Fenemore (1970)
<i>D. pierrei</i>		+	Hou <i>et al.</i> (1982)
<i>Dactylis glomerata</i>	Gramineae (M) [Poaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Dactylorhiza fuchsia</i>	Orchidaceae (M)	-	Dinan <i>et al.</i> (2020b)
<i>D. incarnata</i>	[Orchidaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>D. maculata</i>		-	Volodin <i>et al.</i> (2002)
<i>Dalbergia hupeana</i>	Leguminosae-P. (D) [Fabaceae (E)]	6.0 +	Wong <i>et al.</i> (1979) Chou & Lu (1980)
<i>Danae racemosa</i>	Asparagaceae (M) [Asparagaceae (M)]	-	Dinan <i>et al.</i> (2020b)
<i>Daphne mezereum</i>	Thymelaeaceae (D) [Thymelaeaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Datura arborea</i>	Solanaceae (D)	-	Savchenko <i>et al.</i> (2000)
<i>D. ferox</i>	[Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)

<i>D. innoxia</i>		-	Savchenko <i>et al.</i> (2000)
<i>D. metel</i> (<i>D. fastuosa</i>)		-	Savchenko <i>et al.</i> (2000)
<i>D. meteloides</i>		-	Savchenko <i>et al.</i> (2000)
<i>D. quercifolia</i>		-	Savchenko <i>et al.</i> (2000)
<i>D. sanguinea</i>		-	Savchenko <i>et al.</i> (2000)
<i>D. stramonium</i>		(+)	Blackford & Dinan (1997c)
		-/(+)	Savchenko <i>et al.</i> (2000)
		+	Rufaie <i>et al.</i> (2011)
<i>D. suaveolens</i>		-	Savchenko <i>et al.</i> (2000)
<i>D. tatula</i>		-	Savchenko <i>et al.</i> (2000)
<i>Daucus carota</i>	Umbelliferae (D)	1.0	Wong <i>et al.</i> (1979)
	[Apiaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>D. carota</i>		-	Blackford <i>et al.</i> (1996)
<i>Davallia formosana</i>	Davalliaceae (F)	+	Yen <i>et al.</i> (1974)
<i>D. mariesii</i>	[Davalliaceae (F)]	+	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
<i>D. tasmanii</i>		-	Russell & Fenemore (1971)
<i>Delphinium cardinale</i>	Ranunculaceae (D)	-	Dinan <i>et al.</i> (2002a)
<i>D. chinense</i>	[Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2002a)
<i>D. consolida</i>		-	Dinan <i>et al.</i> (2002a)
<i>D. cuneatum</i>		-	Dinan <i>et al.</i> (2002a)
<i>D. elatum</i>		-	Dinan <i>et al.</i> (2002a)
		-	Volodin <i>et al.</i> (2002)
<i>D. grandiflorum</i>		-	Dinan <i>et al.</i> (2002a)
<i>D. hybridum</i> (= <i>D. schmallhauseni</i>)		-	Dinan <i>et al.</i> (2002a)
<i>D. nudicaule</i>		-	Dinan <i>et al.</i> (2002a)
<i>D. requienii</i>		-	Dinan <i>et al.</i> (2002a)
<i>D. staphisagria</i>		-	Dinan <i>et al.</i> (2002a)
<i>D. tatsienense</i>		-	Dinan <i>et al.</i> (2002a)
<i>D. zalil</i>		-	Dinan <i>et al.</i> (2002a)
<i>Dennstaedtia hirsuta</i>	Dennstaedtiaceae (F)	-	Hikino <i>et al.</i> (1973)
	[Dennstaedtiaceae (F)]	-	Takemoto <i>et al.</i> (1967c)
<i>D. scabra</i>		-	Yen <i>et al.</i> (1974)
<i>D. scabra</i>		-	Hikino <i>et al.</i> (1973)
<i>D. scabra</i> var. <i>glabrescens</i>		-	Hikino <i>et al.</i> (1973)
<i>D. scandens</i>		-	Yen <i>et al.</i> (1974)
<i>D. wilfordii</i>		+	Takemoto <i>et al.</i> (1967c)
		+/-	Hikino <i>et al.</i> (1973)
<i>Deschampsia caespitosa</i>	Gramineae (M)	-	Dinan (1995a)
<i>D. parviflora</i>	[Poaceae (M)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Desmodium elegans</i>	Leguminosae-P. (D)	(+)	Dinan <i>et al.</i> (2001d)
	[Fabaceae (E)]		
<i>Dianthus amurensis</i>	Caryophyllaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
	[Caryophyllaceae (E)]	-	Zibareva <i>et al.</i> (2003)
<i>D. arenarius</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. armeria</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. arvenensis</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. barbatus</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. borbasii</i>		+	Agabekova <i>et al.</i> (2010)
<i>D. carthusianorum</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. caryophyllus</i>		-	Blackford & Dinan (1997b)
		-	Zibareva <i>et al.</i> (2003)
<i>D. chinensis</i>		-	Zibareva <i>et al.</i> (2003)

<i>D. deltoides</i>		-	Volodin <i>et al.</i> (2002)
		-	Zibareva <i>et al.</i> (2003)
		+	Zibareva <i>et al.</i> (2007a)
<i>D. giganteiformis</i>		+	Bespayeva <i>et al.</i> (2012)
<i>D. gratianopolitanus</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. helenae</i>		+	Yusupova <i>et al.</i> (2019)
		+	Yusupova <i>et al.</i> (2020)
<i>D. hoeltzeri</i>		+	Saatov <i>et al.</i> (1990b)
		+	Saatov <i>et al.</i> (1999)
<i>D. hungaricus</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. kitaibelii</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. knappii</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. monspessulanus</i>		-	Dinan <i>et al.</i> (2020b)
<i>D. plumarius</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. seguieri</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. shinanensis</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. subacaulis</i>		-	Zibareva <i>et al.</i> (2003)
<i>D. superbus</i>		-	Revina <i>et al.</i> (1988)
		-	Volodin <i>et al.</i> (2002)
<i>D. versicolor</i>		-	Revina <i>et al.</i> (1988)
<i>Diapensia lapponica</i>	Diapensiaceae (D) [Diapensiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Dichodon (Cerastium) cerastoides</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	-	Revina <i>et al.</i> (1988)
		-	Volodin <i>et al.</i> (2002)
<i>Dichorisandra hexandra</i>	Commelinaceae (M)	+	Calderón <i>et al.</i> (2009)
<i>D. reginae</i>	[Commelinaceae (M)]	(+)	Crouzet <i>et al.</i> (2009)
<i>D. thyrsiflora</i>		-(+)	Crouzet <i>et al.</i> (2009)
<i>Dicksonia squarrosa</i>	Dicksoniaceae (F)	-	Russell & Fenemore (1971)
<i>D. fibrosa</i>	[Dicksoniaceae (F)]	-	Russell & Fenemore (1971)
<i>Dicranopteris dichotoma (D. linearis)</i>	Gleicheniaceae (F)	-	Yen <i>et al.</i> (1974)
<i>D. linearis</i>	[Gleicheniaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>Dictyocline griffithii</i> var. <i>pinnatifida</i>	Thelypteridaceae (F)	+	Hikino <i>et al.</i> (1973)
<i>D. (Stenogramma) griffithii</i> var. <i>wilfordii</i>	[Thelypteridaceae (F)]	+	Yen <i>et al.</i> (1974)
<i>Didymanthus roei</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Digitalis ciliata</i>	Plantaginaceae (D)	+	Gvazava & Kukoladze (2010)
<i>D. davisiana</i>	[Plantaginaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>D. purpurea</i>		+	Gvazava & Kukoladze (2010)
<i>Dioscorea dumentorum</i>	Dioscoreaceae (M)	+	Sautour <i>et al.</i> (2007)
	[Dioscoreaceae (M)]	+	Sautour <i>et al.</i> (2008)
<i>D. persimilis</i>		+	Volodin <i>et al.</i> (2018)
<i>Diplazium dilatatum (Athyrium maximum)</i>	Aspleniaceae (F)	+	Yen <i>et al.</i> (1974)
<i>D. donianum (A. aphanoneuron)</i>	[Athyriaceae (F)]	+	Yen <i>et al.</i> (1974)
		+	Hikino <i>et al.</i> (1976)
<i>D. esculentum (A. esculentum)</i>		+	Yen <i>et al.</i> (1974)
		+	Watanabe <i>et al.</i> (2021)
<i>D. kawakami (A. procerum)</i>		-	Yen <i>et al.</i> (1974)
<i>D. mettenianum (A. mettenianum)</i>		+	Yen <i>et al.</i> (1974)
<i>D. owaseanum</i>		+	Hikino <i>et al.</i> (1973)
<i>D. sibiricum</i>		+	Revina & Gureeva (1985)

<i>D. taiwanense</i>		(+)	Olennikov & Kashchenko (2018)
<i>D. yakumontanum</i>		+	Hikino <i>et al.</i> (1973)
		+	Hikino <i>et al.</i> (1973)
<i>Diploclisia glaucescens</i>	Menispermaceae (D) [Menispermaceae (E)]	+	Miller <i>et al.</i> (1985)
		+	Bandara <i>et al.</i> (1989)
		+	Jayasinghe (1995)
		+	Jayasinghe <i>et al.</i> (2002)
		+	Jayasinghe <i>et al.</i> (2003a)
		+	Jayasinghe <i>et al.</i> (2003b)
		+	Huang <i>et al.</i> (2003)
		+	Jayasinghe <i>et al.</i> (2005)
		+	Mao <i>et al.</i> (2013)
		+	Sreejit (2014)
		+	Fang <i>et al.</i> (2017)
		+	Sreejit <i>et al.</i> (2018)
		+	Sreejit <i>et al.</i> (2019)
<i>Diplopterygium rufopilosum</i>	Gleicheniaceae (F) [Gleicheniaceae (F)]	+	Hu <i>et al.</i> (2014)
<i>Disporum cantoniense</i>	Convallariaceae (M)	+	Chen <i>et al.</i> (2018)
<i>D. smithii</i>	[Colchicaceae (M)]	-	Dinan <i>et al.</i> (2020b)
<i>Diospyros kaki</i>	Ebenaceae (D)	+	Matsuoka <i>et al.</i> (1969)
	[Ebenaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>D. lotus</i>		-	Dinan <i>et al.</i> (2020b)
<i>Doellingeria scabra</i>	Compositae (D) [Asteraceae (E)]	24	Wong <i>et al.</i> (1979)
<i>Doodia aspersa</i>	Blechnaceae (F)	+	Bergamasco & Horn (1983)
<i>D. media</i>	[Blechnaceae (F)]	+	Russell & Fenemore (1971)
<i>Draba languinosa</i>	Cruciferae (D) [Brassicaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Dracocephalum thymiflorum</i>	Labiatae (D) [Lamiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Dracontomelon duperreanum</i>	Anacardiaceae (D) [Anacardiaceae (E)]	+	Volodin <i>et al.</i> (2018)
<i>Drosera rotundifolia</i>	Droseraceae (D) [Droseraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Drymaria cordata</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	-	Sreejit (2014)
<i>Dryopteris arguta</i>	Dryopteridaceae (F)	+	Dreier (1987)
<i>D. austriaca</i>	[Dryopteridaceae (F)]	+	Hikino <i>et al.</i> (1973)
		-	Revina & Gureeva (1985)
<i>D. bissetiana</i>		+	Hikino <i>et al.</i> (1973)
		+	Takemoto <i>et al.</i> (1967c)
<i>D. carthusiana</i>		-	Revina & Gureeva (1985)
<i>D. championi</i>		-	Hikino <i>et al.</i> (1973)
<i>D. chinensis</i>		+	Takemoto <i>et al.</i> (1967c)
		+	Hikino <i>et al.</i> (1973)
		0.2	Wong <i>et al.</i> (1979)

<i>D. commixta</i>		-	Hikino <i>et al.</i> (1973)
<i>D. crassirhizoma</i>		-	Takemoto <i>et al.</i> (1967c)
		+	Hikino <i>et al.</i> (1973)
<i>D. cycadina</i>		+	Hikino <i>et al.</i> (1973)
<i>D. dickinsii</i>		-	Hikino <i>et al.</i> (1973)
<i>D. erythrosora</i>		-	Hikino <i>et al.</i> (1973)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
		+	Yen <i>et al.</i> (1974)
<i>D. erythrosora</i> var. <i>cystolepidota</i>		-	Hikino <i>et al.</i> (1973)
<i>D. erythrosora</i> var. <i>koidzumiana</i>		+	Hikino <i>et al.</i> (1973)
<i>D. filix-mas</i>		-	Revina & Gureeva (1985)
<i>D. formosana</i>		+	Hikino <i>et al.</i> (1973)
		-	Takemoto <i>et al.</i> (1967c)
<i>D. fragrans</i>		-	Revina & Gureeva (1985)
<i>D. fuscipes</i>		-	Hikino <i>et al.</i> (1973)
<i>D. gymnosora</i>		-	Hikino <i>et al.</i> (1973)
<i>D. hayatai</i>		+	Hikino <i>et al.</i> (1973)
<i>D. hondoensis</i>		+	Hikino <i>et al.</i> (1973)
<i>D. indusiata</i>		-	Hikino <i>et al.</i> (1973)
<i>D. lacera</i>		+	Hikino <i>et al.</i> (1973)
		+	Takemoto <i>et al.</i> (1967c)
<i>D. laeta</i>		+	Hikino <i>et al.</i> (1973)
<i>D. lepidopoda</i>		+	Yen <i>et al.</i> (1974)
<i>D. melanocarpa</i>		+	Hikino <i>et al.</i> (1973)
<i>D. monticola</i>		+	Hikino <i>et al.</i> (1973)
<i>D. polita</i>		+	Hikino <i>et al.</i> (1973)
<i>D. polylepis</i>		-	Hikino <i>et al.</i> (1973)
<i>D. pycnopteroides</i>		-	Hikino <i>et al.</i> (1973)
<i>D. sabacai</i>		-	Hikino <i>et al.</i> (1973)
<i>D. scottii</i>		-	Yen <i>et al.</i> (1974)
<i>D. sieboldii</i>		-	Yen <i>et al.</i> (1974)
		-	Hikino <i>et al.</i> (1973)
<i>D. sordidipes</i>		+	Hikino <i>et al.</i> (1973)
<i>D. sparsa</i>		-	Hikino <i>et al.</i> (1973)
<i>D. tasiroi</i>		+	Hikino <i>et al.</i> (1973)
<i>D. thelypteris</i>		+	Imai <i>et al.</i> (1969d)
<i>D. tokyoensis</i>		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
		+	Hikino <i>et al.</i> (1973)
<i>D. uniformis</i>		+	Hikino <i>et al.</i> (1973)
		-	Takemoto <i>et al.</i> (1967c)
<i>D. varia</i>		+	Matsuoka <i>et al.</i> (1969)
		+	Yen <i>et al.</i> (1974)
<i>D. varia</i> var. <i>sacrosancta</i>		+	Hikino <i>et al.</i> (1973)
<i>D. varia</i> var. <i>setosa</i>		+	Imai <i>et al.</i> (1969d)
		+	Hikino <i>et al.</i> (1973)
<i>D. varia</i> var. <i>subtripinnata</i>		+	Hikino <i>et al.</i> (1973)
<i>Drypis spinosa</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	-	Zibareva <i>et al.</i> (2003)
<i>Duboisia cleichhardtii</i>	Solanaceae (D)	-	Savchenko <i>et al.</i> (2000)
<i>D. hopwoodii</i>	[Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
<i>Duschekia fruticosa</i>	Betulaceae (D) [Betulaceae (E)]	+	Volodin <i>et al.</i> (2002)
<i>Dysphania glomulifera</i>	Chenopodiaceae (D)	-	Dinan <i>et al.</i> (1998)
<i>D. inflata</i>	[Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)

<i>D. kalpari</i>		-	Dinan <i>et al.</i> (1998)
<i>D. littoralis</i>		-	Dinan <i>et al.</i> (1998)
<i>D. plantaginella</i>		-	Dinan <i>et al.</i> (1998)
<i>D. rhadinostachya</i>		-	Dinan <i>et al.</i> (1998)
<i>D. simulans</i>		-	Dinan <i>et al.</i> (1998)
E			
<i>Ecballium elaterium</i>	Cucurbitaceae (D) [Cucurbitaceae (E)]	+	Matsuoka <i>et al.</i> (1969)
<i>Echinops grijisii</i>	Compositae (D)	2.0	Wong <i>et al.</i> (1979)
<i>E. tienschanicus</i>	[Asteraceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Echium pininana</i>	Boraginaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>E. vulgare</i>	[Boraginaceae (E)]	-	Blackford & Dinan (1997a)
<i>Edraianthus pumilio</i>	Campanulaceae (D) [Campanulaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Einadia nutans</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	+	Dinan <i>et al.</i> (1998)
<i>Elaphoglossum tosaense</i>	Lomariopsidaceae (F)	+	Hikino <i>et al.</i> (1973)
<i>E. yoshinagae</i>	[Dryopteridaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>Elasis hirsuta</i>	Commelinaceae (M) [Commelinaceae (M)]	(+)	Crouzet <i>et al.</i> (2009)
<i>Eleocharis palustris</i>	Cyperaceae (M) [Cyperaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Elettaria cardamomum</i>	Zingiberaceae (M) [Zingiberaceae (M)]	-	Dinan <i>et al.</i> (2020a)
<i>Elisanthe (Silene) viscosa</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	-	Revina <i>et al.</i> (1988)
<i>Elodea canadensis</i>	Hydrocharitaceae (M) [Hydrocharitaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Elymus fibrosus</i>	Gramineae (M) [Poaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Elytrigia repens</i>	Gramineae (M) [Poaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Empetrum hermaphroditum</i>	Empetraceae (D) [Ericaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Encephalartos</i> sp.	Zamiaceae (G) [Zamiaceae (Cyc)]	-	Hoffmeister <i>et al.</i> (1967)
<i>Enchylaena tomentosa</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Epilobium angustifolium</i>	Onagraceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>E. davuricum</i>	[Onagraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>E. hirsutum</i>		-	Dinan <i>et al.</i> (2001d)
<i>E. montanum</i>		-	Dinan <i>et al.</i> (2001d)
<i>E. palustre</i>		-	Volodin <i>et al.</i> (2002)

<i>E. tetragonium</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>Equisetum arvense</i>	Equisetaceae (FA) [Equisetaceae (Equ)]	- +	Hikino <i>et al.</i> (1973) Clément & Dinan (1991)
<i>E. hyemale</i>		-	Hikino <i>et al.</i> (1973)
<i>E. ramosissimum</i> ssp. <i>debile</i>		-	Yen <i>et al.</i> (1974)
<i>E. ramosissimum</i> var. <i>japonicum</i>		-	Hikino <i>et al.</i> (1973)
<i>Eragrostis abessinica</i>	Gramineae (M)	-	Dinan (1995a)
<i>E. interrupta</i>	[Poaceae (M)]	-	Dinan (1995a)
<i>Eranthis hyemalis</i>	Ranunculaceae (D) [Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2002a)
<i>Eremogone (Arenaria) formosa</i>	Caryophyllaceae (D)	-	Revina <i>et al.</i> (1988)
<i>E. saxatilis</i>	[Caryophyllaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Erigeron alpinus</i>	Compositae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>E. acris</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>E. glabratus</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>E. uniflorus</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>Eriophorum scheuchzeri</i>	Cyperaceae (M)	-	Volodin <i>et al.</i> (2002)
<i>E. vaginatum</i>	[Cyperaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Eriophyton wallchii</i>	Labiatae (D) [Lamiaceae (E)]	+	Yi <i>et al.</i> (2004)
<i>Eryngium agavifolium</i>	Umbelliferae (D)	-	Blackford & Dinan (1997a)
<i>E. maritimum</i>	[Apiaceae (E)]	+	Dinan <i>et al.</i> (2001d)
<i>E. planum</i>		-	Blackford & Dinan (1997a)
<i>Erysimum cheiranthoides</i>	Cruciferae (D)	-	Volodin <i>et al.</i> (2002)
<i>E. perofskianum</i>	[Brassicaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Erythronium dens-canis</i>	Liliaceae (M)	-	Dinan <i>et al.</i> (2001c)
<i>E. montanum</i>	[Liliaceae (M)]	-	Dinan <i>et al.</i> (2001c)
<i>E. revolutum</i> var. <i>smithii</i>		-	Dinan <i>et al.</i> (2001c)
<i>Eucryphia glutinosa</i>	Eucrypiaceae (D) [Cunoniaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Euonymus alatus</i>	Celastraceae (D)	-	Takemoto <i>et al.</i> (1967c)
<i>E. europaeus</i>	[Celastraceae (E)]	-	Blackford & Dinan (1997c)
<i>E. japonicus</i>		-	Takemoto <i>et al.</i> (1967c)
<i>E. oxyphyllus</i>		-	Takemoto <i>et al.</i> (1967c)
<i>Euphoria (Dimocarpus) longan</i>	Sapindaceae (D) [Sapindaceae (E)]	1.9	Wong <i>et al.</i> (1979)
<i>Euphorbia borodini</i>	Euphorbiaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>E. characias wulfenii</i>	[Euphorbiaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>E. cognata</i>		-	Dinan <i>et al.</i> (2020b)
<i>E. myrsinites</i>		-	Dinan <i>et al.</i> (2020b)
<i>E. platyphyllos</i>		-	Dinan <i>et al.</i> (2001d)
<i>E. villosa</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>E. watanabei</i>		-	Takemoto <i>et al.</i> (1967c)
<i>Euphrasia frigida</i>	Scrophulariaceae (D) [Orabachaceae (E)]	-	Volodin <i>et al.</i> (2002)

F

<i>Fagopyrum esculentum</i>	Polygonaceae (D) [Polygonaceae (E)]	-	Takemoto <i>et al.</i> (1967c)
<i>Fallopia convolvulus</i>	Polygonaceae (D) [Polygonaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Fatsia japonica</i>	Araliaceae (D) [Araliaceae (E)]	-	Takemoto <i>et al.</i> (1967c)
<i>Festuca amethystina</i>	Gramineae (M)	-	Dinan (1995a)
<i>F. ovina</i>	[Poaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>F. ovina glauca</i>		(+)	Dinan (1995a)
<i>Fibraurea chloroleuca</i>	Menispermaceae (D)	+	Dai <i>et al.</i> (1993)
<i>F. tinctoria</i>	[Menispermaceae (E)]	+	Su <i>et al.</i> (2007)
<i>Ficus carica</i>	Moraceae (D)	-	Takemoto <i>et al.</i> (1967c)
	[Moraceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>F. microcarpa</i>		-	Takemoto <i>et al.</i> (1967c)
<i>Filaginella uliginosa</i>	Compositae (D) [Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Filipendula ulmaria</i>	Rosaceae (D) [Rosaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Foeniculum vulgare</i>	Umbelliferae (D) [Apiaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Fornicium uniflorum</i> (= <i>Stemmacantha uniflorum</i>)	Compositae (D) [Asteraceae (E)]	+	Nikolaeva <i>et al.</i> (2017)
<i>Fragaria vesca</i>	Rosaceae (D) [Rosaceae (E)]	-	Blackford & Dinan (1997c)
		-	Volodin <i>et al.</i> (2002)
<i>F. x ananassa</i>		-	Dinan <i>et al.</i> (2020a)
<i>Frangula (Rhamnus) alnus</i>	Rhamnaceae (D) [Rhamnaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Fraxinus chinensis</i>	Oleaceae (D)	3.0	Wong <i>et al.</i> (1979)
<i>F. excelsior</i>	[Oleaceae (E)]	-	Blackford & Dinan (1997c)
<i>F. ornus</i>		(+)	Blackford & Dinan (1997c)
<i>Fritillaria acropetala</i>	Liliaceae (M)	-	Dinan <i>et al.</i> (2001c)
<i>F. biflora</i>	[Liliaceae (M)]	-	Dinan <i>et al.</i> (2001c)
<i>F. imperialis</i> var. <i>lutea maxima</i>		-	Dinan <i>et al.</i> (2001c)
<i>F. involucrata</i>		-	Dinan <i>et al.</i> (2001c)
<i>F. lanceolata</i>		-	Dinan <i>et al.</i> (2001c)
<i>F. meleagris</i>		-	Dinan <i>et al.</i> (2001c)
<i>F. pallidiflora</i>		-	Dinan <i>et al.</i> (2001c)
<i>F. persica</i>		-	Dinan <i>et al.</i> (2001c)
<i>F. pudica</i>		-	Dinan <i>et al.</i> (2001c)
<i>F. tubiformis</i>		-	Dinan <i>et al.</i> (2001c)
<i>Froehlichia floridana</i>	Amaranthaceae (D) [Amaranthaceae (E)]	+ +	Sarker <i>et al.</i> (1998d) Wang <i>et al.</i> (2009)
<i>Fumaria officinalis</i>	Papaveraceae (D)	-	Volodin <i>et al.</i> (2002)

[Papaveraceae (E)]

G

<i>Galega officinalis</i>	Leguminosae-P. (D) [Fabaceae (E)]	+	Matsuoka <i>et al.</i> (1969)
<i>Galeopsis bifida</i>	Labiatae (D)	-	Volodin <i>et al.</i> (2002)
<i>G. speciosa</i>	[Lamiaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
		-	Volodin <i>et al.</i> (2002)
<i>G. tetrahit</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>Galium boreale</i>	Rubiaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>G. mollugo</i>	[Rubiaceae (E)]	-	Blackford & Dinan (1997c)
		-	Volodin <i>et al.</i> (2002)
<i>Galtonia candicans</i>	Hyacinthaceae (M)	(+)	Dinan <i>et al.</i> (2001d)
<i>G. princeps</i>	[Asparagaceae (M)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Garcinia mangostana</i>	Guttiferae (D) [Clusiaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Garrya elliptica</i>	Garryaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>G. fremontii</i>	[Garryaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Gastrolychnis angustiflora</i>	Caryophyllaceae (D)	+	Volodin <i>et al.</i> (2002)
<i>G. apetala</i>	[Caryophyllaceae (E)]	+	Revina <i>et al.</i> (1988)
<i>G. brachypetala</i>		+	Revina <i>et al.</i> (1988)
		+	Olennikov & Kashchenko (2018)
<i>G. gracilis</i>		+	Olennikov & Kashchenko (2018)
<i>G. saxatilis</i>		+	Olennikov & Kashchenko (2018)
<i>G. tristis</i>		+	Revina <i>et al.</i> (1988)
		+	Olennikov (2018a)
		+	Olennikov & Kashchenko (2018)
<i>Gentiana algida</i>	Gentianaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>G. asclepiadea</i>	[Gentianaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>G. cruciata</i>		-	Volodin <i>et al.</i> (2002)
<i>G. scabra</i> var. <i>buergeri</i>		-	Takemoto <i>et al.</i> (1967c)
<i>G. tibetica</i>		-	Dinan <i>et al.</i> (2020b)
<i>Geranium maderense</i>	Geraniaceae (D)	-	Dinan <i>et al.</i> (2001d)
<i>G. molle</i>	[Geraniaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>G. renardii</i>		-	Dinan <i>et al.</i> (2020b)
<i>G. rotundifolium</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>G. sylvaticum</i>		-	Volodin <i>et al.</i> (2002)
<i>G. versicolor</i>		-	Dinan <i>et al.</i> (2020b)
<i>Geum allepicum</i>	Rosaceae (D)	-	Dinan <i>et al.</i> (2001d)
<i>G. magellanicum</i>	[Rosaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>G. rivale</i>		-	Volodin <i>et al.</i> (2002)
<i>Gibasis geniculata</i>	Commelinaceae (M) [Commelinaceae (M)]	(+)	Crouzet <i>et al.</i> (2009)
<i>Gilia millefoliata</i>	Polemoniaceae (D) [Polemoniaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Ginkgo biloba</i>	Ginkgoaceae (G) [Ginkgoaceae (Gin)]	-	Imai <i>et al.</i> (1967)
		-	Takemoto <i>et al.</i> (1967c)
		-	Hoffmeister <i>et al.</i> (1967)

<i>Girgensohnia minima</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Glechoma hederacea</i>	Labiatae (D) [Lamiaceae (E)]	-	Takemoto <i>et al.</i> (1967c)
<i>G. hederacea</i>		-	Volodin <i>et al.</i> (2002)
		4.0	Wong <i>et al.</i> (1979)
<i>Gleichenia glauca</i>	Gleicheniaceae (F)	+	Hikino & Hikino (1970)
<i>G. microphylla</i>	[Gleicheniaceae (F)]	-	Russell & Fenemore (1971)
<i>G. circinata</i>		-	Russell & Fenemore (1971)
<i>G. cunninghamii</i>		+	Russell & Fenemore (1971)
<i>G. linearis</i>		-	Russell & Fenemore (1971)
<i>Globularia bisnagarica</i>	Globulariaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>G. repens</i>	[Plantaginaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>G. trichosantha</i>		-	Dinan <i>et al.</i> (2020b)
<i>G. valentina</i>		-	Dinan <i>et al.</i> (2020b)
<i>G. vulgaris</i>		-	Dinan <i>et al.</i> (2001d)
<i>Glyceria notata</i>	Gramineae (M) [Poaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Glycine max</i>	Leguminosae-P. (D) [Fabaceae (E)]	-	Takemoto <i>et al.</i> (1967c)
		-	Blackford <i>et al.</i> (1996)
<i>Gnetum</i> sp.	Gnetaceae (G) [Gnetaceae (G)]	-	Hoffmeister <i>et al.</i> (1967)
<i>Gomphrena affinis</i>	Amaranthaceae (D)	+	Sarker <i>et al.</i> (1996a)
	[Amaranthaceae (E)]	+	Savchenko <i>et al.</i> (1998b)
<i>G. canescens</i>		+	Sarker <i>et al.</i> (1996a)
		+	Savchenko <i>et al.</i> (1998b)
<i>G. celosioides</i>		+	Banerji <i>et al.</i> (1971)
		+	Sreejit (2014)
		+	Sreejit <i>et al.</i> (2018)
<i>G. cunninghamii</i>		+	Savchenko <i>et al.</i> (1998b)
<i>G. dispersa</i>		+	Sarker <i>et al.</i> (1996a)
		+	Savchenko <i>et al.</i> (1998b)
<i>G. globosa</i>		2.7	Wong <i>et al.</i> (1979)
		-	Sarker <i>et al.</i> (1996a)
		-	Savchenko <i>et al.</i> (1998b)
<i>G. haageana</i>		+	Sarker <i>et al.</i> (1996a)
		+	Savchenko <i>et al.</i> (1998b)
<i>G. hybrida</i>		-	Sarker <i>et al.</i> (1996a)
		-	Savchenko <i>et al.</i> (1998b)
<i>G. virgata</i>		+	Marinho <i>et al.</i> (2021)
<i>Gonocormus minutus</i> (syn. <i>Crepidomanes minutum</i>)	Hymenophyllaceae (F) [Hymenophyllaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>Goodyera repens</i>	Orchidaceae (M) [Orchidaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Gossypium hirsutum</i>	Malvaceae (D) [Malvaceae (E)]	(+)	Blackford <i>et al.</i> (1996)
<i>Grammitis billardieri</i>	Grammitidaceae (F) [Polypodiaceae (F)]	+	Russell & Fenemore (1971)

<i>Grewia biloba</i>	Tiliaceae (D) [Malvaceae (E)]	2.0	Wong <i>et al.</i> (1979)
<i>Grindelia nana</i>	Compositae (D) [Asteraceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Gymnosphaera (Cyathea) denticulata</i>	Cyatheaceae (F) [Cyatheaceae (F)]	+/-	Hikino <i>et al.</i> (1973)
<i>Gypsophila acutifolia</i>	Caryophyllaceae (D)	+	Bespayeva <i>et al.</i> (2012)
<i>G. alsinoides</i>	[Caryophyllaceae (E)]	+	Agabekova <i>et al.</i> (2010)
<i>G. altissima</i>		-	Revina <i>et al.</i> (1988)
<i>G. cephalotes</i>		-	Revina <i>et al.</i> (1988)
<i>G. elegans</i>		-	Zibareva <i>et al.</i> (2003)
		+	Bespayeva <i>et al.</i> (2012)
<i>G. pacifica</i>		-	Zibareva <i>et al.</i> (2003)
<i>G. paniculata</i>		-	Revina <i>et al.</i> (1988)
		-	Zibareva <i>et al.</i> (2003)
<i>G. patrinii</i>		-	Revina <i>et al.</i> (1988)
		+	Bespayeva <i>et al.</i> (2012)
<i>G. perfoliata</i>		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
<i>G. repens</i>		-	Zibareva <i>et al.</i> (2003)
<i>G. rupestris</i>		+	Agabekova <i>et al.</i> (2010)
<i>G. sericea</i>		-	Revina <i>et al.</i> (1988)
H			
<i>Hablitzia tamnoides</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Halimione portulacoides</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	+	Clément & Dinan (1991)
		+	Dinan <i>et al.</i> (1991)
		+	Dinan <i>et al.</i> (1998)
<i>Halimium halimifolium</i>	Cistaceae (D) [Cistaceae (E)]	+	Imai <i>et al.</i> (1969d)
<i>Halocnemum strobilaceum</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Halosarcia doleiformis</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Haloxylon salicornicum</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Hammada articulata ssp. scoparia</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Haplopappus rehderi</i>	Compositae (D) [Asteraceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Hedera helix</i>	Aquifoliaceae (D) [Araliaceae (E)]	-	Clément & Dinan (1991)
<i>Helianthus annuus</i>	Compositae (D)	(+)	Blackford <i>et al.</i> (1996)
<i>H. tuberosus</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (1993)
		-	Dinan <i>et al.</i> (2020a)

<i>Helleborus abchasicus</i>	Ranunculaceae (D)	+	Hardman & Benjamin (1976)
	[Ranunculaceae (E)]	+	Beridze <i>et al.</i> (2020)
<i>H. argutifolius</i>		-	Dinan <i>et al.</i> (2002a)
<i>H. atrorubens</i>		+	Hardman & Benjamin (1976)
<i>H. atrorubens</i> f. <i>cupreus</i>		+	Dinan <i>et al.</i> (2002a)
<i>H. bocconeii</i> ssp. <i>bocconeii</i>		+	Hardman & Benjamin (1976)
<i>H. caucasicus</i>		+	Muzashvili & Kemertelidze (2015)
		+	Bassarello <i>et al.</i> (2008)
		+	Martucciello <i>et al.</i> (2017)
		+	Beridze <i>et al.</i> (2020)
<i>H. corsicus</i>		(+)	Dinan <i>et al.</i> (2002a)
<i>H. croaticus</i>		+	Dinan <i>et al.</i> (2002a)
<i>H. cyclophyllus</i>		+	Hardman & Benjamin (1976)
		+	Hardman & Benjamin (1980)
		+	Dinan <i>et al.</i> (2002a)
<i>H. dumentorum</i>		+	Dinan <i>et al.</i> (2002a)
<i>H. dumentorum</i> ssp. <i>dumentorum</i>		+	Hardman & Benjamin (1976)
<i>H. dumentorum</i> ssp. <i>atorrubens</i>		+	Hardman & Benjamin (1976)
<i>H. foetidissima</i>		+	Dinan <i>et al.</i> (2001d)
		+	Dinan <i>et al.</i> (2002a)
<i>H. foetidus</i>		-	Hardman & Benjamin (1976)
		(+)	Dinan <i>et al.</i> (2002a)
		-	Dinan <i>et al.</i> (2020b)
<i>H. guttatus</i>		+	Hardman & Benjamin (1976)
<i>H. lividus</i>		+	Dinan <i>et al.</i> (2002a)
<i>H. lividus</i> ssp. <i>lividus</i>		-	Hardman & Benjamin (1976)
<i>H. lividus</i> ssp. <i>corsicus</i>		-	Hardman & Benjamin (1976)
<i>H. multifidus</i>		+	Dinan <i>et al.</i> (2002a)
<i>H. multifidus</i> ssp. <i>multifidus</i>		+	Hardman & Benjamin (1976)
<i>H. niger</i>		+	Imai <i>et al.</i> (1969c)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
		+	Glombitza <i>et al.</i> (1989)
		+	Dinan <i>et al.</i> (2002a)
		+	Yokosuka <i>et al.</i> (2021)
<i>H. niger</i> ssp. <i>niger</i>		+	Liedtke <i>et al.</i> (1997)
<i>H. odorus</i>		+	Kissmer & Wichtl (1987)
		+	Dinan <i>et al.</i> (2002a)
<i>H. odorus</i> ssp. <i>cyclophyllus</i>		+	Tsiftoglou <i>et al.</i> (2018)
		+	Brillatz <i>et al.</i> (2020)
<i>H. odorus</i> ssp. <i>laxus</i>		+	Colombo <i>et al.</i> (1990)
		+	Colombo & Tome (1993)
<i>H. orientalis</i>		+	Hardman & Benjamin (1976)
		+	Dinan <i>et al.</i> (2002a)
		+	Akin & Anil (2007)
<i>H. purpurascens</i>		+	Kissmer & Wichtl (1987)
		+	Dinan <i>et al.</i> (2002a)
<i>H. x sternii</i>		-	Dinan <i>et al.</i> (2002a)
<i>H. thibetanus</i>		+	Yang <i>et al.</i> (2010)
<i>H. torquatus</i>		+	Meng <i>et al.</i> (2001a)
		+	Dinan <i>et al.</i> (2002a)
<i>H. vesicarius</i>		(+)	Dinan <i>et al.</i> (2002a)
<i>H. viridis</i>		+	Colombo <i>et al.</i> (1990)
		+	Dinan <i>et al.</i> (2002a)
<i>H. viridis</i> ssp. <i>occidentalis</i>		+	Hardman & Benjamin (1976)
<i>H. viridis</i> ssp. <i>viridis</i>		+	Colombo <i>et al.</i> (1990)
<i>Hemerocallis lilioasphodelus</i>	Hemerocalidaceae (M)	-	Dinan <i>et al.</i> (2020b)
	[Asphodelaceae (M)]		

<i>Hepatica triloba</i>	Ranunculaceae (D) [Ranunculaceae (E)]	+	Dinan <i>et al.</i> (2002a)
<i>Heracleum sibiricum</i>	Umbelliferae (D) [Apiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Heuchera americana</i> <i>H. cylindrica</i>	Saxifragaceae (D) [Saxifragaceae (E)]	- (+)	Dinan <i>et al.</i> (2020b) Dinan <i>et al.</i> (2001d)
<i>Hibiscus esculentus</i> <i>H. mutabilis</i> <i>H. splendens</i>	Malvaceae (D) [Malvaceae (E)]	- + + +	Blackford <i>et al.</i> (1996) Saeng-ngam <i>et al.</i> (on-line) Wong <i>et al.</i> (1979) Imai <i>et al.</i> (1969d) Matsuoka <i>et al.</i> (1969)
<i>Hicriopteris (Dicranopteris) glauca</i> <i>H. glauca</i>	Gleicheniaceae (F) [Gleichenaceae (F)]	- + +	Takemoto <i>et al.</i> (1967c) Takemoto <i>et al.</i> (1973) Hikino <i>et al.</i> (1973)
<i>Hieracium alpinum</i> <i>H. altipes</i> <i>H. bombycinum</i> <i>H. humile</i> <i>H. murorum</i> <i>Hieracium</i> sp. <i>Hieracium</i> sp. <i>H. umbellatum</i>	Compositae (D) [Asteraceae (E)]	- - - (+) + - - -	Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002) Dinan <i>et al.</i> (2001d) Dinan <i>et al.</i> (2001d) Dinan <i>et al.</i> (2001d) Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002)
<i>Hippuris tetraphylla</i> <i>H. vulgaris</i>	Hippuridaceae (D) [Plantaginaceae (E)]	- -	Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002)
<i>Histiopteris incisa</i>	Dennstaedtiaceae (F) [Dennstaedtiaceae (F)]	- + -	Russell & Fenemore (1971) Hikino <i>et al.</i> (1973) Yen <i>et al.</i> (1974)
<i>Holboellia coriacea</i>	Lardizabalaceae (D) [Lardizabalaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Holmbergia tweedii</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	+	Dinan <i>et al.</i> (1998)
<i>Hordeum vulgare</i>	Gramineae (M) [Poaceae (M)]	- -	Clément & Dinan (1991) Dinan (1995a)
<i>Hovenia dulcis</i>	Rhamnaceae (D) [Rhamnaceae (E)]	-	Takemoto <i>et al.</i> (1967c)
<i>Humata (Davallia) repens</i>	Davalliaceae (F) [Davalliaceae (F)]	+	Hikino <i>et al.</i> (1973)
<i>Humulus lupulus</i> var. <i>cordifolius</i> <i>H. lupulus</i> <i>H. scandens</i>	Cannabaceae (D) [Cannabaceae (E)]	- - + +	Takemoto <i>et al.</i> (1967c) Blackford & Dinan (1997a) Blackford & Dinan (1997c) Xu <i>et al.</i> (2014)
<i>Hydrangea heteromalia</i>	Hydrangaceae (D) [Hydrangeaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Hydrocharis morsus-ranae</i>	Hydrocharitaceae (M) [Hydrocharitaceae (M)]	-	Volodin <i>et al.</i> (2002)

<i>Hylocereus (Selenicereus) megalanthus</i>	Cactaceae (D)	-	Dinan <i>et al.</i> (2020a)
<i>H.(Selenicereus) undatus</i>	[Cactaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Hymenophyllum barbatum</i>	Hymenophyllaceae (F)	-	Hikino <i>et al.</i> (1973)
<i>H. sanguinolentum</i>	[Hymenophyllaceae (F)]	-	Russell & Fenemore (1971)
<i>H. scabrum</i>		-	Russell & Fenemore (1971)
<i>H. malingii</i>		-	Russell & Fenemore (1971)
<i>H. bivalve</i>		-	Russell & Fenemore (1971)
<i>Hyoscyamus albus</i>	Solanaceae (D)	-	Savchenko <i>et al.</i> (2000)
<i>H. niger</i>	[Solanaceae (E)]	-	Blackford & Dinan (1997c)
		-	Savchenko <i>et al.</i> (2000)
<i>Hypericum japonicum</i>	Guttiferae (D)	1.4	Wong <i>et al.</i> (1979)
<i>H. perforatum</i>	[Hypericaceae (E)]	-	Volodin <i>et al.</i> (2002)
		-	Dinan <i>et al.</i> (2020b)
<i>H. quadrangulum</i>		-	Volodin <i>et al.</i> (2002)
<i>H. tetrapterum</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>Hypolepis alte-gracillima</i>	Dennstaedtiaceae (F)	-	Yen <i>et al.</i> (1974)
<i>H. alte-gracilis</i>	[Dennstaedtiaceae (F)]	+	Hikino <i>et al.</i> (1973)
<i>H. millefolium</i>		+	Russell & Fenemore (1971)
<i>H. punctata</i>		+/-	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)
<i>H. tenuifolia</i>		-	Russell & Fenemore (1971)
I			
<i>Ilex cinerea</i>	Aquifoliaceae (D)	+	Volodin <i>et al.</i> (2018)
<i>I. crenata</i>	[Aquifoliaceae (E)]	-	Takemoto <i>et al.</i> (1967c)
<i>I. integra</i>		+	Matsuoka <i>et al.</i> (1969)
<i>I. latifolia</i>		-	Takemoto <i>et al.</i> (1967c)
<i>I. purpurea</i>		(+)	Volodin <i>et al.</i> (2018)
<i>I. rotunda</i>		+	Volodin <i>et al.</i> (2018)
<i>Incarvillea forestii</i>	Bignoniaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
	[Bignoniaceae (E)]		
<i>Inula helenium</i>	Compositae (D)	+	Volodin <i>et al.</i> (1993)
<i>I. salicina</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Iochroma gesnerioides</i>	Solanaceae (D)	+	Savchenko <i>et al.</i> (2000)
	[Solanaceae (E)]		
<i>Ipheion uniflorum</i>	Alliaceae (M)	+	Nakamura <i>et al.</i> (1994)
	[Amaryllidaceae (M)]	+	Dinan <i>et al.</i> (2020b)
<i>Ipomoea alba</i>	Convolvulaceae (D)	-	Sreejit (2014)
<i>I. batatus</i>	[Convolvulaceae (E)]	0.9	Wong <i>et al.</i> (1979)
		-	Blackford <i>et al.</i> (1996)
		-	Dinan <i>et al.</i> (2020a)
<i>I. calonyction</i>		+	Canonica <i>et al.</i> (1973)
		+	Canonica <i>et al.</i> (1972)
		+	Canonica <i>et al.</i> (1973b)
		+	Sardini & Krepinsky (1974)
		+	Canonica <i>et al.</i> (1975)
		+	Canonica <i>et al.</i> (1977a)
		+	Canonica <i>et al.</i> (1977b)
		+/-	Austin (2000)

<i>I. congesta</i>		+	Volodin <i>et al.</i> (2018)
<i>I. hederacea</i>		+	Ghosh & Ladda (2006)
		+	Sumayya (2005)
		+	Zia-Ul-Haq (2012)
<i>I. lindheimeri</i>		-	Dinan <i>et al.</i> (2020b)
<i>I. obscura</i>		-	Sreejit (2014)
<i>I. pes-caprae ssp. pes-caprae</i>		-	Sreejit (2014)
<i>Iresine herbstii</i>	Amaranthaceae (D)	+	Takemoto <i>et al.</i> (1967d)
<i>I. lindenii</i>	[Amaranthaceae (E)]	+	Takemoto <i>et al.</i> (1967d)
<i>Iris chrysographes</i>	Iridaceae (M)	-	Dinan <i>et al.</i> (2001d)
<i>I. crocea</i>	[Iridaceae (M)]	+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
<i>I. sibirica</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>I. spuria</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>I. ventricosa</i>		-	Dinan <i>et al.</i> (2001d)
<i>I. vicaria</i>		-	Dinan <i>et al.</i> (2020b)
<i>I. warleyensis</i>		-	Dinan <i>et al.</i> (2020b)
J			
<i>Jasione laevis (J. perennis)</i>	Campanulaceae (D) [Campanulaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Jasminum beesianum</i>	Oleaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>J. nudiflorum</i>	[Oleaceae (E)]	-	Blackford & Dinan (1997c)
<i>J. officinale</i>		-	Blackford & Dinan (1997c)
<i>Juglans regia</i>	Juglanaceae (D) [Juglandaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Juncus filiformis</i>	Juncaceae (M)	-	Volodin <i>et al.</i> (2002)
<i>J. nodulosus</i>	[Juncaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Juniperus communis</i>	Cupressaceae (G) [Cupressaceae (G)]	-	Volodin <i>et al.</i> (2002)
<i>Justicia adhatoda</i>	Acanthaceae (D)	-	Sreejit & Nelshi (2019)
<i>J. gendarussa</i>	[Acanthaceae (E)]	-	Sreejit (2014)
K			
<i>Kickxia elatine</i>	Scrophulariaceae (D) [Plantaginaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Kishazi (Bergenia) ciliata</i>	Saxifragaceae (D) [Saxifragaceae (E)]	1.3	Wong <i>et al.</i> (1979)
<i>Kitaibelia vitifolia</i>	Malvaceae (D) [Malvaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Klaseopsis chinensis</i> (syn. <i>Serratula chinensis</i>)	Compositae (D) [Asteraceae (E)]	+	Ling <i>et al.</i> (2009)
		+	Yang <i>et al.</i> (2010)
<i>Knautia sp.</i>	Dipsacaceae (D) [Caprifoliaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Kniphofia typhoides</i>	Asphodelaceae (M) [Asphodelaceae (M)]	-	Dinan <i>et al.</i> (2020b)

<i>Kochia laniflora</i>	Chenopodiaceae (D)	-	Báthory <i>et al.</i> (1984)
<i>K. prostrata</i>	[Amaranthaceae (E)]	+	Bespayeva <i>et al.</i> (2012)
<i>K. scoparia</i>		+	Báthory <i>et al.</i> (1984)
		5.0	Wong <i>et al.</i> (1979)
		+	Chou & Lu (1980)
		+	Dinan (1994)
		+	Dinan (1995b)
		+	Sakimbai <i>et al.</i> (2019)
<i>Koeleria pohleana</i>	Gramineae (M)	-	Volodin <i>et al.</i> (2002)
	[Poaceae (M)]		
L			
<i>Lachenalia reflexa</i>	Hyacinthaceae (M)	-	Dinan <i>et al.</i> (2020b)
	[Asparagaceae (M)]		
<i>Lactuca sativa</i>	Compositae (D)	+	Whitehead & Sellheyer (1982)
	[Asteraceae (E)]	-	Blackford & Dinan (1997b)
		-	Volodin <i>et al.</i> (2002)
<i>L. tatarica</i>		-	Volodin <i>et al.</i> (2002)
<i>Lagurus ovatus</i>	Gramineae (M)	-	Dinan (1995a)
	[Poaceae (M)]		
<i>Lamarckia aurea</i>	Gramineae (M)	-	Dinan (1995a)
	[Poaceae (M)]		
<i>Lamium album</i>	Labiatae (D)	(+)	Savchenko <i>et al.</i> (2001)
	[Lamiaceae (E)]	+	Volodin <i>et al.</i> (2002)
<i>L. barbatum</i>		15	Wong <i>et al.</i> (1979)
		+	Chou & Lu (1980)
<i>L. galeobdolon</i>		(+)	Savchenko <i>et al.</i> (2001)
<i>L. maculatum</i>		+	Savchenko <i>et al.</i> (2001)
		+	Cui <i>et al.</i> (2003)
		+	Deng <i>et al.</i> (2003)
<i>L. maculatum</i> var. <i>kansuense</i>		+	Deng <i>et al.</i> (2005)
<i>L. purpureum</i>		(+)	Savchenko <i>et al.</i> (2001)
		+	Alipieva <i>et al.</i> (2003)
<i>L. takesimense</i>		+	Nugroho <i>et al.</i> (2018)
<i>Lansium parasiticum</i>	Meliaceae (D)	-	Dinan <i>et al.</i> (2020b)
	[Meliaceae (E)]		
<i>Lastrea (Coryphopteris) angulariloba</i>	Thelypteridaceae (F)	+	Hikino <i>et al.</i> (1973)
<i>L. (Oreopteris) beddomei</i>	[Thelypteridaceae (F)]	+	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
<i>L. cystopteroides</i>		+	Hikino <i>et al.</i> (1973)
<i>L. (Oreopteris) decursive-pinnata</i>		+/-	Hikino <i>et al.</i> (1973)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
<i>L. dryopteris</i>		-	Hikino <i>et al.</i> (1973)
<i>L. glanduligera</i>		+/-	Hikino <i>et al.</i> (1973)
<i>L. glanduligera</i> var. <i>hyalostegia</i>		+	Hikino <i>et al.</i> (1973)
<i>L. gracilescens</i>		+	Hikino <i>et al.</i> (1973)
<i>L. japonica</i>		+	Hikino <i>et al.</i> (1973)
		+	Matsuoka <i>et al.</i> (1969)
		+	Imai <i>et al.</i> (1969d)
		+	Takemoto <i>et al.</i> (1967c)
		+	Hikino & Hikino (1970)
		+	Takemoto <i>et al.</i> (1973)

<i>L. laxa</i>		-	Hikino <i>et al.</i> (1973)
<i>L. oligophlebia</i>		+	Hikino <i>et al.</i> (1973)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
<i>L. oligophlebia</i> var. <i>lasiocarpa</i>		+	Hikino <i>et al.</i> (1973)
<i>L. omeiensis</i>		-	Hikino <i>et al.</i> (1973)
<i>L. phlegopteris</i>		-	Hikino <i>et al.</i> (1973)
<i>L. quelpaertensis</i>		+	Hikino <i>et al.</i> (1973)
<i>L. subochthodes</i>		+/-	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
<i>L. thelypteris</i>		+	Matsuoka <i>et al.</i> (1969)
		+	Takemoto <i>et al.</i> (1967)
		+	Takemoto <i>et al.</i> (1968b)
		+	Takemoto <i>et al.</i> (1967c)
		+	Hikino & Hikino (1970)
		+	Takemoto <i>et al.</i> (1973)
		+	Hikino <i>et al.</i> (1973)
<i>L. totta</i>		+/-	Hikino <i>et al.</i> (1973)
<i>L. uraiensis</i>		+	Hikino <i>et al.</i> (1973)
<i>Lathyrus pratensis</i>	Leguminosae-P. (D)	-	Volodin <i>et al.</i> (2002)
<i>L. sativus</i>	[Fabaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>L. sylvestris</i>		-	Dinan <i>et al.</i> (2001d)
<i>L. tuberosus</i>		-	Dinan <i>et al.</i> (2020b)
<i>Lavandula angustifolia</i>	Labiatae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>L. latifolia</i>	[Lamiaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>L. multifida</i>		-	Dinan <i>et al.</i> (2001d)
<i>L. stoechas</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>Ledum decumbens</i>	Ericaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>L. palustre</i> (syn. <i>Rhododendron tomentosum</i>)	[Ericaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Legousia speculatum-veneris</i>	Campanulaceae (D) [Campanulaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Leibnitzia anandria</i>	Compositae (D) [Asteraceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Lemmaphyllum microphyllum</i>	Polypodiaceae (F) [Polypodiaceae (F)]	+	Takemoto <i>et al.</i> (1968a)
		+	Takemoto <i>et al.</i> (1967c)
		+	Takemoto <i>et al.</i> (1968g)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
		+	Takemoto <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
		+	Hikino <i>et al.</i> (1973)
<i>Lemna minor</i>	Lemnaceae (M)	-	Volodin <i>et al.</i> (2002)
<i>L. trisulca</i>	[Araceae (M)]	+	Volodin <i>et al.</i> (2002)
<i>Leonorus sibiricus</i>	Labiatae (D) [Lamiaceae (E)]	24 +	Wong <i>et al.</i> (1979) Chou & Lu (1980)
<i>Leonurus cardiaca</i>	Labiatae (D) [Lamiaceae (E)]	(+)	Volodin <i>et al.</i> (2002)
<i>Leontodon autumnalis</i>	Compositae (D) [Asteraceae (E)]	+	Volodin <i>et al.</i> (2002)

<i>Ligustrum congestum</i>	Oleaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>L. lucidum</i>	[Oleaceae (E)]	4.2	Wong <i>et al.</i> (1979)
		+	Chou & Lu (1980)
<i>L. obtusifolium</i>		-	Takemoto <i>et al.</i> (1967c)
<i>L. ovalifolium</i>		(+)	Blackford & Dinan (1997c)
<i>Lilium auratum</i> var. <i>platyphyllum</i>	Liliaceae (M)	-	Dinan <i>et al.</i> (2001c)
<i>L. carniolicum</i>	[Liliaceae (M)]	-	Dinan <i>et al.</i> (2001c)
<i>L. columbianum</i>		-	Dinan <i>et al.</i> (2001c)
<i>L. cordatum</i> var. <i>glehnii</i>		-	Takemoto <i>et al.</i> (1967c)
<i>L. davidii</i>		-	Dinan <i>et al.</i> (2001c)
<i>L. formosanum</i> var. <i>pricei</i>		-	Dinan <i>et al.</i> (2001c)
		(+)	Dinan <i>et al.</i> (2001d)
<i>L. giganteum</i> var. <i>yunnanense</i>		-	Dinan <i>et al.</i> (2001c)
<i>L. martagon</i>		-	Dinan <i>et al.</i> (2001c)
<i>L. pilosiusculum</i>		-	Dinan <i>et al.</i> (2001c)
<i>L. pyrenaicum</i>		-	Dinan <i>et al.</i> (2001c)
<i>L. regale</i>		-	Dinan <i>et al.</i> (2001c)
		(+)	Dinan <i>et al.</i> (2001d)
<i>Limnanthes alba</i>	Limnanthaceae (D)	+	Sarker <i>et al.</i> (1997b)
	[Limnanthaceae (E)]	+	Meng <i>et al.</i> (2001c)
		+	Preston-Mafham & Dinan (2002)
		+	Stevens <i>et al.</i> (2008)
<i>L. bakeri</i>		+	Sarker <i>et al.</i> (1997b)
<i>L. douglasii</i>		+	Sarker <i>et al.</i> (1997b)
		+	Girault (1998)
<i>L. floccosa</i>		+	Sarker <i>et al.</i> (1997b)
<i>L. gracilis</i>		+	Sarker <i>et al.</i> (1997b)
<i>L. montana</i>		+	Sarker <i>et al.</i> (1997b)
<i>Limonium bellidifolium</i>	Plumbaginaceae (D)	(+)	Whiting <i>et al.</i> (1998)
<i>L. binervosum</i>	[Plumbaginaceae (E)]	+	Whiting <i>et al.</i> (1998)
<i>L. bonduelli</i>		-	Whiting <i>et al.</i> (1998)
<i>L. cosyrense</i>		-	Whiting <i>et al.</i> (1998)
<i>L. gmelinii</i>		-	Whiting <i>et al.</i> (1998)
<i>L. gerberi</i>		+	Whiting <i>et al.</i> (1998)
<i>L. otolepis</i>		+	Whiting <i>et al.</i> (1998)
<i>L. peregrinum</i>		+	Whiting <i>et al.</i> (1998)
<i>L. perezii</i>		+	Whiting <i>et al.</i> (1998)
<i>L. puberulum</i>		-	Whiting <i>et al.</i> (1998)
<i>L. purpuratum</i>		-	Whiting <i>et al.</i> (1998)
<i>L. ramosissimum</i>		+	Whiting <i>et al.</i> (1998)
		+	Dinan <i>et al.</i> (2001d)
<i>L. sinense</i>		-	Whiting <i>et al.</i> (1998)
<i>L. sinuatum</i>		-	Whiting <i>et al.</i> (1998)
<i>L. suworowii</i>		-	Whiting <i>et al.</i> (1998)
<i>L. tataricum</i>		-	Whiting <i>et al.</i> (1998)
<i>L. tetragonum</i>		-	Whiting <i>et al.</i> (1998)
<i>Limosella aquatica</i>	Scrophulariaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Scrophulariaceae (E)]		
<i>Linaria purpurea</i>	Scrophulariaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>L. vulgaris</i>	[Plantaginaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Lindsaea commixta</i>	Dennstaedtiaceae (F)	-	Hikino <i>et al.</i> (1973)
<i>L. cultrata</i>	[Lindsaeaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>L. javanensis</i>		-	Hikino <i>et al.</i> (1973)

<i>L. orbiculata</i> var. <i>chienenii</i>		-	Hikino <i>et al.</i> (1973)
<i>L. trichomanoides</i>		-	Russell & Fenemore (1971)
<i>Linnaea borealis</i>	Caprifoliaceae (D) [Caprifoliaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Linum usitatissimum</i>	Linaceae (D) [Linaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Liquidambar formosana</i>	Hamamelidaceae (D) [Altingiaceae (E)]	1.5	Wong <i>et al.</i> (1979)
<i>Listera (Neottia) ovata</i>	Orchidaceae (M) [Orchidaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Litchi chinensis</i>	Sapindaceae (D) [Sapindaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Lloydia serotina</i>	Liliaceae (M) [Liliaceae (M)]	+	Dinan <i>et al.</i> (2001c)
<i>Loasa triphylla</i>	Loasaceae (D) [Loasaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Lobelia chinensis</i>	Campanulaceae (D) [Campanulaceae (E)]	5.0	Wong <i>et al.</i> (1979)
<i>L. siphilitica</i>		+	Chou & Lu (1980)
		(+)	Dinan <i>et al.</i> (2001d)
<i>Lonicera japonica</i>	Caprifoliaceae (D)	-	Takemoto <i>et al.</i> (1967c)
<i>L. gracilipes</i> var. <i>glabra</i>	[Caprifoliaceae (E)]	-	Takemoto <i>et al.</i> (1967c)
<i>L. pallasii</i>		-	Volodin <i>et al.</i> (2002)
<i>L. periclymenum</i>		-	Blackford & Dinan (1997c)
<i>Loxogramme grammitoides</i>	Polypodiaceae (F)	+	Hikino <i>et al.</i> (1973)
<i>L. salicifolia</i>	[Polypodiaceae (F)]	+	Hikino <i>et al.</i> (1973)
<i>L. saziran</i>		+	Hikino <i>et al.</i> (1973)
<i>Loxsoma cunninghamii</i>	Loxsomataceae (F) [Loxsomataceae (F)]	-	Russell & Fenemore (1971)
<i>Lupinaster pentaphyllus</i>	Leguminosae-P. (D) [Fabaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Lupinus chamissonis</i>	Leguminosae-P. (D) [Fabaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Luzula confusa</i>	Juncaceae (M)	-	Volodin <i>et al.</i> (2002)
<i>L. parviflora</i>	[Juncaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Lychnis alba</i>	Caryophyllaceae (D)	+	Bespayeva <i>et al.</i> (2012)
<i>L. alpina</i>	[Caryophyllaceae (E)]	-	Zibareva <i>et al.</i> (1995)
		-	Dinan <i>et al.</i> (2020b)
<i>L. apetala</i>		-	Zibareva <i>et al.</i> (2003)
<i>L. arkwrightii</i>		+	Zibareva <i>et al.</i> (1995)
		+	Zibareva <i>et al.</i> (2003)
		+	Zibareva <i>et al.</i> (2007a)
		+	Dinan <i>et al.</i> (2020b)
<i>L. chalconica</i>		+	Imai <i>et al.</i> (1969c)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)

	+	Revina <i>et al.</i> (1988)
	+	Zibareva & Sviridova (1989)
	+	Zibareva <i>et al.</i> (1991a)
	+	Zibareva <i>et al.</i> (1991b)
	+	Zibareva <i>et al.</i> (1995)
	+	Zibareva <i>et al.</i> (1995)
	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007a)
	+	Bespayeva <i>et al.</i> (2012)
	+	Dinan <i>et al.</i> (2020b)
<i>L. cognata</i>	+	Zibareva <i>et al.</i> (1995)
	+	Zibareva <i>et al.</i> (2007a)
	+	Novozhilova <i>et al.</i> (2014)
<i>L. compacta</i>	+	Novozhilova <i>et al.</i> (2015)
<i>L. coronaria</i>	-	Zibareva <i>et al.</i> (1995)
	+	Abubakirov (1984)
	+	Dai <i>et al.</i> (2002)
	+	Zibareva <i>et al.</i> (2003)
	+	Bespayeva <i>et al.</i> (2012)
	+	Dinan <i>et al.</i> (2020b)
<i>L. coronaria occulata</i>	+	Zibareva <i>et al.</i> (2003)
<i>L. flos-cuculi</i>	+	Girault <i>et al.</i> (1990)
	+	Revina <i>et al.</i> (1988)
	+	Louden <i>et al.</i> (2001)
	+	Bathori <i>et al.</i> (2001)
	+	Zibareva <i>et al.</i> (2003)
	+	Malinski <i>et al.</i> (2019)
	+	Dinan <i>et al.</i> (2020b)
	+	Malinski <i>et al.</i> (2021)
<i>L. flos-cuculi nana</i>	+	Zibareva <i>et al.</i> (2003)
<i>L. flos-jovis</i>	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007a)
	+	Dinan <i>et al.</i> (2020b)
<i>L. fulgens</i>	+	Baltaev <i>et al.</i> (1986)
	+	Abubakirov (1984)
	+	Zibareva <i>et al.</i> (1995)
	+	Novozhilova <i>et al.</i> (2014)
	+	Novozhilova <i>et al.</i> (2015)
<i>L. gracilis</i>	-	Zibareva <i>et al.</i> (1995)
<i>L. haageana</i>	+	Zibareva <i>et al.</i> (1995)
	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
<i>L. kiusiana</i>	+	Zibareva <i>et al.</i> (2007a)
<i>L. miqueliana</i>	+	Imai <i>et al.</i> (1969c)
	+	Imai <i>et al.</i> (1969d)
	+	Matsuoka <i>et al.</i> (1969)
	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007a)
<i>L. plena</i>	+	Zibareva <i>et al.</i> (2003)
<i>L. samojedorum</i>	+	Volodin <i>et al.</i> (2002)
<i>L. sibirica</i>	+	Zibareva <i>et al.</i> (1995)
	+	Zibareva <i>et al.</i> (2007a)
	+	Novozhilova <i>et al.</i> (2015)
<i>L. villosula</i>	+	Zibareva <i>et al.</i> (1995)
	+	Zibareva <i>et al.</i> (2007a)
<i>L. viscaria</i>	+	Zibareva <i>et al.</i> (2003)
	-	Dinan <i>et al.</i> (2020b)
<i>L. wilfordii</i>	+	Zibareva <i>et al.</i> (1991a)
	+	Zibareva <i>et al.</i> (1995)

			+	Zibareva <i>et al.</i> (2003)
			+	Zibareva <i>et al.</i> (2007a)
			+	Novozhilova <i>et al.</i> (2014)
			+	Novozhilova <i>et al.</i> (2015)
<i>L. yunnanensis</i>			-	Zibareva <i>et al.</i> (2003)
			+	Bespayeva <i>et al.</i> (2012)
			(+)	Dinan <i>et al.</i> (2020b)
<i>Lycium australe</i>	Solanaceae (D)		-	Savchenko <i>et al.</i> (2000)
<i>L. barbarum</i>	[Solanaceae (E)]		-	Savchenko <i>et al.</i> (2000)
			-	Dinan <i>et al.</i> (2020b)
<i>L. chinese</i>			-	Savchenko <i>et al.</i> (2000)
<i>L. ferocissimum</i>			-(+)	Savchenko <i>et al.</i> (2000)
<i>Lycopersicon esculentum</i>	Solanaceae (D)		-	Blackford <i>et al.</i> (1996)
	[Solanaceae (E)]		-	Blackford & Dinan (1997b)
			-	Savchenko <i>et al.</i> (2000)
			-	Dinan <i>et al.</i> (2020a)
<i>Lycopodium alpinum</i> var. <i>planiramulosum</i>	Lycopodiaceae (FA)		-	Hikino <i>et al.</i> (1973)
<i>L. annotinum</i>	[Lycopodiaceae (FA)]		-	Hikino <i>et al.</i> (1973)
<i>L. cernuum</i>			-	Hikino <i>et al.</i> (1973)
			-	Yen <i>et al.</i> (1974)
<i>L. clavatum</i>			-	Yen <i>et al.</i> (1974)
<i>L. clavatum</i> var. <i>nipponicum</i>			-	Hikino <i>et al.</i> (1973)
<i>L. complanatum</i>			-	Hikino <i>et al.</i> (1973)
			-	Yen <i>et al.</i> (1974)
<i>L. fargesii</i>			-	Hikino <i>et al.</i> (1973)
			-	Yen <i>et al.</i> (1974)
<i>L. fordii</i>			-	Hikino <i>et al.</i> (1973)
			-	Yen <i>et al.</i> (1974)
<i>L. inundatum</i>			-	Hikino <i>et al.</i> (1973)
<i>L. obscurum</i>			-	Hikino <i>et al.</i> (1973)
<i>L. phlegmaria</i>			-	Hikino <i>et al.</i> (1973)
			-	Yen <i>et al.</i> (1974)
<i>L. serratum</i>			+	Matsuoka <i>et al.</i> (1969)
			+	Imai <i>et al.</i> (1969d)
			-	Takemoto <i>et al.</i> (1967c)
			-	Hikino <i>et al.</i> (1973)
<i>L. sieboldi</i>			-	Hikino <i>et al.</i> (1973)
<i>L. sitchense</i>			-	Hikino <i>et al.</i> (1973)
<i>Lycopus americanus</i>	Labiatae (D)		(+)	Dinan <i>et al.</i> (2001d)
<i>L. lucidus</i> var. <i>hirtus</i>	[Lamiaceae (E)]		2.2	Wong <i>et al.</i> (1979)
<i>Lygodium japonicum</i>	Schizaeaceae (F)		-	Takemoto <i>et al.</i> (1967c)
	[Lygodiaceae (F)]		5.1	Wong <i>et al.</i> (1979)
			-	Hikino <i>et al.</i> (1973)
			-	Yen <i>et al.</i> (1974)
			+	Chou & Lu (1980)
			+	Zhu <i>et al.</i> (2009)
<i>L. microstachyum</i>			-	Yen <i>et al.</i> (1974)
<i>L. scandens</i> var. <i>microphyllum</i>			-	Yen <i>et al.</i> (1974)
<i>Lysimachia clethroides</i>	Primulaceae (D)		-	Dinan <i>et al.</i> (2020b)
<i>L. mauritiana</i>	[Primulaceae (E)]		-	Takemoto <i>et al.</i> (1967c)
<i>L. vulgaris</i>			-	Volodin <i>et al.</i> (2002)

M

<i>Magnolia champaca</i>	Magnoliaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>M. obobata</i>	[Magnoliaceae (Mag)]	-	Takemoto <i>et al.</i> (1967c)
<i>Maianthemum bifolium</i>	Convallariaceae (M)	-	Volodin <i>et al.</i> (2002)
	[Asparagaceae (M)]		
<i>Maireana aphylla</i>	Chenopodiaceae (D)	-	Dinan <i>et al.</i> (1998)
<i>M. appressa</i>	[Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>M. astrostricha</i>		-	Dinan <i>et al.</i> (1998)
<i>M. atkinsiana</i>		-	Dinan <i>et al.</i> (1998)
<i>M. brevifolia</i>		-	Dinan <i>et al.</i> (1998)
<i>M. carnosa</i>		-	Dinan <i>et al.</i> (1998)
<i>M. enchylaenoides</i>		-	Dinan <i>et al.</i> (1998)
<i>M. erioclada</i>		-	Dinan <i>et al.</i> (1998)
<i>M. excavata</i>		-	Dinan <i>et al.</i> (1998)
<i>M. georgei</i>		-	Dinan <i>et al.</i> (1998)
<i>M. platycarpa</i>		-	Dinan <i>et al.</i> (1998)
<i>M. polypterygia</i>		-	Dinan <i>et al.</i> (1998)
<i>M. pyrimidata</i>		-	Dinan <i>et al.</i> (1998)
<i>M. sedifolia</i>		-	Dinan <i>et al.</i> (1998)
<i>M. tomentosa</i>		-	Dinan <i>et al.</i> (1998)
<i>M. triptera</i>		-	Dinan <i>et al.</i> (1998)
<i>Malus domestica</i>	Rosaceae (D)	-	Dinan <i>et al.</i> (2020a)
<i>M. sikkimensis</i>	[Rosaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>M. sylvestris</i>		-	Blackford & Dinan (1997c)
<i>Malva moschata</i>	Malvaceae (D)	(+)	Blackford & Dinan (1997a)
<i>M. officinalis</i>	[Malvaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>M. robusta</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>M. sylvestris</i>		-	Blackford & Dinan (1997a)
<i>Mandragora officinarum</i>	Solanaceae (D)	-	Savchenko <i>et al.</i> (2000)
	[Solanaceae (E)]		
<i>Mangifera indica</i>	Anacardaceae (D)	-	Dinan <i>et al.</i> (2020a)
	[Anacardiaceae (E)]		
<i>Manihot esculentum</i>	Euphorbiaceae (D)	-	Blackford <i>et al.</i> (1996)
	[Euphorbiaceae (E)]		
<i>Manochlamys albicans</i>	Chenopodiaceae (D)	+	Dinan <i>et al.</i> (1998)
	[Amaranthaceae (E)]		
<i>Marattia salicina</i>	Marattiaceae (F)	-	Russell & Fenemore (1971)
	[Marattiaceae (F)]		
<i>Marsilea quadrifolia</i>	Marsileaceae (F)	+	Hikino <i>et al.</i> (1973)
	[Marsilieaceae (F)]		
<i>Matricaria chamomile</i>	Compositae (D)	-	Dinan <i>et al.</i> (2020b)
<i>M. recutita</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Matteuccia orientalis</i>	Woodsiaceae (F)	-	Takemoto <i>et al.</i> (1967c)
	[Onocleaceae (F)]	+	Takemoto <i>et al.</i> (1967a)
		+	Hikino & Hikino (1970)
		+	Hikino <i>et al.</i> (1973)
<i>M. struthiopteris</i>		+	Hikino <i>et al.</i> (1973)
		+	Takemoto <i>et al.</i> (1973)
		+/-	Revina & Gureeva (1985)

		+	Shin & Lee (2011)
		-	Petruk <i>et al.</i> (2013)
<i>Mecodium (Hymenophyllum) flexile</i>	Hymenophyllaceae (F)	-	Hikino <i>et al.</i> (1973)
<i>M. polyanthos</i>	[Hymenophyllaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>M. wrightii</i>		-	Hikino <i>et al.</i> (1973)
<i>Meconopsis superba</i>	Papaveraceae (D) [Papaveraceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Medicago arabica</i>	Leguminosae-P. (D) [Fabaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Melampyrum nemorosum</i>	Scrophulariaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>M. roseum</i>	[Orobanchaceae (E)]	0.7	Wong <i>et al.</i> (1979)
<i>Melandrium (Vaccaria) album</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	-?	Revina <i>et al.</i> (1988)
		-	Volodin <i>et al.</i> (2002)
		+	Zibareva <i>et al.</i> (2007a)
<i>M. apetala</i>		+	Zibareva <i>et al.</i> (2007a)
<i>M. erubescens</i>		+	Abubakirov (1984)
<i>M. firmum</i>		+	Zheng <i>et al.</i> (2008)
		+	Novozhilova <i>et al.</i> (2014)
		+	Novozhilova <i>et al.</i> (2015)
<i>M. noctiflorum</i>		+	Bespayeva <i>et al.</i> (2012)
<i>M. (= Silene) nutans</i>		+	Abubakirov (1984)
		+	Baltaev <i>et al.</i> (1984)
<i>M. ruinarum</i>		+	Abubakirov (1984)
<i>M. sachalinense</i>		+	Novozhilova <i>et al.</i> (2015)
<i>M. tristis</i>		+	Zibareva <i>et al.</i> (2007a)
<i>M. turkestanicum</i>		+	Abubakirov (1984)
		+	Saatov <i>et al.</i> (1990c)
		+	Saatov <i>et al.</i> (1991)
		+	Saatov <i>et al.</i> (1999)
<i>Melica ciliata</i>	Gramineae (M) [Poaceae (M)]	-	Dinan (1995a)
<i>Melothria scabra</i>	Cucurbitaceae (D) [Cucurbitaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Melilotus albus</i>	Leguminosae-P. (D) [Fabaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Menispermum dauricum</i>	Menispermaceae (D) [Menispermaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Mentha arvensis</i>	Labiatae (D)	-	Volodin <i>et al.</i> (2002)
<i>M. piperata</i>	[Lamiaceae (E)]	-	Blackford & Dinan (1997b)
<i>Menyanthes trifoliata</i>	Menyanthaceae (D) [Menyanthaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Mercurialis annua</i>	Euphorbiaceae (D) [Euphorbiaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Metaplexis japonica</i>	Asclepiadaceae (D) [Apocynaceae (E)]	3.4	Wong <i>et al.</i> (1979)
<i>Microlepis hookeriana</i>	Dennstaedtiaceae (F)	+	Yen <i>et al.</i> (1974)

<i>M. marginata</i>	[Dennstaedtiaceae (F)]	-	Hikino <i>et al.</i> (1973)
		+/-	Yen <i>et al.</i> (1974)
<i>M. obtusifolia</i>		+	Hikino <i>et al.</i> (1973)
<i>M. pilosula</i>		-	Yen <i>et al.</i> (1974)
<i>M. speluncae</i>		+	Yen <i>et al.</i> (1974)
<i>M. strigosa</i>		-	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)
<i>M. substrigosa</i>		-	Hikino <i>et al.</i> (1973)
<i>Microsorium buergerianum</i>	Polypodiaceae (F)	+	Hikino <i>et al.</i> (1973)
	[Polypodiaceae (F)]	-	Yen <i>et al.</i> (1974)
<i>M. commutatum</i>		+	Ho <i>et al.</i> (2007)
<i>M. hancockii</i>		+	Hikino <i>et al.</i> (1973)
<i>M. fortunei</i>		-	Yen <i>et al.</i> (1974)
<i>M. insigne</i>		+	Jitchum <i>et al.</i> (2016)
<i>M. maximum</i>		+	Ho <i>et al.</i> (2007)
<i>M. membranaceum</i>		-	Jitchum <i>et al.</i> (2016)
<i>M. membranifolium</i>		-	Yen <i>et al.</i> (1974)
		+	Ho <i>et al.</i> (2007)
		+	Ho <i>et al.</i> (2008)
		+	Ho <i>et al.</i> (2012)
<i>M. punctatum</i>		+	Ho <i>et al.</i> (2007)
		+	Sripinyowanich <i>et al.</i> (2021)
		+	Jitchum <i>et al.</i> (2016)
<i>M. rubidum</i>		-/(+)	Ho <i>et al.</i> (2007)
<i>M. scolopendria</i>		+	Snogan <i>et al.</i> (2007)
		+	Ho <i>et al.</i> (2007)
		+	Sripinyowanich <i>et al.</i> (2021)
<i>M. superficiale</i>		+	Jitchum <i>et al.</i> (2016)
<i>Milium effusum</i>	Gramineae (M)	-	Volodin <i>et al.</i> (2002)
<i>M. effusum aureum</i>	[Poaceae (M)]	-	Dinan (1995a)
<i>Mimulus guttatus</i>	Scrophulariaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
	[Phrymaceae (E)]		
<i>Mimusops elanji</i>	Sapotaceae (D)	-	Sreejit & Nelshi (2019)
	[Sapotaceae (E)]		
<i>Minuartia biflora</i>	Caryophyllaceae (D)	-	Revina <i>et al.</i> (1988)
<i>M. kyriloviana</i>	[Caryophyllaceae (E)]	+	Bespayeve <i>et al.</i> (2012)
<i>M. laricifolia</i>		-	Zibareva <i>et al.</i> (2003)
<i>M. verna</i>		-	Revina <i>et al.</i> (1988)
		-	Zibareva <i>et al.</i> (2003)
<i>Mirabilis dichotoma</i>	Nyctaginaceae (D)	-	Báthori <i>et al.</i> (1987)
<i>M. jalapa</i>	[Nyctaginaceae (E)]	+	Takemoto <i>et al.</i> (1967c)
		-	Báthori <i>et al.</i> (1987)
<i>M. nyctaginea</i>		-	Báthori <i>et al.</i> (1987)
<i>Miscanthus sinensis</i>	Gramineae (M)	-	Dinan (1995a)
	[Poaceae (M)]		
<i>Moehringia lateriflora</i>	Caryophyllaceae (D)	-	Revina <i>et al.</i> (1988)
	[Caryophyllaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>M. umbrosa</i>		-	Revina <i>et al.</i> (1988)
<i>Monachosorum arakii</i>	Adiantaceae (F)	-	Hikino <i>et al.</i> (1973)
<i>M. flagellare</i>	[Dennstaedtiaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>M. maximowiczii</i>		-	Hikino <i>et al.</i> (1973)

<i>Moneses uniflora</i>	Ericaceae (D) [Ericaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Monolepis trifida</i>	Chenopodiaceae (D)	-	Dinan <i>et al.</i> (1998)
<i>M. nutalliana</i>	[Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Morinda citrifolia</i>	Rubiaceae (D) [Rubiaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Moringa pterygosperma</i>	Moringaceae (D) [Moringaceae (E)]	-	Sreejit (2014)
<i>Morus</i> sp.	Moraceae (D)	+	Takemoto <i>et al.</i> (1967e)
<i>M. alba</i>	[Moraceae (E)]	-	Blackford & Dinan (1997b)
		+	Zhou <i>et al.</i> (2005)
		+	Qiu <i>et al.</i> (2008)
<i>M. atropurpurea</i>		+	Ya <i>et al.</i> (2006)
<i>Morus nigra</i>		-	Blackford & Dinan (1997b)
		-	Blackford & Dinan (1997c)
<i>Mosla punctata</i>	Labiatae (D) [Lamiaceae (E)]	1.8	Wong <i>et al.</i> (1979)
<i>Murdannia triquetra</i>	Commelinaceae (M) [Commelinaceae (M)]	+	Hou <i>et al.</i> (1980)
		+	Hou <i>et al.</i> (1981)
		+	Wang <i>et al.</i> (1984)
<i>M. scapiflora</i>		-	Dreier (1987)
<i>Musa x paradisiaca</i>	Musaceae (M) [Musaceae (M)]	-	Dinan <i>et al.</i> (2020a)
<i>Muscari leucostomum</i>	Hyacinthaceae (M) [Asparagaceae (M)]	-	Dinan <i>et al.</i> (2020b)
<i>Myosotis arvensis</i>	Boraginaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>M. cespitosa</i>	[Boraginaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Myosoton aquaticum</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	-	Dinan <i>et al.</i> (2001d)
		-	Zibareva <i>et al.</i> (2003)
<i>Myrciaria cauliflora</i>	Myrtaceae (D) [Myrtaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Myriophyllum spicatum</i>	Haloragaceae (D) [Haloragaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Myristica fragrans</i>	Myristicaceae (D) [Myristicaceae (Mag)]	-	Dinan <i>et al.</i> (2020a)
N			
<i>Napaea dioica</i>	Malvaceae (D) [Malvaceae (E)]	+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
<i>Naumburgia (Lysimachia) thyrsiflora</i>	Primulaceae (D) [Primulaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Nectaroscordum siculum</i> (syn. <i>Allium bulgaricum</i>)	Alliaceae (M) [Amaryllidaceae (M)]	-	Dinan <i>et al.</i> (2020b)

<i>Nelumbo nucifera</i>	Nelumbonaceae (D) [Nelumbonaceae (E)]	-	Sreejit (2014)
<i>Neobassia astrocarpa</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Neocheiropteris ensata</i>	Polypodiaceae (F) [Polypodiaceae (F)]	+ + + +	Takemoto <i>et al.</i> (1968g) Hikino & Hikino (1970) Takemoto <i>et al.</i> (1973) Hikino <i>et al.</i> (1973)
<i>N. normalis</i>		+	Jitchum <i>et al.</i> (2016)
<i>Nepeta camphorata</i>	Labiatae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>N. cataria</i>	[Lamiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>N. italica</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>N. subsessilis</i>		-	Dinan <i>et al.</i> (2001d)
<i>Nephelium lappaceum</i>	Sapindaceae (D) [Sapindaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Nephrolepis auriculata</i>	Oleandraceae (F) [Nephrolepidaceae (F)]	- -	Hikino <i>et al.</i> (1973) Yen <i>et al.</i> (1974)
<i>N. biserrata</i>		+	Yen <i>et al.</i> (1974)
<i>N. hirsutula</i>		-	Yen <i>et al.</i> (1974)
<i>Nephrolepis</i> sp.		+	Saeng-ngam <i>et al.</i> (on-line)
<i>Nerium indicum</i>	Apocynaceae (D)	-	Takemoto <i>et al.</i> (1967c)
<i>N. oleander</i>	[Apocynaceae (E)]	-	Blackford & Dinan (1997c)
<i>Nicandra physaloides</i>	Solanaceae (D) [Solanaceae (E)]	(+)	Savchenko <i>et al.</i> (2000)
<i>Nicotiana affinis</i>	Solanaceae (D)	-(+)	Savchenko <i>et al.</i> (2000)
<i>N. glauca</i>	[Solanaceae (E)]	(+)	Savchenko <i>et al.</i> (2000)
<i>N. alata</i>		-	Savchenko <i>et al.</i> (2000)
<i>N. knightiana</i>		-	Savchenko <i>et al.</i> (2000)
<i>N. langsdorfii</i>		-	Savchenko <i>et al.</i> (2000)
<i>N. rustica</i>		-	Dinan <i>et al.</i> (2020b)
		-	Blackford & Dinan (1997c)
		-(+)	Savchenko <i>et al.</i> (2000)
<i>N. sylvestris</i>		-(+)	Savchenko <i>et al.</i> (2000)
<i>N. tabacum</i>		-	Blackford <i>et al.</i> (1996)
		-	Blackford & Dinan (1997c)
		-	Savchenko <i>et al.</i> (2000)
<i>Niedenzuella multiglandulosa</i>	Malpighiaceae (D) [Malpighiaceae (E)]	+	Russo <i>et al.</i> (2020)
<i>Nierembergia caeru</i>	Solanaceae (D)	+	Savchenko <i>et al.</i> (2000)
<i>N. hippomanica</i>	[Solanaceae (E)]	+	Pomilio <i>et al.</i> (1996)
<i>N. hippomanica</i> var. <i>violacea</i>		+	Savchenko <i>et al.</i> (2000)
<i>N. solanacea</i>		+	Savchenko <i>et al.</i> (2000)
<i>Nigella damascena</i>	Ranunculaceae (D)	-	Dinan <i>et al.</i> (2002a)
<i>N. hispanica</i>	[Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2002a)
<i>N. orientalis</i>		-	Dinan <i>et al.</i> (2002a)
<i>N. sativa</i>		-	Dinan <i>et al.</i> (2002a)
		-	Dinan <i>et al.</i> (2020a)

<i>Nitrophila mohavensis</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Nolana paradoxa</i>	Solanaceae (D) [Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
<i>Nonea pulla</i>	Boraginaceae (D) [Boraginaceae (E)]	- -	Blackford & Dinan (1997a) Volodin <i>et al.</i> (2002)
<i>Notholirion campanulatum</i>	Liliaceae (M) [Liliaceae (M)]	-	Dinan <i>et al.</i> (2001c)
<i>Nuphar lutea</i>	Nymphaeaceae (D) [Nymphaeaceae (Nym)]	-	Volodin <i>et al.</i> (2002)
<i>Nymphaea</i> sp.	Nymphaeaceae (D) [Nymphaeaceae (Nym)]	-	Volodin <i>et al.</i> (2002)
O			
<i>Oberna behen</i> (syn. <i>Silene vulgaris</i>)	Caryophyllaceae (D) [Caryophyllaceae (E)]	- -	Revina <i>et al.</i> (1988) Volodin <i>et al.</i> (2002)
<i>Ochna kirkii</i>	Ochnaceae (D) [Ochnaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Oldenlandia chrysotricha</i>	Rubiaceae (D) [Rubiaceae (E)]	1.0	Wong <i>et al.</i> (1979)
<i>Olearia macrodontata</i>	Compositae (D) [Asteraceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Omalotheca supina</i> <i>O. sylvatica</i>	Compositae (D) [Asteraceae (E)]	- -	Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002)
<i>Onoclea sensibilis</i>	Woodsiaceae (F) [Onocleaceae (F)]	+ + + + + +	Takemoto <i>et al.</i> (1967a) Kaplanis <i>et al.</i> (1967) Takemoto <i>et al.</i> (1968b) Hikino & Hikino (1970) Takemoto <i>et al.</i> (1973) Hikino <i>et al.</i> (1973)
<i>Onopordum acanthium</i>	Compositae (D) [Asteraceae (E)]	+ -	Volodin <i>et al.</i> (1993) Blackford & Dinan (1997a)
<i>Onychium japonicum</i>	Adiantaceae (F) [Pteridaceae (F)]	+ - -	Takemoto <i>et al.</i> (1967c) Yen <i>et al.</i> (1974) Hikino <i>et al.</i> (1973)
<i>Ophioglossum vulgatum</i>	Ophioglossaceae (F) [Ophioglossaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>Opuntia ficus-indica</i>	Cactaceae (D) [Cactaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Origanum vulgare</i>	Labiatae (D) [Lamiaceae (E)]	3.0 -	Wong <i>et al.</i> (1979) Volodin <i>et al.</i> (2002)
<i>Orlaya grandiflora</i>	Umbelliferae (D) [Apiaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)

<i>Orthilia secunda</i>	Ericaceae (D) [Ericaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Oryza sativa</i>	Gramineae (M) [Poaceae (M)]	- (+) -	Takemoto <i>et al.</i> (1967c) Blackford <i>et al.</i> (1996) Dinan <i>et al.</i> (2020a)
<i>Osmunda asiatica</i>	Osmundaceae (F) [Osmundaceae (F)]	+ +	Takemoto <i>et al.</i> (1968c) Takemoto <i>et al.</i> (1973)
<i>O. banksiaefolia</i>		+	Hikino <i>et al.</i> (1973)
<i>O. cinnamomea</i>		+	Hikino <i>et al.</i> (1973)
<i>O. claytoniana</i>		+	Kaplanis <i>et al.</i> (1967)
<i>O. japonica</i>		+	Hikino <i>et al.</i> (1973)
		+	Takemoto <i>et al.</i> (1968c)
		+	Takemoto <i>et al.</i> (1967c)
		+	Imai <i>et al.</i> (1969c)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
		+	Takemoto <i>et al.</i> (1973)
		+	Hikino <i>et al.</i> (1973)
		+	Shin & Lee (2011)
<i>O. lancea</i>		+	Hikino <i>et al.</i> (1973)
<i>O. lancea</i> var. <i>latipinnula</i>		+	Hikino <i>et al.</i> (1973)
<i>Osteocarpum dipterocarpum</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Otites parviflorus</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	+	Darmogray <i>et al.</i> (2015)
<i>Ourisia caespitosa</i>	Scrophulariaceae (D)	+	Sarker <i>et al.</i> (1996c)
<i>O. macrocarpa</i>	[Plantaginaceae (E)]	+	Sarker <i>et al.</i> (1996c)
<i>O. macrophylla</i>		+	Sarker <i>et al.</i> (1996c)
		+	Dinan <i>et al.</i> (2020b)
<i>O. sessilifolia</i>		-	Sarker <i>et al.</i> (1996c)
<i>Oxalis acetosella</i>	Oxalidaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>O. corymbosa</i>	[Oxalidaceae (E)]	+	Volodin <i>et al.</i> (2018)
<i>Oxycoccus microcarpus</i> (syn. <i>Vaccinium microcarpum</i>)	Ericaceae (D) [Ericaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Oxytropis vicida</i>	Leguminosae-P. (D) [Fabaceae (E)]	-	Dinan <i>et al.</i> (2020b)
P			
<i>Pachypleurum alpinum</i>	Umbelliferae (D) [Apiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Padus avium</i>	Rosaceae (D) [Rosaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Paederia scandens</i>	Rubiaceae (D) [Rubiaceae (E)]	0	Wong <i>et al.</i> (1979)
<i>Paeonia anomala</i>	Paeoniaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>P. delavayi</i>	[Paeoniaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Paesia scaberula</i>	Dennstaedtiaceae (F) [Dennstaedtiaceae (F)]	-	Russell & Fenemore (1971)

<i>Palbergia (Dalbergia?) hupeana</i>	Leguminosae (D) [Fabaceae (E)]	+	Chou & Lu (1980)
<i>Palisota ambigua</i>	Commelinaceae (M)	+	Kusamba <i>et al.</i> (1995)
<i>P. barteri</i>	[Commelinaceae (M)]	+	Crouzet <i>et al.</i> (2009)
<i>P. hirsuta</i>		+	Kusamba <i>et al.</i> (1995)
		+	Sarpong (2012)
		+	Sarpong <i>et al.</i> (2016)
<i>P. mannii</i>		+	Crouzet <i>et al.</i> (2009)
<i>P. pynaerti</i>		+	Crouzet <i>et al.</i> (2009)
<i>P. schweinfurthii</i>		+	Kusamba <i>et al.</i> (1995)
<i>Papaver argemone</i>	Papaveraceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>P. dubium</i>	[Papaveraceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Papaver</i> sp.		-	Dinan <i>et al.</i> (2020a)
<i>P. nudicaule</i>		-	Dinan <i>et al.</i> (2020b)
<i>P. sendtneri</i>		-	Dinan <i>et al.</i> (2020b)
<i>Panderia pilosa</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Pandiaka involucrata</i>	Amaranthaceae (D) [Amaranthaceae (E)]	+	Harman & Mahar (1978)
<i>Panicum miliaceum</i>	Gramineae (M) [Poaceae (M)]	-	Dinan (1995a)
		-	Blackford <i>et al.</i> (1996)
<i>P. violaceum</i>		-	Dinan (1995a)
<i>Paradisea lusitanica</i>	Asphodelaceae (M) [Asparagaceae (M)]	-	Dinan <i>et al.</i> (2020b)
<i>Parietaria diffusa</i>	Urticaceae (D) [Urticaceae (E)]	-	Blackford & Dinan (1997a)
		-	Blackford & Dinan (1997c)
<i>Paris axialis</i>	Trilliaceae (M)	+	Huang <i>et al.</i> (2010)
<i>P. bashanensis</i>	[Melanthiaceae (M)]	+	Zhang <i>et al.</i> (2011)
<i>P. delavayi</i>		+	Liu <i>et al.</i> (2006)
<i>P. dulongensis</i>		+	Wang <i>et al.</i> (2017)
<i>P. formosana</i>		+	Yeh & Chiang (1982)
		+	Chiang <i>et al.</i> (1992)
<i>P. polyphylla</i>		+	Singh & Thakur (1982)
		+	Zhang <i>et al.</i> (2017)
<i>P. pollyphylla</i> var. <i>chinensis</i>		+	Yin <i>et al.</i> (2015)
		+	Zhao <i>et al.</i> (2018)
<i>P. polyphylla</i> var. <i>pseudotibetica</i>		+	Xiao <i>et al.</i> (2009)
		+	Zhao <i>et al.</i> (2011)
<i>P. polyphylla</i> var. <i>yunnanensis</i>		+	Zhang <i>et al.</i> (2014)
		+	Guo <i>et al.</i> (2021)
<i>P. pubescens</i>		+	Huang <i>et al.</i> (2009)
<i>P. quadrifolia</i>		+	Abubakirov (1982)
		+	Novoselskya <i>et al.</i> (1981a)
		+	Nohara <i>et al.</i> (1982)
		+	Abubakirov (1984)
		+	Volodin <i>et al.</i> (2002)
		+	Jenett-Siems <i>et al.</i> (2012)
		+	Dinan <i>et al.</i> (2020b)
<i>P. tetraphylla</i>		+	Imai <i>et al.</i> (1969c)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)

<i>P. verticillata</i>		+	Nakano <i>et al.</i> (1981)
		+	Huang <i>et al.</i> (2009)
		+	Jiang <i>et al.</i> (2021)
<i>Paris spp.</i>		+	Wang <i>et al.</i> (2017)
<i>Parnassia palustris</i>	Parnassiaceae (D) [Celastraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Passiflora edulis</i>	Passifloraceae (D)	-	Dinan <i>et al.</i> (2020a)
<i>P. quadrangularis</i>	[Passifloraceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Pastinaca sativa</i>	Umbelliferae (D) [Apiaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Paulownia fortunei</i>	Scrophulariaceae (D) [Paulowniaceae (E)]	0.7	Wong <i>et al.</i> (1979)
<i>Pedicularis lapponica</i>	Scrophulariaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>P. sceptrum-carolinum</i>	[Orobanchaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Pedicularis sp.</i>		-	Volodin <i>et al.</i> (2002)
<i>Pelargonium peltatum</i>	Geraniaceae (D) [Geranaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Pellaea falcata</i>	Adiantaceae (F)	+	Bergamasco & Horn (1983)
<i>P. rotundifolia</i>	[Pteridaceae (F)]	+	Russell & Fenemore (1971)
<i>Peltaria alliacea</i>	Cruciferae (D) [Brassicaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Penianthus longifolius</i>	Menispermaceae (D)	+	Tabekoueng <i>et al.</i> (2019)
<i>P. zenkeri</i>	[Menispermaceae (E)]	+	Tabekoueng <i>et al.</i> (2020)
<i>Peniocereus greggii</i>	Cactaceae (D) [Cactaceae (E)]	+	Knight & Pettit (1969)
<i>Pennisetum ruppelianum</i>	Gramineae (M) [Poaceae (M)]	-	Dinan (1995a)
<i>Penstemon alpinus</i>	Scrophulariaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>P. azureus</i>	[Plantaginaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>P. laevigatus</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>P. lyallii</i>		-	Dinan <i>et al.</i> (2020b)
<i>P. procerus</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>P. serrulatus</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>P. venustus</i>		+	Roth <i>et al.</i> (1995)
<i>Peperomia pellucida</i>	Piperaceae (D) [Piperaceae (Mag)]	-	Sreejit (2014)
<i>Perilla frutescens</i> var. <i>crispa</i>	Labiatae (D) [Lamiaceae (E)]	1.0	Wong <i>et al.</i> (1979)
<i>Persea americana</i>	Lauraceae (D) [Lauraceae (Mag)]	-	Dinan <i>et al.</i> (2020a)
<i>Persicaria amphibia</i>	Polygonaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>P. hydropiper</i>	[Polygonaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Petasites radiatus</i>	Compositae (D)	-	Volodin <i>et al.</i> (2002)

<i>P. spurius</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Petrocoptis glaucifolia</i>	Caryophyllaceae (D)	+	Zibareva <i>et al.</i> (2003)
	[Caryophyllaceae (E)]	+	Zibareva <i>et al.</i> (2007a)
<i>P. hispanica</i>		+	Zibareva <i>et al.</i> (2003)
<i>P. pyrenaica</i>		+	Zibareva <i>et al.</i> (2003)
		+	Zibareva <i>et al.</i> (2007a)
<i>Petrorhagia prolifera</i>	Caryophyllaceae (D)	-	Zibareva <i>et al.</i> (2003)
<i>P. saxifraga</i>	[Caryophyllaceae (E)]	-	Zibareva <i>et al.</i> (2003)
<i>Petunia integrifolia</i>	Solanaceae (D)	-	Savchenko <i>et al.</i> (2000)
<i>P. pendula</i>	[Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
<i>Pfaffia glomerata</i>	Amaranthaceae (D)	+	Shiobara <i>et al.</i> (1993)
	[Amaranthaceae (E)]	+	Gosmann <i>et al.</i> (2003)
		+	Zimmer <i>et al.</i> (2006)
		+	Flores <i>et al.</i> (2009)
		+	Nakamura <i>et al.</i> (2010)
		+	Flores <i>et al.</i> (2010)
		+	Serra <i>et al.</i> (2012)
		+	Rostagno <i>et al.</i> (2014)
		+	Vasconcelos <i>et al.</i> (2014)
		+	Debien & Meireles (2014)
		+	Felipe <i>et al.</i> (2014)
		+	Flores <i>et al.</i> (2015)
		+	Correa <i>et al.</i> (2016)
		+	Vardanega <i>et al.</i> (2017)
		+	Vardanega <i>et al.</i> (2019)
		+	Felipe <i>et al.</i> (2019)
		+	Ferreira <i>et al.</i> (2019)
		+	Silva <i>et al.</i> (2019)
		+	Zhao <i>et al.</i> (2019)
		+	Silva <i>et al.</i> (2020a)
		+	Silva <i>et al.</i> (2020b)
		+	da Silva Souza <i>et al.</i> (2021)
		+	Oliveira da Silva <i>et al.</i> (2021)
<i>P. iresinoides</i>		+	Nishimoto <i>et al.</i> (1987)
		+	Nishimoto <i>et al.</i> (1988)
<i>P. paniculata</i>		-	Gosmann <i>et al.</i> (2003)
		+	Li <i>et al.</i> (2009)
		+	Gao <i>et al.</i> (2009)
<i>P. pulverulenta</i>		+	Shiobara <i>et al.</i> (1992)
<i>P. tuberosa</i>		+	Nishimoto <i>et al.</i> (1986)
		+	Flores <i>et al.</i> (2009)
		+	Flores <i>et al.</i> (2010)
<i>Phalaris canariensis</i>	Gramineae (M)	-	Dinan (1995a)
	[Poaceae (M)]		
<i>Phalaroides arundinacea</i>	Gramineae (M)	+	Volodin <i>et al.</i> (2002)
	[Poaceae (M)]		
<i>Phanerophlebia falcata</i>	Dryopteridaceae (F)	+/-	Hikino <i>et al.</i> (1973)
<i>P. fortunei</i>	[Dryopteridaceae (F)]	-	Takemoto <i>et al.</i> (1967c)
		+	Matsuoka <i>et al.</i> (1969)
		+/-	Hikino <i>et al.</i> (1973)
<i>P. fortunei</i> var. <i>clivicola</i>		+	Hikino <i>et al.</i> (1973)
<i>P. fortunei</i> var. <i>intermedia</i>		-	Hikino <i>et al.</i> (1973)

<i>P. macrophylla</i>		+/-	Hikino <i>et al.</i> (1973)
<i>P. tachiroana</i>		+	Hikino <i>et al.</i> (1973)
<i>P. vittata</i>		+	Hikino <i>et al.</i> (1973)
<i>Phaseolus aureus</i>	Leguminosae-P. (D)	(+)	Blackford <i>et al.</i> (1996)
<i>P. coccineus</i>	[Fabaceae (E)]	-	Blackford <i>et al.</i> (1996)
		-	Blackford & Dinan (1997a)
<i>P. vulgaris</i>		-	Dinan <i>et al.</i> (2020a)
<i>Phegopteris (Lastrea) decursive-pinnata</i>	Thelypteridaceae (F)	-	Yen <i>et al.</i> (1974)
<i>P. subaurata</i>	[Thelypteridaceae (F)]	+	Yen <i>et al.</i> (1974)
<i>P. subaurita</i>		-	Hikino <i>et al.</i> (1973)
<i>Philadelphus coronarius</i>	Hydrangeaceae (D)	-	Blackford & Dinan (1997c)
<i>P. x virginalis</i>	[Hydrangeaceae (E)]	-	Blackford & Dinan (1997c)
<i>Phleum alpinum</i>	Gramineae (M)	-	Volodin <i>et al.</i> (2002)
<i>P. pratense</i>	[Poaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Phlomis umbrosa</i> var. <i>ovalifolia</i>	Labiatae (D)	3.4	Wong <i>et al.</i> (1979)
	[Lamiaceae (E)]		
<i>Phlox drummondii</i>	Polemoniaceae (D)	+	Matsuoka <i>et al.</i> (1969)
	[Polemoniaceae (E)]		
<i>Phoenix dactylifera</i>	Palmae (M)	-	Dinan <i>et al.</i> (2020b)
	[Arecaceae (M)]		
<i>Phyllanthus acidus</i>	Euphorbiaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>P. amarus</i>	[Phyllanthaceae (E)]	-	Sreejit & Nelshi (2019)
<i>Phyllanthus</i> sp.		+	Saeng-ngam <i>et al.</i> (on-line)
<i>Phyllocladus alpinus</i>	Phyllocladaceae (G)	+	Russell & Fenemore (1970)
<i>P. aspleniifolius</i>	[Podocarpaceae (G)]	+	Gerard <i>et al.</i> (1997)
<i>P. trichomanoides</i>		+	Russell & Fenemore (1970)
		+	Gerard <i>et al.</i> (1997)
<i>Phymatodes (Dendroconche, Microsorium) scandens</i>	Dipteridaceae (F)	-	Russell & Fenemore (1971)
<i>P. diversifolium</i>	[Polypodiaceae (F)]	-	Russell & Fenemore (1971)
<i>P. novae-zelandiae</i>		+	Russell & Fenemore (1971)
		+	Russell (1972)
<i>Phymatosorus (Microsorium) cuspidatus</i>	Polypodiaceae (F)	+	Jitchum <i>et al.</i> (2016)
<i>P. scolopendria</i>	[Polypodiaceae (F)]	+	Jitchum <i>et al.</i> (2016)
<i>Physalis alkekengi</i>	Solanaceae (D)	(+)	Blackford & Dinan (1997c)
	[Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
<i>P. franchetii</i>		-	Savchenko <i>et al.</i> (2000)
<i>P. gigantea</i>		-/(+)	Savchenko <i>et al.</i> (2000)
<i>P. ixocarpa</i>		-/(+)	Savchenko <i>et al.</i> (2000)
		-	Dinan <i>et al.</i> (2020b)
<i>P. minima</i>		-/(+)	Savchenko <i>et al.</i> (2000)
<i>P. nicandroides</i>		-	Savchenko <i>et al.</i> (2000)
<i>P. peruviana</i>		-	Savchenko <i>et al.</i> (2000)
<i>P. philadelphica</i>		-	Savchenko <i>et al.</i> (2000)
<i>P. pruinosa</i>		-/(+)	Savchenko <i>et al.</i> (2000)
<i>P. pubescens</i>		-	Savchenko <i>et al.</i> (2000)
<i>P. subglabrata</i>		-	Savchenko <i>et al.</i> (2000)
<i>P. viscosa</i>		-	Savchenko <i>et al.</i> (2000)

<i>Phytolacca acinosa</i>	Phytolaccaceae (D)	1.1	Wong <i>et al.</i> (1979)
<i>P. americana</i>	[Phytolaccaceae (E)]	-	Takemoto <i>et al.</i> (1967c)
		-	Dinan <i>et al.</i> (2020b)
<i>P. clavigera</i>		-	Báthori <i>et al.</i> (1987)
<i>Picea abies</i>	Pinaceae (G)	-	Hoffmeister <i>et al.</i> (1967)
<i>P. obovata</i>	[Pinaceae (G)]	-	Volodin <i>et al.</i> (2002)
<i>Picris hieracioides</i>	Compositae (D)	-	Volodin <i>et al.</i> (2002)
<i>P. umbellatum</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Pilea microphylla</i>	Urticaceae (D)	-	Sreejit (2014)
	[Urticaceae (E)]		
<i>Pimpinella anisum</i>	Umbelliferae (D)	-	Dinan <i>et al.</i> (2020a)
<i>P. saxifraga</i>	[Apiaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Pinguicula</i> sp.	Lentibulariaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Lentibulariaceae (E)]		
<i>Pinus pinaster</i>	Pinaceae (G)	-	Clément & Dinan (1991)
<i>P. pinea</i>	[Pinaceae (G)]	-	Dinan <i>et al.</i> (2020a)
<i>P. sibirica</i>		-	Volodin <i>et al.</i> (2002)
<i>P. silvestris</i>		-	Hoffmeister <i>et al.</i> (1967)
<i>P. sylvestris</i>		-	Volodin <i>et al.</i> (2002)
<i>P. thunbergia</i>		0.7	Wong <i>et al.</i> (1979)
<i>Pistacia integrima</i>	Anacardiaceae (D)	-	Sreejit (2014)
	[Anacardiaceae (E)]		
<i>Pittosporum tenuifolium</i>	Pittosporaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
	[Pittosporaceae (E)]		
<i>Pityrogramma calomelanos</i>	Adiantaceae (F)	-	Yen <i>et al.</i> (1974)
<i>P. triangularis</i>	[Pteridaceae (F)]	-	Dreier (1987)
<i>Plagiogyria aduata</i>	Plagiogyriaceae (F)	-	Hikino <i>et al.</i> (1973)
<i>P. euphlebia</i>	[Plagiogyriaceae (F)]	+	Hikino <i>et al.</i> (1973)
<i>P. japonica</i>		+	Hikino <i>et al.</i> (1973)
		+	Takemoto <i>et al.</i> (1967c)
<i>P. matsumureana</i>		+/-	Hikino <i>et al.</i> (1973)
<i>P. stenoptera</i>		-	Hikino <i>et al.</i> (1973)
<i>Plantago coronopus</i>	Plantaginaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>P. major</i>	[Plantaginaceae (E)]	-	Blackford & Dinan (1997b)
<i>P. media</i>		-	Volodin <i>et al.</i> (2002)
<i>Platanus acerifolia</i>	Platanaceae (D)	1.3	Wong <i>et al.</i> (1979)
	[Platanaceae (E)]		
<i>Plectranthus amboinicus</i>	Labiatae (D)	-	Sreejit & Nelshi (2019)
<i>P. glaucocalyx</i>	[Lamiaceae (E)]	5.0	Wong <i>et al.</i> (1979)
<i>P. kameba</i>		-	Takemoto <i>et al.</i> (1967c)
<i>P. tomentosus</i>		-	Dinan <i>et al.</i> (2001d)
<i>Plagiogyria formosana</i>	Plagiogyriaceae (F)	+	Yen <i>et al.</i> (1974)
	[Plagiogyriaceae (F)]		
<i>Platanthera bifolia</i>	Orchidaceae (M)	+	Volodin <i>et al.</i> (2002)
	[Orchidaceae (M)]		

<i>Plenasium (Osmunda) banksiaefolium</i>	Osmundaceae (F) [Osmundaceae (F)]	+ +	Yen <i>et al.</i> (1974) Murakami <i>et al.</i> (1980)
<i>Pleopeltis annuifrons</i>	Polypodiaceae (F)	+	Hikino <i>et al.</i> (1973)
<i>P. onoei</i>	[Polypodiaceae (F)]	+	Hikino <i>et al.</i> (1973)
<i>P. thunbergiana</i>		+	Hikino <i>et al.</i> (1973)
		+	Takemoto <i>et al.</i> (1968g)
		+	Takemoto <i>et al.</i> (1967c)
		+	Matsuoka <i>et al.</i> (1969)
		+	Takemoto <i>et al.</i> (1973)
<i>P. ussuriensis</i> var. <i>distans</i>		+	Hikino <i>et al.</i> (1973)
<i>Poa alpina</i> var. <i>nodosa</i>	Gramineae (M)	-	Dinan (1995a)
<i>P. flexuosa</i>	[Poaceae (M)]	-	Dinan <i>et al.</i> (2001d)
<i>P. pratensis</i>		-	Dinan (1995a)
<i>Podocarpus acutifolius</i>	Podocarpaceae (G)	-	Russell & Fenemore (1970)
<i>P. amarus</i>	[Podocarpaceae (G)]	+	Bergamasco & Horn (1983)
<i>P. andina</i>		+	Galbraith & Horn (1969)
		+	Poyser <i>et al.</i> (1973)
		+	Bhakuni <i>et al.</i> (1974)
		+	Silva & Bittner (1986)
		+	Bittner & Silva (1992)
<i>P. andinus</i>		+	Staal (1967)
<i>P. blumei</i>		-	Galbraith & Horn (1969)
<i>P. compactus</i>		+	Bergamasco & Horn (1983)
<i>P. chinensis</i>		+	Imai <i>et al.</i> (1967)
		+	Takemoto <i>et al.</i> (1967d)
		+	Hoffmeister <i>et al.</i> (1967)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
<i>P. dacrydioides</i>		+	Russell & Fenemore (1970)
		+	Galbraith & Horn (1969)
<i>P. dispermus</i>		+	Bergamasco & Horn (1983)
<i>P. drouyanus</i> (<i>P. gracilor</i>)		+	Galbraith & Horn (1969)
<i>P. elatus</i>		+	Galbraith & Horn (1966)
		+	Galbraith & Horn (1968)
		+	Galbraith <i>et al.</i> (1969)
		+	Staal (1967)
		+	Sauer <i>et al.</i> (1968)
		+	Heftmann <i>et al.</i> (1968)
		+	Galbraith & Horn (1969)
		+	Galbraith <i>et al.</i> (1973)
		+	Bergamasco & Horn (1983)
		+	Joly <i>et al.</i> (1969)
<i>P. elongata</i>		+	Galbraith & Horn (1969)
<i>P. hallii</i>		-	Russell & Fenemore (1970)
<i>P. imbricatus</i>		+	Gu <i>et al.</i> (1997)
<i>P. falcatus</i>		-	Staal (1967)
		+	Galbraith & Horn (1969)
		+	Addo <i>et al.</i> (2015)
<i>P. ferrugineus</i>		-	Russell & Fenemore (1970)
		+	Galbraith & Horn (1969)
<i>P. gracilior</i>		+	Sihra (1974)
<i>P. latifolius</i>		-	Galbraith & Horn (1969)
<i>P. macrophyllus</i>		+	Hikino <i>et al.</i> (1970)
		+	Staal (1967)
		+	Takemoto <i>et al.</i> (1967d)
		+	Hoffmeister <i>et al.</i> (1967)

			+	Imai <i>et al.</i> (1968a)
			+	Imai <i>et al.</i> (1968b)
			+	Hikino <i>et al.</i> (1969b)
			+	Imai <i>et al.</i> (1967)
			+	Imai <i>et al.</i> (1969d)
			+	Takemoto <i>et al.</i> (1967c)
			+	Matsuoka <i>et al.</i> (1969)
<i>P. nagi</i>			-	Imai <i>et al.</i> (1967)
			+	Takemoto <i>et al.</i> (1967c)
			-	Staal (1967)
<i>P. nakaii</i>			+	Nakanishi <i>et al.</i> (1966)
			+	Takemoto <i>et al.</i> (1967c)
			+	Nakanishi <i>et al.</i> (1968)
			+	Moriyama & Nakanishi (1968)
			+	Nakanishi (1969)
			+	Schooley <i>et al.</i> (1972)
<i>P. neriiifolius</i>			+	Staal (1967)
			+	Hoffmeister <i>et al.</i> (1967)
			+	Galbraith & Horn (1969)
			+	Bergamasco & Horn (1983)
<i>P. nivalis</i>			+	Russell & Fenemore (1970)
			+	Staal (1967)
			-	Galbraith & Horn (1969)
<i>P. nubigena</i>			+	Takemoto <i>et al.</i> (1967d)
			+	Hoffmeister <i>et al.</i> (1967)
			-	Bhakuni <i>et al.</i> (1974)
			-	Bittner & Silva (1992)
<i>P. rospigliosii</i>			+	Staal (1967)
<i>P. saligna</i>			-	Bhakuni <i>et al.</i> (1974)
.			-	Bittner & Silva (1992)
<i>P. sellowii</i>			+	Sanchez <i>et al.</i> (1970)
<i>P. spicatus</i>			+	Russell & Fenemore (1970)
<i>P. spinulosus</i>			-	Staal (1967)
			+	Bergamasco & Horn (1983)
<i>P. spinulpa</i>			+	Takemoto <i>et al.</i> (1967d)
			+	Hoffmeister <i>et al.</i> (1967)
<i>P. totara</i>			-	Russell & Fenemore (1970)
			-	Staal (1967)
			+	Galbraith & Horn (1969)
<i>Pogonotherum paniceum</i>	Gramineae (M) [Poaceae (M)]		+	Ma <i>et al.</i> (2018)
<i>Polemonium boreale</i>	Polemoniaceae (D)	(+)		Volodin <i>et al.</i> (2002)
<i>P. caeruleum</i>	[Polemoniaceae (E)]	-		Volodin <i>et al.</i> (2002)
<i>Polemonium</i> sp.		-		Volodin <i>et al.</i> (2002)
<i>Pollia japonica</i>	Commelinaceae (M) [Comelinaceae (M)]	(+)		Crouzet <i>et al.</i> (2009)
<i>Polycnemum arvense</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-		Dinan <i>et al.</i> (1998)
<i>Polygala amarella</i>	Polygalaceae (D) [Polygalaceae (E)]	-		Volodin <i>et al.</i> (2002)
<i>Polygonum</i> sp.	Polygonaceae (D)	2.2		Wong <i>et al.</i> (1979)
<i>P. aviculare</i>	[Polygonaceae (E)]	(+)		Blackford & Dinan (1997b)
		-		Volodin <i>et al.</i> (2002)
<i>P. longisetum</i>		-		Takemoto <i>et al.</i> (1967c)

<i>Polypodium aureum</i>	Polypodiaceae (F)	+	Jizba <i>et al.</i> (1974a)
	[Polypodiaceae (F)]	+	Jizba <i>et al.</i> (1974b)
<i>P. fauriei</i>		+	Hikino <i>et al.</i> (1973)
<i>P. formosanum</i>		+	Hikino <i>et al.</i> (1973)
<i>P. glycyrrhiza</i>		+	Dreier (1987)
<i>P. japonicum</i>		+	Imai <i>et al.</i> (1969c)
		+	Imai <i>et al.</i> (1969d)
		+	Matsuoka <i>et al.</i> (1969)
<i>P. niponicum</i>		+/-	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)
<i>P. taiwanianum</i>		+	Yen <i>et al.</i> (1974)
<i>P. virginianum</i>		+	Kaplanis <i>et al.</i> (1967)
		+	Hikino (1976)
<i>P. vulgare</i>		+	Camps <i>et al.</i> (1990)
		+	Jizba <i>et al.</i> (1967b)
		+	de Souza <i>et al.</i> (1970)
		+	Sardini & Krepinsky (1974)
		+	Coll <i>et al.</i> (1994)
		+	Camps <i>et al.</i> (1990)
		+	Jizba <i>et al.</i> (1967)
		+	Heinrich & Hoffmeister (1967)
		+	Jizba <i>et al.</i> (1971)
		+	Heinrich & Hoffmeister (1968)
		+	Marco <i>et al.</i> (1993)
		+	Cook <i>et al.</i> (1973)
		+	Davies <i>et al.</i> (1980)
		+	Camps <i>et al.</i> (1990b)
		+	Hikino <i>et al.</i> (1973)
		+	Reixach <i>et al.</i> (1996)
		+	Reixach <i>et al.</i> (1997)
		+	Messeguer <i>et al.</i> (1998)
		+	Reixach <i>et al.</i> (1999)
		+	Simon <i>et al.</i> (2011)
		+	Vanyolos (2012)
		+	Olennikov & Kashchenko (2018)
<i>Polypogon monspeliensis</i>	Gramineae (M)	-	Dinan (1995a)
	[Poaceae (M)]		
<i>Polystichopsis (Rumohra) amabilis</i>	Dryopteridaceae (F)	-	Yen <i>et al.</i> (1974)
<i>P. quadripinnata</i>	[Dryopteridaceae (F)]	-	Yen <i>et al.</i> (1974)
<i>Polystichum acrostichoides</i>	Dryopteridaceae (F)	+	Kaplanis <i>et al.</i> (1967)
<i>P. craspedosorum</i>	[Dryopteridaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>P. deltodon</i>		+	Hikino <i>et al.</i> (1973)
<i>P. eximium</i> var. <i>minus</i>		+	Hikino <i>et al.</i> (1973)
<i>P. hancockii</i>		+	Hikino <i>et al.</i> (1973)
<i>P. makinoi</i>		+	Hikino <i>et al.</i> (1973)
<i>P. microchlamys</i>		+	Hikino <i>et al.</i> (1973)
<i>P. munitum</i>		-	Dreier (1987)
<i>P. nepalense</i>		+	Yen <i>et al.</i> (1974)
<i>P. otomasui</i>		+	Hikino <i>et al.</i> (1973)
<i>P. parvipinnulum</i>		+	Yen <i>et al.</i> (1974)
<i>P. polyblepharum</i>		+	Imai <i>et al.</i> (1969d)
		+	Takemoto <i>et al.</i> (1967c)
		+	Matsuoka <i>et al.</i> (1969)
		+	Hikino <i>et al.</i> (1973)
<i>P. pseudo-makinoi</i>		+	Hikino <i>et al.</i> (1973)

<i>P. pseudo-makinoi</i> var. <i>ambiguum</i>		-	Hikino <i>et al.</i> (1973)
<i>P. retroso-paleaceum</i>		+	Hikino <i>et al.</i> (1973)
<i>P. retroso-paleaceum</i> var. <i>coraiense</i>		-	Hikino <i>et al.</i> (1973)
<i>P. retroso-paleaceum</i> var. <i>coreiense</i>		-	Takemoto <i>et al.</i> (1967c)
<i>P. retroso-paleaceum</i> var. <i>ovata-paleaceum</i>		-	Hikino <i>et al.</i> (1973)
<i>P. richardii</i>		-	Russell & Fenemore (1971)
<i>P. rigens</i>		-	Hikino <i>et al.</i> (1973)
<i>P. sylvaticum</i>		-	Russell & Fenemore (1971)
<i>P. tripteron</i>		+	Imai <i>et al.</i> (1969d)
		+	Takemoto <i>et al.</i> (1967c)
		-	Hikino <i>et al.</i> (1973)
		+	Matsuoka <i>et al.</i> (1969)
<i>P. tsussimense</i>		-	Hikino <i>et al.</i> (1973)
<i>P. vestitium</i>		+	Russell & Fenemore (1971)
<i>Porana discifera</i>	Convolvulaceae (D) [Convolvulaceae (E)]	+	Zhu <i>et al.</i> (2000)
<i>Portulaca grandiflora</i>	Portulacaceae (D)	-	Báthori <i>et al.</i> (1987)
<i>P. oleracea</i>	[Portulacaceae (E)]	-	Báthori <i>et al.</i> (1987)
		+	Saeng-ngam <i>et al.</i> (on-line)
		+	Daniel & Mammen (2014)
<i>Potamogeton alpinus</i>	Potamogetonaceae (M)	(+)	Chadin <i>et al.</i> (2003)
	[Potamogetonaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>P. berchtoldii</i>		(+)	Chadin <i>et al.</i> (2003)
		-	Volodin <i>et al.</i> (2002)
<i>P. compressus</i>		(+)	Chadin <i>et al.</i> (2003)
		-	Volodin <i>et al.</i> (2002)
<i>P. gramineus</i>		(+)	Chadin <i>et al.</i> (2003)
		(+)	Volodin <i>et al.</i> (2002)
<i>P. lucens</i>		(+)	Chadin <i>et al.</i> (2003)
		-	Volodin <i>et al.</i> (2002)
<i>P. natans</i>		(+)	Chadin <i>et al.</i> (2003)
		+	Volodin <i>et al.</i> (2002)
<i>P. pectinatus</i>		(+)	Chadin <i>et al.</i> (2003)
		(+)	Volodin <i>et al.</i> (2002)
<i>P. perfoliatus</i>		(+)	Chadin <i>et al.</i> (2003)
		+	Volodin <i>et al.</i> (2002)
<i>Potentilla glandulosa</i>	Rosaceae (D)	-	Dinan <i>et al.</i> (2001d)
<i>P. norvegica</i>	[Rosaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>P. pectinata</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>P. rupestris</i>		-	Dinan <i>et al.</i> (2001d)
<i>Premna serratifolia</i>	Verbenaceae (D) [Lamiaceae (E)]	+	Zhu <i>et al.</i> (2000)
<i>Primula capitata</i>	Primulaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>P. farinosa</i>	[Primulaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>P. heucherifolia</i>		-	Dinan <i>et al.</i> (2020b)
<i>P. pulverulenta</i>		-	Dinan <i>et al.</i> (2020b)
<i>P. sikkimensis pudibunda</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>P. sino-purpurea</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>P. veris</i>		-	Volodin <i>et al.</i> (2002)
<i>P. vialii</i>		-	Dinan <i>et al.</i> (2020b)
<i>P. wilsonii</i>		-	Dinan <i>et al.</i> (2001d)
<i>Proboscidea louisianica</i>	Pedaliaceae (D) [Martyniaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)

<i>Prostanthera cuneata</i>	Labiatae (D) [Lamiaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Protea coronata</i>	Proteaceae (D) [Proteaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Prumnopitys andina</i>	Podocarpaceae (G) [Podocarpaceae (G)]	+	Jiminez-Aspee <i>et al.</i> (2019)
<i>Prunella vulgaris</i>	Labiatae (D) [Lamiaceae (E)]	3.5 - -	Wong <i>et al.</i> (1979) Blackford & Dinan (1997b) Volodin <i>et al.</i> (2002)
<i>Prunus</i> sp.	Rosaceae (D)	-	Takemoto <i>et al.</i> (1967c)
<i>P. armeniaca</i>	[Rosaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>P. avium</i>		-	Dinan <i>et al.</i> (2020b)
<i>P. domestica</i>		-	Blackford & Dinan (1997c)
<i>P. dulcis</i>		-	Dinan <i>et al.</i> (2020a)
<i>P. mume</i>		5.3	Wong <i>et al.</i> (1979)
<i>Pseudodrynaria (Aglaomorpha) coronans</i>	Polypodiaceae (F) [Polypodiaceae (F)]	+	Yen <i>et al.</i> (1974)
<i>Ptarmica cartilaginea</i>	Compositae (D)	-	Volodin <i>et al.</i> (2002)
<i>P. vulgaris</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Pteridium aquilinum</i>	Dennstaedtiaceae (F) [Dennstaedtiaceae (F)]	+ + + + +/- + + + + + +	Russell & Fenemore (1971) Kaplanis <i>et al.</i> (1967) McMorris and Voeller (1971) Jones & Firn (1978) Revina & Gureeva (1985) Svatos & Macek (1994) Vanek <i>et al.</i> (1990) Blackford & Dinan (1997b) Pemryk <i>et al.</i> (2013) Olennikov & Kashchenko (2018)
<i>P. aquilinum</i> var. <i>latiusculum</i>		(+) + + + 0.2 + + + + + + +	Takemoto <i>et al.</i> (1968e) Takemoto <i>et al.</i> (1967c) Takemoto <i>et al.</i> (1968j) Wong <i>et al.</i> (1979) Yen <i>et al.</i> (1974) Hikino <i>et al.</i> (1969c) Takemoto <i>et al.</i> (1973) Macek and Vanek (1994) Hikino <i>et al.</i> (1973)
<i>Pteridium esculentum</i> var. <i>yarrabense</i>		+	Saeng-ngam <i>et al.</i> (on-line)
<i>Pteris cretica</i>	Pteridaceae (F) [Pteridaceae (F)]	+ + + +/-	Imai <i>et al.</i> (1969c) Imai <i>et al.</i> (1969d) Matsuoka <i>et al.</i> (1969) Hikino <i>et al.</i> (1973)
<i>P. cretica</i> var. <i>albolineata</i>		+	Hikino <i>et al.</i> (1973)
<i>P. dispar</i>		+	Yen <i>et al.</i> (1974)
<i>P. ensiformis</i>		-	Hikino <i>et al.</i> (1973)
<i>P. fauriei</i> (<i>P. quadriaurita</i>)		-	Yen <i>et al.</i> (1974)
<i>P. grevilleana</i>		+	Yen <i>et al.</i> (1974)
<i>P. inaequalis</i>		+	Hikino <i>et al.</i> (1973)
<i>P. longipes</i>		+/- +	Hikino <i>et al.</i> (1973) Yen <i>et al.</i> (1974)

<i>P. macilentata</i>		-	Russell & Fenemore (1971)
<i>P. multifida</i>		+	Matsuoka <i>et al.</i> (1969)
		+	Imai <i>et al.</i> (1969d)
			Takemoto <i>et al.</i> (1967c)
		+	Hikino <i>et al.</i> (1973)
<i>P. quadriaurita</i>		-	Hikino <i>et al.</i> (1973)
<i>P. ryukyuensis</i>		+	Hikino <i>et al.</i> (1973)
<i>P. semipinnata</i>		-	Hikino <i>et al.</i> (1973)
		+/-	Yen <i>et al.</i> (1974)
<i>P. setuloso-costulata</i>		+	Hikino <i>et al.</i> (1973)
<i>P. tokioi</i>		+	Hikino <i>et al.</i> (1973)
<i>P. tremula</i>		-	Russell & Fenemore (1971)
<i>P. tripartita</i>		+	Yen <i>et al.</i> (1974)
<i>P. vittata</i>		+	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
<i>P. wallichiana</i>		+	Yen <i>et al.</i> (1974)
		-	Hikino <i>et al.</i> (1973)
<i>P. yakuinsularis</i>		+	Hikino <i>et al.</i> (1973)
<i>Pterocarya stenoptera</i>	Juglandaceae (D) [Juglandaceae (E)]	2.0	Wong <i>et al.</i> (1979)
<i>Pterospermum rubiginosum</i>	Sterculiaceae (D) [Malvaceae (E)]	-	Sreejit (2014)
<i>Pueraria lobata</i>	Leguminosae-P. (D) [Fabaceae (E)]	+	Matsuoka <i>et al.</i> (1969)
<i>Pulicaria dysenterica</i>	Compositae (D) [Asteraceae (E)]	(+)	Blackford & Dinan (1997c)
<i>Pulmonaria obscura</i>	Boraginaceae (D) [Boraginaceae (E)]	+	Volodin <i>et al.</i> (2002)
<i>Pulsatilla alba</i>	Ranunculaceae (D)	+	Sarker <i>et al.</i> (1997c)
<i>P. alpina</i>	[Ranunculaceae (E)]	+	Sarker <i>et al.</i> (1997c)
<i>P. ambigua</i>		+	Sarker <i>et al.</i> (1997c)
<i>P. halleri</i>		-	Sarker <i>et al.</i> (1997c)
		-	Dinan <i>et al.</i> (2020b)
<i>P. cernua</i>		+	Xu <i>et al.</i> (2011)
<i>P. montana</i>		-	Sarker <i>et al.</i> (1997c)
<i>P. myrrhidifolia</i>		+	Sarker <i>et al.</i> (1997c)
<i>P. occidentalis</i>		+	Sarker <i>et al.</i> (1997c)
<i>P. patens</i>		-	Sarker <i>et al.</i> (1997c)
<i>P. patensis</i>		-	Sarker <i>et al.</i> (1997c)
<i>P. sulphurea</i>		+	Sarker <i>et al.</i> (1997c)
<i>P. turczaninovii</i>		-	Sarker <i>et al.</i> (1997c)
<i>P. vernalis</i>		-	Sarker <i>et al.</i> (1997c)
<i>P. vulgaris</i>		-	Sarker <i>et al.</i> (1997c)
<i>Punica granatum</i>	Lythraceae (D) [Lythraceae (E)]	-	Dinan <i>et al.</i> (2020a)
		-	Dinan <i>et al.</i> (2020b)
<i>Pupalia lappacea</i>	Amaranthaceae (D) [Amaranthaceae (E)]	+	Felix & Domingo (2008)
		+	Sreejit (2014)
<i>P. lappacea</i> var. <i>lappacea</i>		+	Sreejit <i>et al.</i> (2018)
<i>Puya chilensis</i>	Bromeliaceae (M)	-	Dinan <i>et al.</i> (2020b)
<i>P. coerulea</i>	[Bromeliaceae (M)]	-	Dinan <i>et al.</i> (2020b)

<i>Pyrola grandiflora</i>	Ericaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>P. minor</i>	[Ericaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>P. rotundifolia</i>		-	Volodin <i>et al.</i> (2002)
<i>Pyrola</i> sp.		-	Volodin <i>et al.</i> (2002)
<i>Pyrrosia hastata</i>	Polypodiaceae (F)	+	Hikino <i>et al.</i> (1973)
<i>P. linearifolia</i>	[Polypodiaceae (F)]	+	Hikino <i>et al.</i> (1973)
<i>P. lingua</i>		+	Takemoto <i>et al.</i> (1967c)
		+/-	Hikino <i>et al.</i> (1973)
		+/-	Yen <i>et al.</i> (1974)
<i>P. petiolosa</i>		5.4	Wong <i>et al.</i> (1979)
		+	Chou & Lu (1980)
<i>P. polydactylis</i>		+	Yen <i>et al.</i> (1974)
<i>P. serpens</i>		-	Russell & Fenemore (1971)
<i>Pyrus communis</i>	Rosaceae (D)	-	Dinan <i>et al.</i> (2020a)
	[Rosaceae (E)]		
Q			
<i>Quercus acutissima</i>	Fagaceae (D)	-	Takemoto <i>et al.</i> (1967c)
	[Fagaceae (E)]	2.7	Wong <i>et al.</i> (1979)
<i>Q. serrata</i>		-	Takemoto <i>et al.</i> (1967c)
<i>Q. variabilis</i>		2.5	Wong <i>et al.</i> (1979)
R			
<i>Rabdosia (Isodon) macrophylla</i>	Labiatae (D)	3.4	Wong <i>et al.</i> (1979)
	[Lamiaceae (E)]		
<i>Ranunculus aconitifolius</i>	Ranunculaceae (D)	-	Dinan <i>et al.</i> (2002a)
<i>R. alpestris</i>	[Rannunculaceae (E)]	-	Dinan <i>et al.</i> (2002a)
<i>R. arvensis</i>		-	Dinan <i>et al.</i> (2001d)
		-	Dinan <i>et al.</i> (2002a)
<i>R. cortusifolius</i>		(+)	Dinan <i>et al.</i> (2002a)
<i>R. flammula</i>		-	Volodin <i>et al.</i> (2002)
<i>R. glacialis</i>		-	Dinan <i>et al.</i> (2002a)
<i>R. gramineus</i>		-	Dinan <i>et al.</i> (2002a)
<i>R. japonicus</i>		0	Wong <i>et al.</i> (1979)
<i>R. languinosa</i>		-	Dinan <i>et al.</i> (2001d)
<i>R. languinosus</i>		-	Dinan <i>et al.</i> (2002a)
<i>R. pallasii</i>		-	Volodin <i>et al.</i> (2002)
<i>R. parnassifolius</i>		-	Dinan <i>et al.</i> (2002a)
<i>R. pygmaeus</i>		-	Volodin <i>et al.</i> (2002)
<i>R. pyrenaicus</i>		-	Dinan <i>et al.</i> (2002a)
<i>R. repens</i>		-	Volodin <i>et al.</i> (2002)
<i>Ranunculus</i> sp.		-	Volodin <i>et al.</i> (2002)
<i>Raphanus raphanistrum sativa</i>	Cruciferae (D)	-	Dinan <i>et al.</i> (2020a)
	[Brassicaceae (E)]		
<i>Rauvolfia serpentina</i>	Apocynaceae (D)	-	Sreejit & Nelshi (2019)
	[Apocynaceae (E)]		
<i>Reineckea carnea</i>	Convallariaceae (M)	+	Wang <i>et al.</i> (2012)
	[Asparagaceae (M)]		
<i>Rhagodia baccata</i>	Chenopodiaceae (D)	+	Dinan <i>et al.</i> (1998)
	[Amaranthaceae (E)]	+	Dinan <i>et al.</i> (1999)
<i>R. baccata</i> ssp. <i>dioica</i>		+	Dinan <i>et al.</i> (1998)

<i>R. candolleana</i>		+	Dinan (1995b)
		+	Dinan <i>et al.</i> (1998)
<i>R. latifolia</i>		+	Dinan <i>et al.</i> (1998)
<i>R. papabolica</i>		+	Dinan <i>et al.</i> (1998)
<i>R. preissii</i>		+	Dinan <i>et al.</i> (1998)
<i>R. spinescens</i>		+	Bergamasco & Horn (1983)
		+	Dinan <i>et al.</i> (1998)
<i>Rhaponticum acaule</i>	Compositae (D)	+	Zughdani <i>et al.</i> (2020)
<i>Rh. (Leuzea) carthamoides</i>	[Asteraceae (E)]	+	Abubakirov (1982)
		+	Krasnov <i>et al.</i> (1976)
		+	Yakubova & Sakharova (1980)
		+	Mamatkhanov <i>et al.</i> (1981)
		+	Vereskovskii <i>et al.</i> (1983)
		+	Abubakirov (1984)
		+	Mamatkhanov <i>et al.</i> (1984)
		+	Baltaev & Abubakirov (1987)
		+	Volodin <i>et al.</i> (1993)
		+	Orlova <i>et al.</i> (1993)
		+	Baltaev (1991)
		+	Baltaev (1995)
		+	Orlova <i>et al.</i> (1994)
		+	Ramazanov <i>et al.</i> (1997a)
		+	Ramazanov <i>et al.</i> (1997b)
		+	Sadykov <i>et al.</i> (1997)
		+	Timofeev <i>et al.</i> (1998)
		+	Orlova <i>et al.</i> (1998)
		+	Geszprych & Weglarz (2002)
		+	Miliauskas <i>et al.</i> (2005)
		+	Huang <i>et al.</i> (2008)
		+	Wu <i>et al.</i> (2017)
		+	Olennikov & Kashchenko (2018)
		+	Asyakina <i>et al.</i> (2021)
<i>Rh. integrifolium</i>		+	Abubakirov (1982)
		+	Ganiev (1975)
		+	Baltaev <i>et al.</i> (1977)
		+	Baltaev <i>et al.</i> (1978a)
		+	Baltaev <i>et al.</i> (1978b)
		+	Yakubova <i>et al.</i> (1978)
		+	Mamatkhanov <i>et al.</i> (1979)
		+	Abubakirov (1980)
		+	Ganiev (1980)
		+	Abubakirov (1984)
		+	Saatov <i>et al.</i> (1999)
		+	Sagdullaev <i>et al.</i> (1999)
		+	Alieva <i>et al.</i> (2009)
		+	Namuna (2019)
<i>Rh. karatavicum</i>		+	Ganiev (1980)
		+	Tuleuov (2016)
<i>Rh. luratum</i>		+	Abubakirov (1982)
		+	Abubakirov (1984)
<i>Rh. lyratum</i>		+	Vereskovskii <i>et al.</i> (1983)
<i>Rh. nanum</i>		+	Abubakirov (1982)
		+	Baltaev <i>et al.</i> (1981)
		+	Ganiev (1975)
		+	Ganiev (1980)
		+	Abubakirov (1984)
		+	Saatov <i>et al.</i> (1999)
<i>Rh. nitidum</i>		+	Ganiev (1980)
<i>Rh. orientale</i>		+	Yakubova <i>et al.</i> (1978)

<i>Rh. pulchrum</i>		+	Vereskovskii <i>et al.</i> (1983)
<i>Rh. scariosum</i>		+	Vereskovskii <i>et al.</i> (1983)
<i>Rh. serratuloides</i>		+	Tuleuov (2016)
<i>Rh. uniflorum</i>		+	Gao <i>et al.</i> (1991)
		+	Guo <i>et al.</i> (1991)
		+	Guo & Lou (1992)
		+	Jiang & Li (1997)
		+	Chen & Ding (1997)
		+	Li <i>et al.</i> (1998)
		+	Li <i>et al.</i> (2000a)
		+	Li <i>et al.</i> (2000b)
		+	Zhang <i>et al.</i> (2001)
		+	Zhang & Wang (2001)
		+	Wang <i>et al.</i> (2001)
		+	Cheng <i>et al.</i> (2002)
		+	Du <i>et al.</i> (2016)
		+	Chen <i>et al.</i> (2017)
		+	Huang <i>et al.</i> (2017)
		+	Olennikov (2018b)
		+	Olennikov (2018c)
		+	Olennikov & Kashchenko (2018)
		+	Olennikov & Kaschenko (2019a)
		+	Olennikov & Kashenko (2019d)
<i>Rheum moorcroftiana</i>	Polygonaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>R. rhaponticum</i>	[Polygonaceae (E)]	-	Blackford <i>et al.</i> (1996)
		-	Blackford & Dinan (1997b)
<i>Rhinanthus vernalis</i>	Scrophulariaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Orobanchaceae (E)]		
<i>Rhodiola rosea</i>	Crassulaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Crassulaceae (E)]		
<i>Rhododendron albrechtii</i>	Ericaceae (D)	-	Dinan <i>et al.</i> (2001d)
	[Ericaceae (E)]		
<i>Rhus chinensis</i>	Anacardiaceae (D)	0.6	Wong <i>et al.</i> (1979)
	[Anacardiaceae (E)]		
<i>Rhyneosinapsis (Coincya) monensis</i>	Cruciferae (D)	(+)	Dinan <i>et al.</i> (2001d)
	[Brassicaceae (E)]		
<i>Ribes dikuscha</i>	Grossulariaceae (D)	-	Dinan <i>et al.</i> (2001d)
<i>R. hispidulum</i>	[Grossulariaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>R. nigrum</i>		-	Dinan <i>et al.</i> (2020b)
<i>R. rubrum</i>		-	Dinan <i>et al.</i> (2020a)
<i>Ricinus communis</i>	Euphorbiaceae (D)	-	Blackford <i>et al.</i> (1996)
	[Euphorbiaceae (E)]		
<i>Rivina aurantica</i>	Phytolaccaceae (D)	-	Dinan <i>et al.</i> (2001d)
<i>R. humilis</i>	[Petiveriaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Robinia pseudoacacia</i>	Leguminosae-P. (D)	-	Dinan <i>et al.</i> (2020a)
	[Fabaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Rodgersia podophylla</i>	Saxifragaceae (D)	-	Takemoto <i>et al.</i> (1967c)
	[Saxifragaceae (E)]		

<i>Rorippa amphibia</i>	Cruciferae (D)	-	Volodin <i>et al.</i> (2002)
<i>R. palustris</i>	[Brassicaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Rosa acicularis</i>	Rosaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Rosaceae (E)]		
<i>Roscoea scillifolia</i>	Zingiberaceae (M)	-	Dinan <i>et al.</i> (2020b)
	[Zingiberaceae (M)]		
<i>Rubia pergrina</i>	Rubiaceae (D)	-	Blackford & Dinan (1997c)
	[Rubiaceae (E)]		
<i>Rubus arcticus</i>	Rosaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>R. biflorus</i>	[Rosaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>R. chamaemorus</i>		-	Volodin <i>et al.</i> (2002)
<i>R. fruticosus</i>		-	Dinan <i>et al.</i> (2020a)
<i>R. humilifolius</i>		-	Volodin <i>et al.</i> (2002)
<i>R. idaeus</i>		-	Dinan <i>et al.</i> (2020a)
<i>R. ludwigii</i>		-	Dinan <i>et al.</i> (2020b)
<i>R. parvifolius</i>		5.3	Wong <i>et al.</i> (1979)
<i>Rumex acetosella</i>	Polygonaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>R. graminifolius</i>	[Polygonaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>R. lapponicus</i>		-	Volodin <i>et al.</i> (2002)
<i>R. obtusifolius</i>		-	Blackford & Dinan (1997b)
		(+)	Dinan <i>et al.</i> (2001d)
<i>Rumohra adiantiformis</i>	Davalliaceae (F)	+	Russell & Fenemore (1971)
<i>R. amabilis</i>	[Dryopteridaceae (F)]	-/+	Hikino <i>et al.</i> (1973)
<i>R. aristata</i>		-	Hikino <i>et al.</i> (1973)
<i>R. assamica</i>		-	Hikino <i>et al.</i> (1973)
<i>R. hispida</i>		+	Russell & Fenemore (1971)
<i>R. maximowiczii</i>		+	Hikino <i>et al.</i> (1973)
<i>R. miqueliana</i>		+	Hikino <i>et al.</i> (1973)
		+	Takemoto <i>et al.</i> (1967c)
<i>R. nipponica</i>		-	Hikino <i>et al.</i> (1973)
<i>R. pseudo-aristata</i>		-/+	Hikino <i>et al.</i> (1973)
<i>R. simplicior</i>		-/+	Hikino <i>et al.</i> (1973)
<i>R. standishii</i>		+	Hikino <i>et al.</i> (1973)
		-	Takemoto <i>et al.</i> (1967c)
<i>Ruscus aculeatus</i>	Ruscaceae (M)	-	Dinan <i>et al.</i> (2020b)
	[Asparagaceae (M)]		
S			
<i>Sabina (Juniperus) chinensis</i>	Cupressaceae (G)	2.0	Wong <i>et al.</i> (1979)
	[Cupressaceae (G)]		
<i>Saccharum ravennae</i>	Gramineae (M)	-	Dinan (1995a)
	[Poaceae (M)]		
<i>Sagina apetala</i>	Caryophyllaceae (D)	+	Zibareva <i>et al.</i> (2007a)
<i>S. ciliata</i>	[Caryophyllaceae (E)]	+	Zibareva <i>et al.</i> (2007a)
<i>S. japonica</i>		+	Jia <i>et al.</i> (2010)
<i>S. maritima</i>		+	Zibareva <i>et al.</i> (2007a)
<i>S. maxima</i>		+	Novozhilova <i>et al.</i> (2015)
		+	Novozhilova <i>et al.</i> (2014)
<i>S. procumbens</i>		+	Volodin <i>et al.</i> (2002)
		+	Zibareva <i>et al.</i> (2007a)
<i>S. subulata</i>		-	Zibareva <i>et al.</i> (2003)

<i>Sagittaria sagittifolia</i> <i>Sagittaria</i> sp.	Alismataceae (M)- [Alismataceae (M)]	-	Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002)
<i>Salicornia europea</i> <i>S. stricta</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	- -	Dinan <i>et al.</i> (1998) Dinan <i>et al.</i> (1998)
<i>Salicornis perrenis</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1991)
<i>Salix aurita</i> <i>S. caprea</i> <i>S. cinerea</i> <i>S. glauca</i> <i>S. herbacea</i> <i>S. jensseensis</i> <i>S. lanata</i> <i>S. nummularia</i> <i>S. pentandra</i>	Salicaceae (D) [Salicaceae (E)]	- - - - - - - - -	Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002) Blackford & Dinan (1997b) Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002) Volodin <i>et al.</i> (2002)
<i>Salpichroa origanifolia</i>	Solanaceae (D) [Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
<i>Salpiglossis sinuata</i>	Solanaceae (D) [Solanaceae (E)]	-/(+)	Savchenko <i>et al.</i> (2000)
<i>Salsola crassa</i> <i>S. imbricata</i> <i>S. jordanicola</i> <i>S. kali</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	- - - -	Dinan <i>et al.</i> (1998) Dinan <i>et al.</i> (1998) Dinan <i>et al.</i> (1998) Bathory <i>et al.</i> (1984)
<i>S. kali</i> ssp. <i>ruthenica</i> <i>S. rigida</i> <i>S. soda</i>		(+) - - -	Blackford & Dinan (1997c) Dinan <i>et al.</i> (1998) Dinan <i>et al.</i> (1998) Dinan <i>et al.</i> (1998)
<i>Salvia hispanica</i> <i>S. plebia</i> <i>S. multiorrhiza</i> <i>S. roemeriana</i> <i>S. virgata</i>	Labiatae (D) [Lamiaceae (E)]	- 2.0 3.0 (+) -	Dinan <i>et al.</i> (2020a) Wong <i>et al.</i> (1979) Wong <i>et al.</i> (1979) Dinan <i>et al.</i> (2001d) Dinan <i>et al.</i> (2001d)
<i>Salvinia natans</i>	Salviniaceae (F) [Salviniaceae (F)]	+	Hikino <i>et al.</i> (1973)
<i>Samadera (Quassia) indica</i>	Simaroubaceae (D) [Simaroubaceae (E)]	-	Sreejit & Nelshi (2019)
<i>Sambucus racemosa</i>	Caprifoliaceae (D) [Adoxaceae (E)]	5.4	Wong <i>et al.</i> (1979)
<i>Sanguisorba minor</i>	Rosaceae (D) [Rosaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Sapium sebiferum</i>	Euphorbiaceae (D) [Euphorbiaceae (E)]	1.0	Wong <i>et al.</i> (1979)
<i>Saponaria bellidifolia</i> <i>S. calabrica</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	+ - +	Zibareva <i>et al.</i> (2007a) Zibareva <i>et al.</i> (2003) Zibareva <i>et al.</i> (2007a)

<i>S. hausknechtii</i>		+	Bespayeve <i>et al.</i> (2012)
<i>S. lutea</i>		+	Bespayeve <i>et al.</i> (2012)
<i>S. ocymoides</i>		-	Zibareva <i>et al.</i> (2003)
		-	Zibareva <i>et al.</i> (2003)
		+	Bespayeve <i>et al.</i> (2012)
<i>S. officinalis</i>		-	Revina <i>et al.</i> (1988)
		-	Zibareva <i>et al.</i> (2003)
		+	Bespayeve <i>et al.</i> (2012)
<i>S. pumilla</i>		-	Zibareva <i>et al.</i> (2003)
<i>S. vaccaria</i>		-	Zibareva <i>et al.</i> (2003)
<i>Saponaria</i> sp.		-	Volodin <i>et al.</i> (2002)
<i>Saraca asoca</i>	Leguminosae-C. (D) [Fabaceae (E)]	-	Sreejit (2014)
<i>Saussurea alpina</i>	Compositae (D)	-	Volodin <i>et al.</i> (2002)
<i>S. latifolia</i>	[Asteraceae (E)]	+	Volodin <i>et al.</i> (1993)
<i>S. parviflora</i>		-	Volodin <i>et al.</i> (2002)
<i>Saxegothaea conspicua</i>	Podocarpaceae (G) [Podocarpaceae (G)]	+	Staal (1967)
<i>Saxifraga cernua</i>	Saxifragaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>S. hirculus</i>	[Saxifragaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Scabiosa farinosa</i>	Dipsacaceae (D) [Caprifoliaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Schefflera octophylla</i>	Araliaceae (D) [Araliaceae (E)]	-	Volodin <i>et al.</i> (2018)
<i>Schizanthus hookerianus</i>	Solanaceae (D)	-	Savchenko <i>et al.</i> (2000)
<i>S. pinnatus</i>	[Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
<i>S. tricolor</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. x wisetonensis</i>		-/(+)	Savchenko <i>et al.</i> (2000)
<i>Schizea dichotoma</i>	Schizaeaceae (F) [Schizaeaceae (F)]	+	Russell & Fenemore (1971)
		+	Bergamasco & Horn (1983)
		+	Fuchino <i>et al.</i> (1997)
<i>Sciadopytis verticillata</i>	Taxodiaceae (G) [Sciadopytiaceae (G)]	+	Takemoto <i>et al.</i> (1967c)
		+	Matsuoka <i>et al.</i> (1969)
<i>Scirpus lacustris</i>	Cyperaceae (M)	-	Volodin <i>et al.</i> (2002)
<i>S. sylvaticus</i>	[Cyperaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Scleranthus annuus</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Scleroblitum atriplicinum</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Sclerochlamys brachyptera</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Sclerolaena bicornis</i>	Chenopodiaceae (D)	-	Dinan <i>et al.</i> (1998)
<i>S. burbridgeae</i>	[Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>S. caricuspis</i>		-	Dinan <i>et al.</i> (1998)
<i>S. cornishiana</i>		-	Dinan <i>et al.</i> (1998)
<i>S. cuneata</i>		-	Dinan <i>et al.</i> (1998)

<i>S. densiflora</i>		-	Dinan <i>et al.</i> (1998)
<i>S. diacantha</i>		-	Dinan <i>et al.</i> (1998)
<i>S. eriacantha</i>		-	Dinan <i>et al.</i> (1998)
<i>S. euriotioides</i>		-	Dinan <i>et al.</i> (1998)
<i>S. lanicuspis</i>		-	Dinan <i>et al.</i> (1998)
<i>S. microcarpa</i>		-	Dinan <i>et al.</i> (1998)
<i>S. ventricosa</i>		-	Dinan <i>et al.</i> (1998)
<i>Sclerostegia tenuis</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Scopolia carniolica</i>	Solanaceae (D) [Solanaceae (E)]	-	Savchenko <i>et al.</i> (2000)
<i>Scorzonera humilis</i>	Compositae (D) [Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Scrophularia ningpoensis</i>	Scrophulariaceae (D)	3.0	Wong <i>et al.</i> (1979)
<i>S. scopolii</i>	[Scrophulariaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>S. vernalis</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>Scutellaria galericulata</i>	Labiatae (D)	-	Volodin <i>et al.</i> (2002)
<i>S. indica</i>	[Lamiaceae (E)]	4.0	Wong <i>et al.</i> (1979)
<i>Securinega (Flueggea) suffruticosa</i>	Euphorbiaceae (D) [Phyllanthaceae (E)]	0	Wong <i>et al.</i> (1979)
<i>Sechium edule</i>	Cucurbitaceae (D) [Cucurbitaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Sedum acre</i>	Crassulaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>S. album</i>	[Crassulaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>S. onychopetalum</i>		3.0	Wong <i>et al.</i> (1979)
<i>S. sanmentosum</i>		9.0	Wong <i>et al.</i> (1979)
<i>S. stoleniferum</i>		+	Chou & Lu (1980)
<i>S. telephium</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>S. verticillatum</i>		-	Clément & Dinan (1991)
		-	Dinan <i>et al.</i> (2020b)
<i>Selaginella deliculata</i>	Selaginellaceae (FA)	-	Yen <i>et al.</i> (1974)
<i>S. doederleinii</i>	[Selaginellaceae (Lyc)]	-	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)
<i>S. mollendorffii</i>		-	Yen <i>et al.</i> (1974)
<i>S. nipponica</i>		-	Hikino <i>et al.</i> (1973)
<i>S. pachystachys</i>		-	Hikino <i>et al.</i> (1973)
<i>S. pseudo-involvens</i>		-	Yen <i>et al.</i> (1974)
<i>S. remotifolia</i> var. <i>japonica</i>		-	Takemoto <i>et al.</i> (1967c)
		-	Hikino <i>et al.</i> (1973)
<i>S. tamariscina</i>		-	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)
<i>S. uncinata</i>		-	Hikino <i>et al.</i> (1973)
<i>Selenodesmium (Trichomanes) obscurum</i>	Hymenophyllaceae (F) [Hymenophyllaceae (F)]	-	Hikino <i>et al.</i> (1973)
<i>Selliguea hastata</i> (syn. <i>Phymatopteris hastata</i>)	Polypodiaceae (F) [Polypodiaceae (F)]	+	Yang <i>et al.</i> (2014)
<i>Semiaquilegya ecalcarata</i>	Ranunculacgae (D) [Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2002a)

<i>Senecio chrysanthemum</i>	Compositae (D)	(+)	Blackford & Dinan (1997a)
<i>S. jacobaea</i>	[Asteraceae (E)]	-	Blackford & Dinan (1997a)
<i>S. nemorensis</i>		-	Volodin <i>et al.</i> (2002)
<i>S. polydon</i>		-	Dinan <i>et al.</i> (2020b)
<i>S. scandens</i>		-	Blackford & Dinan (1997a)
<i>S. squalidus</i>		-	Blackford & Dinan (1997a)
<i>S. vulgaris</i>		(+)	Blackford & Dinan (1997a)
		-	Volodin <i>et al.</i> (2002)
<i>Senna occidentalis</i>	Leguminosae-C. (D)	-	Sreejit (2014)
<i>S. tora</i>	[Fabaceae (E)]	-	Sreejit (2014)
<i>Serratula</i> sp.	Compositae (D)	+	Zatsny <i>et al.</i> (1975)
<i>Serratula algida</i>	[Asteraceae (E)]	+	Abubakirov (1982)
		+	Ganiev (1980)
		+	Novoselskaya <i>et al.</i> (1981b)
		+	Abubakirov (1984)
		+	Saatov <i>et al.</i> (1999)
<i>S. cardunculus</i>		+	Bespayeva <i>et al.</i> (2012)
		+	Berkenov <i>et al.</i> (2016)
<i>S. centauroides</i>		+	Abubakirov (1982)
		+	Gorovits <i>et al.</i> (1974)
		+	Novoselskaya <i>et al.</i> (1981b)
		+	Abubakirov (1984)
		+	Vorob'eva <i>et al.</i> (2005)
		+	Nikolaeva <i>et al.</i> (2017)
		+	Olennikov & Kashchenko (2018)
		+	Olennikov & Kaschenko (2019c)
<i>S. chicoracea</i>		+	Larguet (2011)
		+	Aliouche <i>et al.</i> (2018)
<i>S. chinensis</i>		+	Chen & Wei (1989)
		+	Ling <i>et al.</i> (2003)
		+	Tang <i>et al.</i> (2014)
		+	Xu <i>et al.</i> (2016)
		+	Zhang <i>et al.</i> (2016)
		+	Zhang <i>et al.</i> (2017)
		+	Wu <i>et al.</i> (2019)
<i>S. coronata</i>		+	Abubakirov (1982)
		+	Novosenskaya <i>et al.</i> (1981b)
		+	Gorovits <i>et al.</i> (1974)
		+	Abubakirov (1984)
		+	Revina <i>et al.</i> (1986)
		+	Anufrieva <i>et al.</i> (1995)
		+	Volodin <i>et al.</i> (1993)
		+	Volodin <i>et al.</i> (1998a)
		+	Volodin <i>et al.</i> (1998b)
		+	Anufrieva <i>et al.</i> (1998)
		+	Kolegova & Volodin (1999)
		+	Galiautdinov <i>et al.</i> (1999)
		+	Báthori <i>et al.</i> (1999b)
		+	Odinokov <i>et al.</i> (2000)
		+	Alexeeva <i>et al.</i> (2000)
		+	Pugenov & Savinovskaya (2001)
		+	Odnikov <i>et al.</i> (2002)
		+	Filippova <i>et al.</i> (2003)
		+	Lomovsky <i>et al.</i> (2003)
		+	Ramazanov (2005b)
		+	Odnikov <i>et al.</i> (2005)
		+	Martinussen <i>et al.</i> (2011)

	+	Bespayeva <i>et al.</i> (2012)
	+	Galyautdinov <i>et al.</i> (2016)
	+	Temirgaziev <i>et al.</i> (2018)
	+	Olennikov & Kashchenko (2018)
	+	Dinan <i>et al.</i> (2020b)
<i>S. cupuliformis</i>	+	Zibareva <i>et al.</i> (2017)
<i>S. erucifolia</i>	+	Bespayeva <i>et al.</i> (2012)
<i>S. gmelini</i>	+	Bespayeva <i>et al.</i> (2012)
	+	Zibareva <i>et al.</i> (2017)
<i>S. inermis</i>	+	Yatsuk & Segal (1970)
	+	Volodin <i>et al.</i> (1993)
<i>S. kirghisorum</i>	+	Bespayeva <i>et al.</i> (2012)
<i>S. komarovii</i>	+	Vorob'eva <i>et al.</i> (2004)
	+	Rybin <i>et al.</i> (2007)
<i>S. longicalycina</i>	+	Ramazanov 2005b)
<i>S. lyratifolia</i>	+	Ganiev (1980)
<i>S. manshurica</i>	+	Zibareva <i>et al.</i> (2017)
<i>S. marginata</i>	+	Olennikov & Kashchenko (2018)
<i>S. procumbens</i>	+	Abubakirov (1982)
	+	Novoselskaya <i>et al.</i> (1981b)
	+	Abubakirov (1984)
<i>S. quinquefolia</i>	+	Abubakirov (1982)
	+	Gorovits <i>et al.</i> (1974)
	+	Novoselskaya <i>et al.</i> (1981b)
	+	Abubakirov (1984)
	+	Pylina <i>et al.</i> (2010)
	+	Bespayeva <i>et al.</i> (2012)
<i>S. radiata</i>	+	Bespayeva <i>et al.</i> (2012)
<i>S. sogdiana</i>	+	Ganiev (1975)
	+	Ganiev (1972)
	+	Abubakirov (1982)
	+	Zatsny <i>et al.</i> (1971)
	+	Zatsny <i>et al.</i> (1973)
	+	Novoselskaya <i>et al.</i> (1975)
	+	Ganiev (1980)
	+	Abubakirov (1980)
	+	Abubakirov (1984)
	+	Saatov <i>et al.</i> (1999)
<i>S. strangulata</i>	+	Dai <i>et al.</i> (2001)
	+	Dai <i>et al.</i> (2002)
	+	Wang <i>et al.</i> (2002)
<i>S. tinctoria</i>	+	Rudel <i>et al.</i> (1992)
	+	Akhmed <i>et al.</i> (1990)
	+	Báthori <i>et al.</i> (1986)
	+	Báthori <i>et al.</i> (1986b)
	+	Delbecque <i>et al.</i> (1995)
	+	Báthori <i>et al.</i> (1990)
	+	Corio-Costet <i>et al.</i> (1993)
	+	Corio-Costet <i>et al.</i> (1996)
	+	Báthori <i>et al.</i> (1996)
	+	Báthori <i>et al.</i> (1998a)
	+	Báthori <i>et al.</i> (1998b)
	+	Corio-Costet <i>et al.</i> (1999)
	+	Báthori <i>et al.</i> (1999b)
	+	Báthori <i>et al.</i> (2003)
<i>S. wolffii</i>	+	Miladera <i>et al.</i> (1992)
	+	Saad <i>et al.</i> (1992)
	+	Akhmed <i>et al.</i> (1990)
	+	Báthori <i>et al.</i> (1998b)
	+	Báthori <i>et al.</i> (1999b)

		+	Báthori <i>et al.</i> (2000a)
		+	Hunyadi <i>et al.</i> (2004)
		+	Kalász <i>et al.</i> (2006)
		+	Liktor-Busa <i>et al.</i> (2007a)
		+	Liktor-Busa <i>et al.</i> (2007b)
		+	Hunyadi <i>et al.</i> (2007a)
		+	Hunyadi <i>et al.</i> (2007b)
		+	Simon <i>et al.</i> (2007)
		+	Liktor-Busa <i>et al.</i> (2008)
		+	Liktor-Busa (2008)
		+	Simon <i>et al.</i> (2008)
		+	Takacs <i>et al.</i> (2010)
		+	Vanyolos (2012)
		+	Vanyolos <i>et al.</i> (2012)
		+	Nowak <i>et al.</i> (2013)
		+	Urbanska <i>et al.</i> (2014)
<i>S. xeranthemoides</i>		+	Kholodova <i>et al.</i> (1979)
		+	Kholodova (1981)
<i>S. xeranthemoides</i> (<i>S. erucifolia</i>)		+	Abubakirov (1982)
		+	Abubakirov (1984)
<i>Sesamum indicum</i>	Pedaliaceae (D) [Pedaliaceae (E)]	-	Dinan <i>et al.</i> (2020a)
<i>Sesuvium portulacastrum</i>	Aizoaceae (D) [Aizoaceae (E)]	+	Banerji <i>et al.</i> (1971)
		+	Sipahimalani <i>et al.</i> (1972)
		18	Wong <i>et al.</i> (1979)
		+	Bergamasco & Horn (1983)
		+	Shivakumar <i>et al.</i> (1995)
		+	Rele <i>et al.</i> (2003)
		+	Saeng-ngam <i>et al.</i> (on-line)
		+	Sreejit (2014)
		+	Kapare <i>et al.</i> (2016)
		+	Muchate <i>et al.</i> (2017)
		+	Kapare <i>et al.</i> (2017)
		+	Sreejit <i>et al.</i> (2018)
		+	Sreejit <i>et al.</i> (2019)
<i>Setaria italica</i>	Gramineae (M)	-	Dinan <i>et al.</i> (2001d)
<i>S. macrochaeta</i>	[Poaceae (M)]	-	Dinan (1995a)
<i>Sibbaldia procumbens</i>	Rosaceae (D) [Rosaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Sida acuta</i>	Malvaceae (D) [Malvaceae (E)]	+	Dinan <i>et al.</i> (2001b)
		+	Volodin <i>et al.</i> (2018)
<i>Sida carpinifolia</i> (<i>S. acuta</i>)		+	Pandit <i>et al.</i> (1976)
<i>S. cordifolia</i>		+	Prakash & Ghosal (1979)
		-	Dinan <i>et al.</i> (2001b)
		+	Dinan <i>et al.</i> (2020b)
<i>S.echinocarpa</i>		-	Dinan <i>et al.</i> (2001b)
<i>S. filicaulis</i>		+	Dinan <i>et al.</i> (2001b)
<i>S. filiformis</i>		-	Dinan <i>et al.</i> (2001b)
<i>S. glutinosa</i>		+	Das <i>et al.</i> (2014)
<i>S. goniocarpa</i>		-	Dinan <i>et al.</i> (2001b)
<i>S. ovata</i>		-	Dinan <i>et al.</i> (2001b)
<i>S. pertrophila</i>		-	Dinan <i>et al.</i> (2001b)
<i>S. rhombifolia</i>		+	Prakash & Ghosal (1979)
		+	Dinan <i>et al.</i> (2001b)

		+	Jadhav <i>et al.</i> (2007)
		+	Wang <i>et al.</i> (2008)
		+	Volodin <i>et al.</i> (2018)
<i>S. rhomboidea</i>		+	Sreejit (2014)
		+	Sreejit <i>et al.</i> (2018)
<i>S. rholenae</i>		-	Dinan <i>et al.</i> (2001b)
<i>S. szechuensis</i>		+	Li <i>et al.</i> (2006)
<i>S. spinosa</i>		+	Prakash & Ghosal (1979)
		+	Darwish & Reinecke (2003)
<i>S. subspicata</i>		-	Dinan <i>et al.</i> (2001b)
<i>S. tuberculata</i>		+	Rosa <i>et al.</i> (2014)
		+	Silva da Rosa <i>et al.</i> (2015)
		+	da Rosa <i>et al.</i> (2016)
		+	da Rosa <i>et al.</i> (2018a)
		+	da Rosa <i>et al.</i> (2018b)
<i>Sidalcea candida</i>	Malvaceae (D) [Malvaceae (E)]	(+)	Dinan <i>et al.</i> (2020b)
<i>Silene acaulis</i>	Caryophyllaceae (D) [Caryophyllaceae (E)]	+	Zibareva (1995)
		+	Zibareva (2000)
		+	Zibareva <i>et al.</i> (2003)
		+	Zibareva <i>et al.</i> (2007a)
<i>S. alba</i>		-	Zibareva (2000)
		-	Zibareva <i>et al.</i> (2003)
<i>S. alpestris</i>		-	Zibareva (2000)
		-	Zibareva <i>et al.</i> (2003)
		(+)	Dinan <i>et al.</i> (2020b)
<i>S. altaica</i>		+	Zibareva (2000)
		+	Zibareva (1999)
		+	Báthori <i>et al.</i> (1995)
		-	Pongracz <i>et al.</i> (2003b)
		+/-	Zibareva <i>et al.</i> (2007b)
		+	Zibareva <i>et al.</i> (2007a)
		+	Agabekova <i>et al.</i> (2010)
		+	Bespayeveva <i>et al.</i> (2012)
<i>S. ambigua</i>		+	Zibareva <i>et al.</i> (2007a)
<i>S. antirrhina</i>		+	Zibareva <i>et al.</i> (2003)
		+	Meng <i>et al.</i> (2001d)
		+	Mamadalieva <i>et al.</i> (2004b)
		+	Zibareva <i>et al.</i> (2007b)
		+	Zibareva <i>et al.</i> (2007a)
<i>S. apetala</i>		+	Zibareva <i>et al.</i> (2003)
		-	Pongracz <i>et al.</i> (2003b)
		+	Zibareva <i>et al.</i> (2007a)
<i>S. aprica (Elisanthe aprica)</i>		-	Olenikov & Kashchenko (2018)
<i>S. armeria</i>		-	Zibareva (1997)
		+	Zibareva (2000)
		+	Báthori <i>et al.</i> (1995)
		-	Zibareva <i>et al.</i> (2003)
		+	Pongracz <i>et al.</i> (2003b)
		+/-	Zibareva <i>et al.</i> (2007b)
		-	Dinan <i>et al.</i> (2020b)
<i>S. asterias</i>		-	Zibareva (2000)
		-	Zibareva <i>et al.</i> (2003)
		-	Dinan <i>et al.</i> (2020b)
<i>S. atigraca</i>		-	Dinan <i>et al.</i> (2020b)
<i>S. atropurpurea</i>		-	Dinan <i>et al.</i> (2020b)
<i>S. balchaschensis</i>		-	Agabekova <i>et al.</i> (2010)
<i>S. bashkirorum</i>		+	Zibareva (2000)

<i>S. bellidifolia</i>	+	Zibareva <i>et al.</i> (2007a)
	+	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2003)
	-	Pongracz <i>et al.</i> (2003b)
	+	Zibareva <i>et al.</i> (2007b)
<i>S. bellidioides</i>	+	Zibareva <i>et al.</i> (2007a)
	-	Zibareva <i>et al.</i> (2003)
	-	Dinan <i>et al.</i> (2020b)
<i>S. bergiana</i>	+	Zibareva <i>et al.</i> (2007a)
	-	Pongracz <i>et al.</i> (2003b)
<i>S. boryi</i>	-	Zibareva (2000)
<i>S. borystenica</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. bourgeaui</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. brachypoda</i>	+	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. brahuica</i>	+	Abubakirov (1982)
	+	Saatov <i>et al.</i> (1981)
	+	Saatov <i>et al.</i> (1982)
	+	Saatov <i>et al.</i> (1982b)
	+	Abubakirov (1984)
	+	Saatov <i>et al.</i> (1984a)
	+	Saatov <i>et al.</i> (1984b)
	+	Saatov <i>et al.</i> (1986b)
	+	Dzhukharova <i>et al.</i> (1991)
	+	Saatov <i>et al.</i> (1993)
	+	Dzhugarova <i>et al.</i> (1993)
	+	Dzhukharova <i>et al.</i> (1994a)
	+	Dzhukharova <i>et al.</i> (1994b)
	+	Dzhukharova <i>et al.</i> (1995)
	+	Báthori <i>et al.</i> (1995)
	+	Sadikov & Saatov (1998)
	+	Saatov <i>et al.</i> (1999)
	+	Sadikov & Saatov (1999)
	+	Mamedov <i>et al.</i> (2017)
<i>S. bupleroides</i>	-	Zibareva (2000)
	-	Pongracz <i>et al.</i> (2003b)
<i>S. burchellii</i>	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. campanula</i>	-	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2003)
<i>S. campanulata</i>	+	Zibareva (1999)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. caramanica</i>	+	Zibareva (1999)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. caroliniana</i>	-	Zibareva <i>et al.</i> (2003)
	-	Dinan <i>et al.</i> (2020b)
<i>S. catholica</i>	+	Zibareva (2000)
	+	Zibareva (1999)
	+	Bathori <i>et al.</i> (1995)
	-	Pongracz <i>et al.</i> (2003b)
	+	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. caucasica</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. chamarensis</i>	+	Saatov <i>et al.</i> (1993)
	+	Revina <i>et al.</i> (1988)
	+	Olennikov & Kashchenko (2018)
<i>S. chlorantha</i>	+	Sviridova <i>et al.</i> (1995)
	+	Zibareva (1995)
	+	Zibareva (2000)
	+	Pongracz <i>et al.</i> (2003b)

	+	Zibareva <i>et al.</i> (2007a)
	+	Olennikov & Kashchenko (2018)
<i>S. chlorifolia</i>	+	Zibareva (2000)
	+	Zibareva (1999)
	+	Meng <i>et al.</i> (2001d)
	+	Mamadaliyeva <i>et al.</i> (2004b)
	+	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. chlorifolia cordifolia</i>	-	Pongracz <i>et al.</i> (2003b)
<i>S. ciliata</i>	+	Zibareva (2000)
	+	Báthori <i>et al.</i> (1995)
	-	Pongracz <i>et al.</i> (2003b)
<i>S. ciliata</i> var. <i>graefferi</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. clandestina</i>	+	Zibareva (2000)
<i>S. claviformis</i>	+	Zibareva <i>et al.</i> (2003)
	+	Sadikov <i>et al.</i> (2001)
	+	Ramazonov <i>et al.</i> (2020)
	+	Yusupova <i>et al.</i> (2020c)
<i>S. coeli-rosa</i>	-	Zibareva (2000)
	+	Báthori <i>et al.</i> (1995)
	-	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
<i>S. colorata</i>	+	Zibareva & Yeryomina (1996)
	+	Zibareva (1995)
	+	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2003)
	-	Pongracz <i>et al.</i> (2003b)
	+	Zibareva <i>et al.</i> (2007a)
	+	Termentzi <i>et al.</i> (2014)
<i>S. colorata</i> ssp. <i>trichocalycina</i>	+	Zibareva (2000)
<i>S. colpophylla</i>	+	Zibareva <i>et al.</i> (2014)
	+	Seliverstova <i>et al.</i> (2014)
	+	Badulina & Zibareva (2015)
<i>S. compacta</i>	-	Zibareva (2000)
	-	Pongracz <i>et al.</i> (2003b)
	-	Dinan <i>et al.</i> (2020b)
<i>S. conica</i>	-	Zibareva (1997)
	-	Zibareva (2000)
	-	Zibareva <i>et al.</i> (2003)
	-	Pongracz <i>et al.</i> (2003b)
<i>S. conoidea</i>	-	Zibareva (2000)
<i>S. coronaria</i>	+	Zibareva (1999)
	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. cretaceae</i>	+	Bespayeva <i>et al.</i> (2012)
	+	Tuleuov <i>et al.</i> (2014)
	+	Temirgazivev <i>et al.</i> (2016)
<i>S. cretica</i>	+	Zibareva & Yeryomina (1996)
	+	Zibareva (1997)
	+	Zibareva <i>et al.</i> (1997)
	+	Zibareva <i>et al.</i> (2003)
	-	Pongracz <i>et al.</i> (2003b)
	+	Meng <i>et al.</i> (2001d)
	+	Mamadaliyeva <i>et al.</i> (2004b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. cucubalus</i>	+	Pongracz <i>et al.</i> (2003b)
<i>S. damboldtiana</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. delavayi</i>	+	Zibareva <i>et al.</i> (2007a)
	+	Dinan <i>et al.</i> (2020b)

<i>S. densiflora</i>	+	Sviridova <i>et al.</i> (1995)
	+	Zibareva (2000)
	-	Pongracz <i>et al.</i> (2003b)
<i>S. dichotoma</i>	+	Zibareva <i>et al.</i> (2007a)
	-	Zibareva (2000)
	-	Pongracz <i>et al.</i> (2003b)
<i>S. dinarica</i>	-	Zibareva (2000)
<i>S. dioica</i>	+	Girault <i>et al.</i> (1996)
	+	Saatov <i>et al.</i> (1993)
	-	Zibareva (1997)
	-	Zibareva (2000)
	-	Zibareva <i>et al.</i> (2003)
	-	Pongracz <i>et al.</i> (2003b)
	-	Dinan <i>et al.</i> (2020b)
<i>S. disticha</i>	+	Zibareva & Yeryomina (1996)
	+	Zibareva (1995)
	+	Zibareva (1997)
	+	Meng <i>et al.</i> (2001d)
	+	Zibareva <i>et al.</i> (2003)
	+	Mamadaliyeva <i>et al.</i> (2004b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. divaricata</i>	-	Zibareva (2000)
<i>S. echinata</i>	+	Zibareva <i>et al.</i> (2003)
	+	Meng <i>et al.</i> (2001d)
	+	Pongracz <i>et al.</i> (2003b)
	+	Mamadaliyeva <i>et al.</i> (2004b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. elegans</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. fabaria</i>	-	Zibareva (2000)
<i>S. fabariodes</i>	-	Zibareva (2000)
	+	Pongracz <i>et al.</i> (2003b)
<i>S. fetissovii</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. fimbriata</i>	+	Dinan <i>et al.</i> (2020b)
<i>S. firma</i>	+	Zibareva (1999)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. flavescens</i>	+	Zibareva (1999)
	+	Báthori <i>et al.</i> (1995)
	+	Pongracz <i>et al.</i> (2003b)
<i>S. foliosa</i>	+	Zibareva <i>et al.</i> (2007a)
	+	Novozhilova <i>et al.</i> (2014)
	+	Novozhilova <i>et al.</i> (2015)
<i>S. fortunei</i>	+	Gaidi <i>et al.</i> (2002)
<i>S. frivaldszkyana</i>	+	Zibareva <i>et al.</i> (1997)
	+	Sviridova <i>et al.</i> (1995)
	+	Zibareva (1997)
	+	Louden <i>et al.</i> (2002)
	+	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
	+	Mamadaliyeva <i>et al.</i> (2004b)
	+	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
	+	Zibareva <i>et al.</i> (2009)
<i>S. fruticosa</i>	+	Zibareva <i>et al.</i> (1997)
	+	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. fruticulosa</i>	+	Bespayeveva <i>et al.</i> (2012)
	-	Pongracz <i>et al.</i> (2003b)
	+	Temirgaziev <i>et al.</i> (2016)
	+	Termirgaziev <i>et al.</i> (2018b)
<i>S. fuscata</i>	-	Zibareva (2000)

<i>S. gallica</i>	+	Bergamasco & Horn (1983)
	+	Báthori <i>et al.</i> (1995)
	+	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
	+	Nair <i>et al.</i> (2010)
<i>S. gallica</i> var. <i>quinquevulnera</i>	+	Zibareva (2000)
	+	Zibareva (1999)
	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. gebleriana</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. gigantea</i>	+	Zibareva (2000)
	+	Zibareva (1999)
	+	Zibareva <i>et al.</i> (2007a)
	+	Zibareva <i>et al.</i> (2009)
<i>S. goulimyi</i>	+	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. graminifolia</i>	+	Saatov <i>et al.</i> (1993)
	+	Revina <i>et al.</i> (1988)
	+	Zibareva (1997)
	+	Zibareva <i>et al.</i> (2007a)
	+	Zibareva <i>et al.</i> (2009)
<i>S. guntensis</i>	+	Mamadalieva <i>et al.</i> (2011)
	+	Glashkin <i>et al.</i> (2014)
	+	Mamedov <i>et al.</i> (2017)
<i>S. hifacensis</i>	+	Zibareva (2000)
<i>S. holopetala</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. hookeri</i>	+	Dinan <i>et al.</i> (2020b)
<i>S. inaperta</i>	-	Zibareva (2000)
<i>S. inflata</i>	+	Shivakumar <i>et al.</i> (1995)
	+	Pongracz <i>et al.</i> (2003b)
<i>S. italica</i>	+	Sviridova <i>et al.</i> (1995)
	+	Zibareva <i>et al.</i> (1997)
	+	Zibareva (1997)
	+	Zibareva <i>et al.</i> (1997)
	+	Meng <i>et al.</i> (2001d)
	+	Zibareva <i>et al.</i> (2003)
	+	Mamadalieva <i>et al.</i> (2004b)
	+	Zibareva <i>et al.</i> (2007a)
	+	Olennikov & Kashchenko (2019)
	+	Kashchenko & Olennikov (2019)
	+	Dinan <i>et al.</i> (2020b)
<i>S. italica</i> ssp. <i>nemoralis</i>	+	Báthori <i>et al.</i> (2000c)
	+	Báthori <i>et al.</i> (2002a)
	+	Báthori <i>et al.</i> (2002b)
	+	Báthori <i>et al.</i> (2002c)
	+	Pongrácz <i>et al.</i> (2003a)
	+	Simon <i>et al.</i> (2004)
<i>S. jenissensis</i>	+	Saatov <i>et al.</i> (1993)
	+	Revina <i>et al.</i> (1988)
	+	Zibareva (1997)
	+	Zibareva <i>et al.</i> (1997)
	+	Zibareva <i>et al.</i> (2007a)
	+	Novozhilova <i>et al.</i> (2014)
	+	Novozhilova <i>et al.</i> (2015)
	+	Olennikov & Kashchenko (2017)
	+	Olennikov & Kashchenko (2018)
<i>S. karkaralensis</i>	-	Agabekova <i>et al.</i> (2010)
<i>S. keiskei</i>	+	Zibareva <i>et al.</i> (2003)
	+	Dinan <i>et al.</i> (2020b)
<i>S. kungessana</i>	+	Zibareva <i>et al.</i> (2007a)

<i>S. laciniata</i>	+	Zibareva <i>et al.</i> (2003)
<i>S. laciniata</i> ssp. <i>angustifolia</i>	+	Dinan <i>et al.</i> (2020b)
<i>S. laeta</i>	-	Zibareva (2000)
<i>S. latifolia</i>	+	Abubakirov (1982)
	+	Abubakirov (1984)
	+	Saatov <i>et al.</i> (1993)
	+	Bathori <i>et al.</i> (1995)
	-	Zibareva <i>et al.</i> (2003)
(<i>Melandrium album</i>)	-	Olennikov & Kashchenko (2018)
<i>S. lerchenfeldiana</i>	-	Dinan <i>et al.</i> (2020b)
<i>S. linicola</i>	+	Zibareva & Yeryomina (1996)
	+	Zibareva (1997)
	+	Zibareva <i>et al.</i> (1997)
	+	Mamadaliyeva <i>et al.</i> (2002b)
	+	Zibareva <i>et al.</i> (2003)
	-	Pongracz <i>et al.</i> (2003b)
	+	Mamadaliyeva <i>et al.</i> (2004b)
	+	Zibareva <i>et al.</i> (2007a)
	+	Erst <i>et al.</i> (2016)
	+	Mamedov <i>et al.</i> (2017)
	+	Erst <i>et al.</i> (2018)
	+	Erst <i>et al.</i> (2019)
<i>S. longicalycina</i>	+	Abubakirov (1982)
	+	Abubakirov (1984)
	+	Saatov <i>et al.</i> (1993)
	+	Báthori <i>et al.</i> (1995)
<i>S. longicilia</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. longiflora</i>	+	Pongracz <i>et al.</i> (2003b)
<i>S. maritima</i>	-	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
	-	Dinan <i>et al.</i> (2020b)
<i>S. mellifera</i>	+	Zibareva <i>et al.</i> (1997)
	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007a)
	+	Zibareva <i>et al.</i> (2009)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. melzheimeri</i>	-	Dinan <i>et al.</i> (2020b)
<i>S. mexicana</i>	+	Zibareva & Yeryomina (1996)
<i>S. micropetala</i>	+	Saatov <i>et al.</i> (1993)
	+	Báthori <i>et al.</i> (1995)
	+	Zibareva (1997)
	-	Pongracz <i>et al.</i> (2003b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. mollissima</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. montbretiana</i>	+	Kilinç <i>et al.</i> (2018)
<i>S. multicaulis</i>	-	Zibareva (2000)
	+	Báthori <i>et al.</i> (1995)
	-	Pongracz <i>et al.</i> (2003b)
	+/-	Zibareva <i>et al.</i> (2007b)
<i>S. multiflora</i>	+	Revina <i>et al.</i> (1988)
	+	Báthori (1998)
	-	Zibareva <i>et al.</i> (2003)
	-	Pongracz <i>et al.</i> (2003b)
	+	Dinan <i>et al.</i> (2020b)
<i>S. nemoralis</i>	+	Sviridova <i>et al.</i> (1995)
	-	Pongracz <i>et al.</i> (2003b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. noctiflora</i>	-	Zibareva (2000)
	-	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)

<i>(Elisanthe noctiflora)</i>	-	Olennikov & Kashchenko (2018)
<i>S. nutans</i>	+	Girault <i>et al.</i> (1990)
	+	Báthori <i>et al.</i> (1986)
	+	Báthori <i>et al.</i> (1987)
	+	Revina <i>et al.</i> (1988)
	+	Baltaev <i>et al.</i> (1985a)
	+	Baltaev <i>et al.</i> (1985b)
	+	Báthori <i>et al.</i> (1986)
	+	Raynor <i>et al.</i> (1989)
	+	Read <i>et al.</i> (1990)
	+	Saatov <i>et al.</i> (1993)
	+	Davis <i>et al.</i> (1993)
	+	Lafont <i>et al.</i> (1993)
	+	Sviridova <i>et al.</i> (1995)
	+	Báthori <i>et al.</i> (1995)
	+	Zibareva (1997)
	+	Zibareva <i>et al.</i> (1997)
	+	Ramazanov <i>et al.</i> (1997c)
	+	Wilson & Morden (1999)
	+	Báthori (1998)
	+	Zibareva (2000)
	+	Louden <i>et al.</i> (2002)
	+	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
	+	Ramazanov <i>et al.</i> (2007)
	+	Olennikov & Kashchenko (2018)
	+	Dinan <i>et al.</i> (2020b)
	+	Zibareva <i>et al.</i> (2007a)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. obovata</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. odoratissima</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. oligantha</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. oreina</i>	-	Mamedov <i>et al.</i> (2017)
<i>S. orphanidis</i>	-	Zibareva (2000)
<i>S. otites</i>	+	Girault <i>et al.</i> (1990)
	+	Báthori <i>et al.</i> (1986)
	+	Báthory <i>et al.</i> (1986)
	+	Báthori <i>et al.</i> (1986b)
	+	Báthori (1986)
	+	Báthori <i>et al.</i> (1988)
	+	Raynor <i>et al.</i> (1989)
	+	Wilson <i>et al.</i> (1990)
	+	Large <i>et al.</i> (1992)
	+	Saatov <i>et al.</i> (1993)
	+	Davis <i>et al.</i> (1993)
	+	Báthori <i>et al.</i> (1995)
	+	Girault <i>et al.</i> (1996)
	+	Báthori <i>et al.</i> (1997)
	+	Zibareva <i>et al.</i> (1997)
	+	Báthori (1998)
	+	Wilson <i>et al.</i> (1998)
	+	Wilson & Morden (1999)
	+	Wilson <i>et al.</i> (1999)
	+	Báthori <i>et al.</i> (1999)
	+	Zibareva (2000)
	+	Báthori <i>et al.</i> (2000b)
	+	Wilson (2000)
	+	Báthori & Kalász (2001)
	+	Louden <i>et al.</i> (2002)
	+	Báthori <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
	+	Zibareva <i>et al.</i> (2007a)

	+	Seliverstova <i>et al.</i> (2014)
	+	Dinan <i>et al.</i> (2020b)
<i>S. otites ssp. hungarica</i>	+	Báthori <i>et al.</i> (1987)
	+	Zibareva <i>et al.</i> (2003)
<i>S. otites ssp. parviflora</i>	+	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. paradoxa</i>	+	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. parnassica</i>	+	Zibareva <i>et al.</i> (1997)
	+/-	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. parviflora</i>	-	Agabekova <i>et al.</i> (2010)
<i>S. patula</i>	+	Zibareva (2000)
<i>S. pendula</i>	-	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
<i>S. petraea</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. polaris</i>	-	Zibareva (1997)
<i>S. popovii</i>	+	Yusupova <i>et al.</i> (2020a)
<i>S. portensis</i>	+	Zibareva (1995)
	+	Zibareva (2000)
	+	Meng <i>et al.</i> (2001d)
	+	Zibareva <i>et al.</i> (2003)
	+	Mamadaliyeva <i>et al.</i> (2004b)
	+	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. praemixta</i>	+	Abubakirov (1982)
	+	Saatov <i>et al.</i> (1979a)
	+	Saatov <i>et al.</i> (1979b)
	+	Abubakirov (1980)
	+	Abubakirov (1984)
	+	Saatov <i>et al.</i> (1985)
	+	Saatov <i>et al.</i> (1993)
	+	Agzamovna <i>et al.</i> (2014)
	+	Mamedov <i>et al.</i> (2017)
<i>S. procumbens</i>	-	Zibareva (2000)
<i>S. pseudotites</i>	+	Zibareva (2000)
	+	Zibareva (1999)
	+	Meng <i>et al.</i> (2001d)
	+	Mamadaliyeva <i>et al.</i> (2004b)
	+	Zibareva <i>et al.</i> (2007a)
	+	Seliverstova <i>et al.</i> (2014)
	+	Mamedov <i>et al.</i> (2017)
<i>S. pseudovelutina</i>	+	Zibareva (2000)
	+	Zibareva (1999)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. pusilla</i>	-	Zibareva (2000)
	+	Dinan <i>et al.</i> (2001d)
	+	Zibareva <i>et al.</i> (2003)
	-	Dinan <i>et al.</i> (2020b)
<i>S. pygmaea</i>	+	Zibareva (1999)
	+	Báthori <i>et al.</i> (1995)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. radicata</i>	+	Zibareva (2000)
	+	Meng <i>et al.</i> (2001d)
	+	Mamadaliyeva <i>et al.</i> (2004b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. regia</i>	+	Zibareva <i>et al.</i> (2003)
	+	Meng <i>et al.</i> (2001d)
	+	Mamadaliyeva <i>et al.</i> (2004b)

<i>S. reichenbachii</i>	+	Zibareva (2000)
<i>S. repens</i>	+	Saatov <i>et al.</i> (1993)
	+	Revina <i>et al.</i> (1988)
	+	Zibareva (2000)
	+	Munkhjargal (2013)
	+	Novozhilova <i>et al.</i> (2014)
	+	Novozhilova <i>et al.</i> (2015)
	+	Zibareva <i>et al.</i> (2009)
	+	Olennikov & Kashchenko (2018)
	+	Kashchenko & Olennikov (2019)
<i>S. requienii</i>	-	Zibareva (2000)
	-	Pongracz <i>et al.</i> (2003b)
<i>S. roemeri</i>	+	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007a)
	+	Seliverstova <i>et al.</i> (2014)
<i>S. rubella</i>	+	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. rupestris</i>	-	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
<i>S. samojedorum</i>	(+)	Olennikov & Kashchenko (2018)
<i>S. saxatilis</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. saxifraga</i>	+	Zibareva <i>et al.</i> (2003)
	-	Pongracz <i>et al.</i> (2003b)
	+	Zibareva <i>et al.</i> (2007a)
	+	Dinan <i>et al.</i> (2020b)
<i>S. scabrifolia</i>	+	Saatov <i>et al.</i> (1986a)
	+	Saatov <i>et al.</i> (1986c)
	+	Saatov <i>et al.</i> (1987a)
	+	Saatov <i>et al.</i> (1987b)
	+	Saatov <i>et al.</i> (1990)
	+	Saatov <i>et al.</i> (1993)
	+	Saatov <i>et al.</i> (1999)
	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. schafta</i>	+	Saatov <i>et al.</i> (1993)
	+	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
	+	Dinan <i>et al.</i> (2020b)
<i>S. schimperiana</i>	+	Hussein <i>et al.</i> (2020)
<i>S. schumacheri</i>	+	Bathori <i>et al.</i> (1995)
<i>S. schmuckeri</i>	+	Zibareva (2000)
	+	Zibareva (1999)
	-	Pongracz <i>et al.</i> (2003b)
	+	Zibareva <i>et al.</i> (2007a)
	+	Zibareva <i>et al.</i> (2009)
<i>S. schwarzenbergeri</i>	-	Zibareva (2000)
<i>S. secundiflora</i>	+	Zibareva & Yeryomina (1996)
	+	Zibareva (1997)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. sendtneri</i>	+	Zibareva (1995)
	+	Bathori <i>et al.</i> (1995)
	+	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
	+	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
	+	Zibareva <i>et al.</i> (2009)
	+	Seliverstova <i>et al.</i> (2014)
<i>S. sericea</i>	+	Zibareva <i>et al.</i> (2007a)

<i>S. sibirica</i>	+	Olennikov & Kashchenko (2020)
<i>S. sieberi</i>	+	Zibareva (2000)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. sobolevskajae</i>	+	Saatov <i>et al.</i> (1993)
	+	Revina <i>et al.</i> (1988)
	+	Ramazanov <i>et al.</i> (1997c)
<i>S. spergulifolia</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. squamigera</i>	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007b)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. stenophylla</i>	+	Zibareva <i>et al.</i> (2007a)
	+	Novozhilova <i>et al.</i> (2014)
	+	Novozhilova <i>et al.</i> (2015)
<i>S. stylosa</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. succulenta</i>	-	Zibareva (2000)
<i>S. suecica</i>	-	Dinan <i>et al.</i> (2020b)
<i>S. supina</i>	+	Ramazanov <i>et al.</i> (1997c)
<i>S. sussamyrica</i>	+	Zibareva <i>et al.</i> (2007a)
<i>S. tatarica</i>	+	Girault <i>et al.</i> (1990)
	+	Baltaev <i>et al.</i> (1987)
	+	Saatov <i>et al.</i> (1993)
	+	Báthori <i>et al.</i> (1995)
	+	Zibareva & Yeryomina (1996)
	+	Báthori & Máthé (1996)
	+	Zibareva (1997)
	+	Baltayev (1998)
	+	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
	+	Ramazanov <i>et al.</i> (2007)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. tatarinowii</i>	+	Liang <i>et al.</i> (2019)
	+	Zhang <i>et al.</i> (2020)
<i>S. thessalonica</i>	+	Zibareva <i>et al.</i> (1997)
	+	Zibareva <i>et al.</i> (2003)
	+	Zibareva <i>et al.</i> (2007a)
<i>S. tomentella</i>	+	Ramazanov <i>et al.</i> (1995)
	+	Ramazanov <i>et al.</i> (1996)
	+	Ramazanov <i>et al.</i> (2007)
<i>S. turgida</i>	-	Revina <i>et al.</i> (1988)
<i>S. undulata</i>	+	Zibareva <i>et al.</i> (2003)
<i>S. uniflora</i>	-	Zibareva <i>et al.</i> (2003)
<i>S. uralensis</i>	+	Zibareva <i>et al.</i> (2007a)
	+	Olennikov & Kashchenko (2018)
<i>S. vallesia</i>	-	Zibareva (2000)
	-	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
<i>S. violascens</i>	+	Olennikov & Kashchenko (2018)
<i>S. virginica</i>	+	Zibareva <i>et al.</i> (2003)
<i>S. viridiflora</i>	+	Zibareva <i>et al.</i> (1997)
	+	Zibareva (1995)
	+	Zibareva (1997)
	+	Ramazanov <i>et al.</i> (1997c)
	+	Zibareva <i>et al.</i> (2003)
	+	Pongracz <i>et al.</i> (2003b)
	+	Mamadaliyeva <i>et al.</i> (2003)
	+	Mamadaliyeva <i>et al.</i> (2004a)
	+	Mamadaliyeva <i>et al.</i> (2004b)
	+	Ramazanov <i>et al.</i> (2007)
	+	Tóth and Bathori (2008)
	+	Zibareva <i>et al.</i> (2007a)

			+	Tóth <i>et al.</i> (2008)
			+	Simon <i>et al.</i> (2009)
			+	Tóth (2010)
			+	Mamadalieva <i>et al.</i> (2010)
			+	Mamedov <i>et al.</i> (2017)
			+	Mamadalieva <i>et al.</i> (2019)
			+	Dinan <i>et al.</i> (2020b)
<i>S. viscaria</i>			-	Dinan <i>et al.</i> (2020b)
<i>S. viscidula</i>			+	Jin <i>et al.</i> (2011)
<i>S. viscosa</i>			-	Volodin <i>et al.</i> (2002)
<i>S. vulgaris</i>			-	Zibareva (1997)
			+	Shivakumar <i>et al.</i> (1995)
			-	Zibareva (2000)
			-	Zibareva <i>et al.</i> (2003)
			+	Pongracz <i>et al.</i> (2003b)
			+	Sidana <i>et al.</i> (2017)
			-	Olennikov & Kashchenko (2018)
<i>S. waldsteinii</i>			-	Zibareva (2000)
			-	Dinan <i>et al.</i> (2020b)
<i>S. wallichiana</i>			+	Abubakirov (1982)
			+	Abubakirov (1984)
			+	Saatov <i>et al.</i> (1987c)
			+	Saatov <i>et al.</i> (1988)
			+	Saatov <i>et al.</i> (1993)
			+	Báthori <i>et al.</i> (1995)
			+	Saatov <i>et al.</i> (1999)
			+	Mamadalieva <i>et al.</i> (2000)
			+	Mamadalieva <i>et al.</i> (2002a)
			+	Ramazanov <i>et al.</i> (2007)
			+	Mamedov <i>et al.</i> (2017)
<i>S. wolgensis</i>			+	Zibareva <i>et al.</i> (2007a)
			+	Bespayeva <i>et al.</i> (2012)
			+	Temirgazivev <i>et al.</i> (2016)
			+	Temirgazivev <i>et al.</i> (2018)
			+	Kozhanova <i>et al.</i> (2020)
<i>S. zawadzki</i>			-	Zibareva (2000)
			+	Báthori <i>et al.</i> (1995)
			-	Zibareva <i>et al.</i> (2003)
			+	Pongracz <i>et al.</i> (2003b)
			-	Dinan <i>et al.</i> (2020b)
<i>Silphium perfoliatum</i>	Compositae (D) [Asteraceae (E)]	(+)		Volodin <i>et al.</i> (1993)
<i>Silybum marianum</i>	Compositae (D) [Asteraceae (E)]	(+)		Volodin <i>et al.</i> (1993)
		-		Blackford & Dinan (1997a)
<i>Sinapis alba</i>	Cruciferae (D) [Brassicaceae (E)]	-		Dinan <i>et al.</i> (2020a)
<i>Siphonostegia chinensis</i>	Scrophulariaceae (D) [Orobanchaceae (E)]	2.0		Wong <i>et al.</i> (1979)
<i>Solanum aculeatissimum</i>	Solanaceae (D)	-/(+)		Savchenko <i>et al.</i> (2000)
<i>S. armatum</i>	[Solanaceae (E)]	(+)		Savchenko <i>et al.</i> (2000)
<i>S. atropurpureum</i>		-		Savchenko <i>et al.</i> (2000)
<i>S. aviculare</i>		-		Savchenko <i>et al.</i> (2000)
<i>S. capsicastrum</i>		-/(+)		Savchenko <i>et al.</i> (2000)
<i>S. carolinense</i>		-		Savchenko <i>et al.</i> (2000)
<i>S. centrale</i>		-		Savchenko <i>et al.</i> (2000)

<i>S. diversifolium</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. dulcamara</i>		+	Matsuoka <i>et al.</i> (1969)
		-	Blackford & Dinan (1997c)
		-	Savchenko <i>et al.</i> (2000)
		-	Volodin <i>et al.</i> (2002)
<i>S. fraxinifolium</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. gabriellae</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. giganteum</i>		(+)	Savchenko <i>et al.</i> (2000)
<i>S. hendersonii</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. heterodoxum</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. hindsianum</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. horridum</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. indicum</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. integrifolium</i>		-/(+)	Savchenko <i>et al.</i> (2000)
<i>S. khasianum</i>		-/(+)	Savchenko <i>et al.</i> (2000)
<i>S. laciniatum</i>		-/(+)	Savchenko <i>et al.</i> (2000)
<i>S. lasiophyllum</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. luteum</i>		(+)	Savchenko <i>et al.</i> (2000)
<i>S. mammosum</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. mauritanum</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. melanocerasum</i>		+	Savchenko <i>et al.</i> (2000)
		-	Dinan <i>et al.</i> (2020b)
<i>S. melongena</i>		(+)	Blackford <i>et al.</i> (1996)
		(+)	Blackford & Dinan (1997c)
		-/(+)	Savchenko <i>et al.</i> (2000)
		-	Dinan <i>et al.</i> (2020a)
<i>S. nigrum</i>		+	Savchenko <i>et al.</i> (2000)
		-	Volodin <i>et al.</i> (2002)
<i>S. petrophilum</i>		-/(+)	Savchenko <i>et al.</i> (2000)
<i>S. phlomoides</i>		(+)	Savchenko <i>et al.</i> (2000)
<i>S. pseudocapsicum</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. quitoense</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. rigescens</i>		(+)	Savchenko <i>et al.</i> (2000)
<i>S. seaforthianum</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. simile</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. sisymbriifolium</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. sodomeum</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. sturtianum</i>		(+)	Savchenko <i>et al.</i> (2000)
<i>S. symonii</i>		(+)	Savchenko <i>et al.</i> (2000)
<i>S. topiro</i>		-	Savchenko <i>et al.</i> (2000)
<i>S. torvum</i>		(+)	Savchenko <i>et al.</i> (2000)
<i>S. tuberosum</i>		-	Blackford & Dinan (1997b)
		-	Blackford & Dinan (1997c)
		-	Dinan <i>et al.</i> (2020a)
<i>S. villosum</i>		-	Dinan <i>et al.</i> (2020b)
<i>S. xanthocarpum</i>		?	Tsay <i>et al.</i> (1970)
		?	Kusano <i>et al.</i> (1975)
		(+)	Savchenko <i>et al.</i> (2000)
<i>S. xantii</i>		-	Savchenko <i>et al.</i> (2000)
<i>Solidago decurens</i>	Compositae (D)	2.2	Wong <i>et al.</i> (1979)
<i>S. virgaurea</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Sonchus arvensis</i>	Compositae (D)	(+)	Volodin <i>et al.</i> (1993)
	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>S. palustris</i>		-	Dinan <i>et al.</i> (2001d)
<i>Sophora japonica</i>	Leguminosae-P. (D)	+	Matsuoka <i>et al.</i> (1969)
	[Fabaceae (E)]		

<i>Sorbus huphenis</i>	Rosaceae (D)	-	Dinan <i>et al.</i> (2001d)
<i>S. munda</i>	[Rosaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Sorghum bicolor</i>	Gramineae (M)	-	Blackford <i>et al.</i> (1996)
	[Poaceae (M)]		
<i>Sparganium emersum</i>	Typhaceae (M)	-	Volodin <i>et al.</i> (2002)
<i>S. hyperboreum</i>	[Typhaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Spergula arvensis</i>	Caryophyllaceae (D)	+	Volodin <i>et al.</i> (2002)
	[Caryophyllaceae (E)]		
<i>Spergularia campestris</i>	Caryophyllaceae (D)	+	Agabekova <i>et al.</i> (2010)
<i>S. media</i>	[Caryophyllaceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
		-	Zibareva <i>et al.</i> (2003)
<i>S. rubra</i>		-	Volodin <i>et al.</i> (2002)
		-	Zibareva <i>et al.</i> (2003)
<i>Sphenocentrum jollyanum</i>	Menispermaceae (D)	+	Ajayi <i>et al.</i> (2019)
	[Menispermaceae (E)]	+	Akinwumi <i>et al.</i> (2020)
<i>Sphenomeris (Odontosoria) chinensis</i>	Dennstaedtiaceae (F)	-	Hikino <i>et al.</i> (1973)
<i>S. chusana (S. chinensis)</i>	[Lindsaeaceae (F)]	-	Yen <i>et al.</i> (1974)
<i>S. chinensis</i> var. <i>littoralis</i>		+	Hikino <i>et al.</i> (1973)
<i>Spinacia oleracea</i>	Chenopodiaceae (D)	+	Báthory <i>et al.</i> (1982)
	[Amaranthaceae (E)]	+	Báthory <i>et al.</i> (1984)
		+	Báthory <i>et al.</i> (1986b)
		+	Báthory <i>et al.</i> (1987)
		+	Dinan <i>et al.</i> (1991)
		+	Grebenok & Adler (1991)
		+	Grebenok <i>et al.</i> (1991)
		+	Gebenok & Adler (1993)
		+	Grebenok <i>et al.</i> (1994)
		+	Dinan (1995b)
		+	Adler & Grebenok (1995)
		+	Grebenok <i>et al.</i> (1996)
		+	Blackford & Dinan (1997b)
		+	Schmelz <i>et al.</i> (1998)
		+	Schmelz <i>et al.</i> (1999)
		+	Dinan <i>et al.</i> (1998)
		+	Schmelz (1999)
		+	Schmelz <i>et al.</i> (2000)
		+	Schmelz <i>et al.</i> (2002)
		+	Saeng-ngam <i>et al.</i> (on-line)
		+	Bakrim <i>et al.</i> (2008)
		+	Cheng <i>et al.</i> (2010)
		+	Cao <i>et al.</i> (2016)
		+	Muchate <i>et al.</i> (2018)
		+	Muchate <i>et al.</i> (2019)
		+	Dinan <i>et al.</i> (2020a)
		+	Gorelick <i>et al.</i> (2020)
		+	Bajkacz <i>et al.</i> (2020)
		+	Bokov <i>et al.</i> (2020)
		+	Chen & Feng (2021)
<i>S. tetrandra</i>		+	Dinan <i>et al.</i> (1998)
<i>Spirea media</i>	Rosaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Rosaceae (E)]		

<i>Stachys annua</i>	Labiatae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>S. betonica</i>	[Lamiaceae (E)]	+	Matsuoka <i>et al.</i> (1969)
<i>S. byzantina</i>		(+)	Dinan <i>et al.</i> (2001d)
<i>S. hissarica</i>		+	Ramazanov <i>et al.</i> (2017)
<i>S. macrantha</i>		-	Dinan <i>et al.</i> (2020b)
<i>S. palustris</i>		-	Volodin <i>et al.</i> (2002)
<i>Stachyurus himalaicus</i> var <i>himalaicus</i>	Stachyuraceae (D)	+	Wang <i>et al.</i> (2006)
<i>S. praecox</i>	[Stachyuraceae (E)]	+	Imai <i>et al.</i> (1969c)
		+	Imai <i>et al.</i> (1969d)
		+	Imai <i>et al.</i> (1970a)
		+	Imai <i>et al.</i> (1970b)
		+	Matsuoka <i>et al.</i> (1969)
<i>S. praecox</i> var. <i>matsuzakii</i>		+	Matsuoka <i>et al.</i> (1969)
<i>Stellaria alsinoides</i>	Caryophyllaceae (D)	-	Agabekova <i>et al.</i> (2010)
<i>S. bungeana</i>	[Caryophyllaceae (E)]	-	Revina <i>et al.</i> (1988)
<i>S. crassifolia</i>		-	Volodin <i>et al.</i> (2002)
<i>S. hebecalyx</i>		-	Volodin <i>et al.</i> (2002)
<i>S. holostea</i>		-	Revina <i>et al.</i> (1988)
		-	Volodin <i>et al.</i> (2002)
		-	Zibareva <i>et al.</i> (2003)
<i>S. media</i>		-	Zibareva <i>et al.</i> (2003)
<i>S. nemorum</i>		-	Volodin <i>et al.</i> (2002)
<i>S. palustris</i>		-	Volodin <i>et al.</i> (2002)
<i>S. umbellata</i>		-	Revina <i>et al.</i> (1988)
<i>Stellaria</i> sp.		-	Volodin <i>et al.</i> (2002)
<i>Stemmacantha carthamoides</i>	Compositae (D)	+	Sun <i>et al.</i> (2012)
<i>S. uniflora</i> ssp. <i>satzyperovii</i>	[Asteraceae (E)]	+	Zarembo <i>et al.</i> (2003)
(syn. <i>Rhaponticum uniflorum</i> ssp. <i>satzyperovii</i> , <i>Rh. satzyperovii</i>)		+	Vorob'eva <i>et al.</i> (2006)
<i>Stenochlaena palustris</i>	Blechnaceae (F)	+	Bergamasco & Horn (1983)
	[Blechnaceae (F)]	+	Pardede <i>et al.</i> (2018)
<i>Stephania japonica</i>	Menispermaceae (D)	-	Sreejit (2014)
<i>S. wightii</i>	[Menispermaceae (E)]	-	Sreejit (2014)
<i>Steris viscaria</i>	Caryophyllaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Caryophyllaceae (E)]		
<i>Stipa arundinacea</i>	Gramineae (M)	-	Dinan (1995a)
<i>S. barbata</i>	[Poaceae (M)]	-	Dinan (1995a)
<i>S. calamagrostis</i>		-	Dinan <i>et al.</i> (2020b)
<i>S. gigantea</i>		-	Dinan (1995a)
<i>S. lessingiana</i>		-	Dinan <i>et al.</i> (2020b)
<i>S. pinnata</i>		-	Dinan (1995a)
<i>S. tenacissima</i>		-	Dinan (1995a)
<i>Stizolophus (Centaurea) balsamita</i>	Compositae (D)	+	Nawrot <i>et al.</i> (2021)
	[Asteraceae (E)]		
<i>Stratiotes aloides</i>	Hydrocharitaceae (M)	(+)	Volodin <i>et al.</i> (2002)
	[Hydrocharitaceae (M)]		
<i>Strobilanthes dalzielli</i>	Acanthaceae (D)	(+)	Volodin <i>et al.</i> (2018)
<i>S. lilacinus</i>	[Acanthaceae (E)]	(+)	Volodin <i>et al.</i> (2018)
<i>S. multangulus</i>		traces	Volodin <i>et al.</i> (2018)
<i>S. oligantha</i>		+	Matsuoka <i>et al.</i> (1969)

<i>Struthiopteris castanea</i>	Blechnaceae/Woodsiaceae/Osmundaceae?(F)+		Imai <i>et al.</i> (1969d)
<i>S. niponica</i>	[Blechnaceae (F)]+		Imai <i>et al.</i> (1969c)
		+	Imai <i>et al.</i> (1969d)
<i>Suaeda aegyptiaca</i>	Chenopodiaceae (D)	-	Dinan <i>et al.</i> (1998)
<i>S. maritima</i>	[Amaranthaceae (E)]	-	Báthory <i>et al.</i> (1984)
		-	Clément & Dinan (1991)
		-	Dinan <i>et al.</i> (1991)
		-	Dinan <i>et al.</i> (1998)
<i>S. pannonica</i>		-	Báthory <i>et al.</i> (1984)
<i>S. vera</i>		-	Dinan <i>et al.</i> (1998)
<i>S. vermiculata</i>		-	Dinan <i>et al.</i> (1998)
<i>Swertia kingii</i>	Gentianaceae (D)	-	Dinan <i>et al.</i> (2020b)
	[Gentianaceae (E)]		
<i>Swida alba</i>	Cornaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Cornaceae (E)]		
<i>Symphoricarpos rivularis</i>	Caprifoliaceae (D)	-	Blackford & Dinan (1997c)
	[Caprifoliaceae (E)]		
<i>Symphytum asperum</i>	Boraginaceae (D)	(+)	Volodin <i>et al.</i> (2002)
<i>S. caucasicum</i>	[Boraginaceae (E)]	(+)	Volodin <i>et al.</i> (2002)
<i>S. officinale</i>		+	Takemoto <i>et al.</i> (1967c)
		-	Volodin <i>et al.</i> (2002)
<i>S. tanaicense</i>		+	Volodin <i>et al.</i> (2002)
<i>Syringa vulgaris</i>	Oleaceae (D)	-	Blackford & Dinan (1997c)
	[Oleaceae (E)]		
T			
<i>Talinum calcynum</i>	Portulacaceae (D)	-	Dinan <i>et al.</i> (2020b)
	[Talinaceae (E)]		
<i>Tamarindus indica</i>	Leguminosae-C. (D)	-	Dinan <i>et al.</i> (2020a)
	[Fabaceae (E)]		
<i>Taraxacum officinale</i>	Compositae (D)	(+)	Volodin <i>et al.</i> (1993)
	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Taxus baccata</i>	Taxaceae (G)	+	Staal (1967)
	[Taxaceae (G)]	+	Takemoto <i>et al.</i> (1967d)
		+	Hoffmeister <i>et al.</i> (1967)
		+	de Souza <i>et al.</i> (1969)
		+	Cook <i>et al.</i> (1973)
		+	Lloyd-Jones <i>et al.</i> (1973)
		+	Dreier (1987)
		+	Chakraborty & Bordoloi (2003)
<i>T. brevifolia</i>		+	Dreier (1987)
<i>T. canadensis</i>		+	Staal (1967)
		+	Dreier (1987)
		+	Fang <i>et al.</i> (2010)
<i>T. cuspidata</i>		+	Burns & Gilgan (1977)
		+	Staal (1967)
		+	Imai <i>et al.</i> (1967)
		+	Imai <i>et al.</i> (1969d)
		+	Ripa <i>et al.</i> (1990)
		+	Matsuoka <i>et al.</i> (1969)

		+	Nakano <i>et al.</i> (1982)
		+	Shi <i>et al.</i> (2007)
		+	Guo <i>et al.</i> (2011)
<i>T. cuspidata</i> var. <i>nana</i>		+	Takemoto <i>et al.</i> (1968f)
		+	Matsuoka <i>et al.</i> (1969)
		+	Imai <i>et al.</i> (1969d)
		+	Takemoto <i>et al.</i> (1967c)
<i>T. floridana</i>		+	Rao & Johnson (1998)
<i>T. mairei</i>		+	Zhang <i>et al.</i> (2007)
<i>T. wallichiana</i>		+	Staal (1967)
		+	Rufaie <i>et al.</i> (2011)
<i>T. yunnanensis</i>		+	Li <i>et al.</i> (2002)
<i>Tectaria</i> sp.	Dryopteridaceae (F)	+	Bergamasco & Horn (1983)
<i>Tectaria kwarensensis</i>	[Tectariaceae (F)]	-	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
<i>T. subtriphylla</i>		+	Yen <i>et al.</i> (1974)
<i>Telekia speciosa</i>	Compositae (D) [Asteraceae (E)]	(+)	Dinan <i>et al.</i> (2001d)
<i>Tephrosieris integrifolia</i>	Compositae (D)	-	Volodin <i>et al.</i> (2002)
<i>T. palustris</i>	[Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Terminalia chebula</i>	Combretaceae (D) [Combretaceae (E)]	-	Sreejit & Nelshi (2019)
<i>Tetragonia tetragonoides</i>	Aizoaceae (D) [Aizoaceae (E)]	-	Takemoto <i>et al.</i> (1967c)
		-	Báthori <i>et al.</i> (1987)
<i>Thalictrum aquilegifolium</i>	Ranunculaceae (D)	-	Dinan <i>et al.</i> (2002a)
<i>T. diptercarpum</i>	[Ranunculaceae (E)]	-	Dinan <i>et al.</i> (2002a)
<i>T. flavum</i>		(+)	Dinan <i>et al.</i> (2001d)
		(+)	Dinan <i>et al.</i> (2002a)
<i>T. minus</i>		-	Dinan <i>et al.</i> (2002a)
		+	Volodin <i>et al.</i> (2002)
<i>T. revolutum</i>		-	Dinan <i>et al.</i> (2002a)
<i>T. simplex</i>		-	Volodin <i>et al.</i> (2002)
<i>T. speciosissimum</i>		(+)	Dinan <i>et al.</i> (2002a)
<i>Thelypteris pennigera</i>	Thelypteridaceae (F)	+	Russell & Fenemore (1971)
<i>T. phegopteris</i>	[Thelypteridaceae (F)]	-	Dreier (1987)
<i>Theobroma cacao</i>	Sterculiaceae (D) [Malvaceae (E)]	(+)	Blackford <i>et al.</i> (1996)
<i>Thlaspi arvense</i>	Cruciferae (D) [Brassicaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Threlkeldia diffusa</i>	Chenopodiaceae (D) [Amaranthaceae (E)]	-	Dinan <i>et al.</i> (1998)
<i>Thuja occidentalis</i>	Cupressaceae (G) [Cupressaceae (G)]	-	Hoffmeister <i>et al.</i> (1967)
<i>Thujopsis dolabrata</i>	Cupressaceae (G) [Cupressaceae (G)]	+	Takemoto <i>et al.</i> (1967c)
<i>Thyselium palustre</i>	Umbelliferae (D) [Apiaceae (E)]	-	Volodin <i>et al.</i> (2002)

<i>Tilia migueliana</i>	Tiliaceae (D) [Malvaceae (E)]	1.8	Wong <i>et al.</i> (1979)
<i>Tiliacora acuminata</i>	Menispermaceae (D) [Menispermaceae (E)]	-	Sreejit (2014)
<i>Tinospora capillipes</i>	Menispermaceae (D) [Menispermaceae (E)]	+	Song & Xu (1991)
<i>T. cordifolia</i>		+	Shi <i>et al.</i> (2006)
		+	Pathak <i>et al.</i> (1995)
		+	Gangan <i>et al.</i> (1997)
		+	Pradhan <i>et al.</i> (1997)
		+	Singh <i>et al.</i> (2003)
		+	Sreejit (2014)
		+	Kumar (2014)
		+	Bajpai <i>et al.</i> (2015)
		+	Abiramasundari <i>et al.</i> (2017)
		+	Sreejit <i>et al.</i> (2018)
		+	Sharma <i>et al.</i> (2019)
<i>T. craveniana</i>		+	Li <i>et al.</i> (2005)
<i>T. hainanensis</i>		+	Guo <i>et al.</i> (1999)
<i>T. sagittata</i>		+	Wang <i>et al.</i> (2011)
<i>T. sagittata</i> var. <i>yunnanensis</i>		+	Cheng <i>et al.</i> (2010)
<i>T. sinensis</i>		-	Sreejit (2014)
<i>Todea barbara</i>	Osmundaceae (F)	+	Russell & Fenemore (1971)
<i>T. superba</i>	[Osmundaceae (F)]	-	Russell & Fenemore (1971)
<i>T. hymenophylloides</i>		+	Russell & Fenemore (1971)
<i>Torreya californica</i>	Taxaceae (G)	-	Staal (1967)
<i>T. grandis</i>	[Taxaceae (G)]	-	Staal (1967)
<i>T. nucifera</i>		+	Imai <i>et al.</i> (1967)
		+	Staal (1967)
		+	Imai <i>et al.</i> (1969d)
		+	Takemoto <i>et al.</i> (1967c)
		+	Matsuoka <i>et al.</i> (1969)
<i>Trachelospermum asiaticum</i>	Apocynaceae (D) [Apocynaceae (E)]	+	Matsuoka <i>et al.</i> (1969)
<i>Tradescantia bractea</i>	Commelinaceae (M)	(+)	Crouzet <i>et al.</i> (2009)
<i>T. brevifolia</i>	[Commelinaceae (M)]	(+)	Crouzet <i>et al.</i> (2009)
<i>T. cerinthoides</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>T. commelinoides</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>T. fluminensis</i> (syn. <i>T. albiflora</i>)		(+)	Crouzet <i>et al.</i> (2009)
<i>T. ohioensis</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>T. pallida</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>T. sillamontana</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>T. spatheca</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>T. virginiana</i>		-	Dreier (1987)
<i>T. viridis</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>T. zebrina</i>		(+)	Crouzet <i>et al.</i> (2009)
<i>T. zebrina purpusii</i>		-(+)	Crouzet <i>et al.</i> (2009)
<i>Tragopogon pratensis</i>	Compositae (D) [Asteraceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Trianthema galericulata</i>	Aizoaceae (D)	+	Bergamasco & Horn (1983)
<i>T. pilosa</i>	[Aizoaceae (E)]	+	Sarker <i>et al.</i> (1998b)
<i>T. portulacastrum</i>		+	Bergamasco & Horn (1983)

			+	Banerji <i>et al.</i> (1971)
			+	Ravishankar & Mehta (1979)
			+	Saeng-ngam <i>et al.</i> (on-line)
			-	Sreejit (2014)
			+	Vaiday <i>et al.</i> (2015)
<i>T. turgidifolia</i>			+	Sarker <i>et al.</i> (1998)
<i>Trichomanes elongatum</i>	Hymenophyllaceae (F) [Hymenophyllaceae (F)]		-	Russell & Fenemore (1971)
<i>Trichosanthes cucumerina</i>	Cucurbitaceae (D)		-	Dinan <i>et al.</i> (2020b)
<i>T. tricuspidata</i>	[Cucurbitaceae (E)]		-	Dinan <i>et al.</i> (2020b)
<i>Tridax procumbens</i>	Compositae (D) [Asteraceae (E)]		+	Saeng-ngam <i>et al.</i> (on-line)
<i>Trientalis europea</i>	Primulaceae (D) [Primulaceae (E)]		-	Volodin <i>et al.</i> (2002)
<i>Trifolium repens</i>	Leguminosae-Pap. (D) [Fabaceae (E)]		-	Takemoto <i>et al.</i> (1967c)
<i>Trigonella foenum-graecum</i>	Leguminosae-P. (D) [Fabaceae (E)]		-	Dinan <i>et al.</i> (2020a)
<i>Trillium cernuum</i>	Trilliaceae (M)		+	Dreier (1987)
<i>T. govanianum</i>	[Melanthiaceae (M)]		+	Rahman <i>et al.</i> (2017)
<i>T. kamtchaticum</i>			+	Ono <i>et al.</i> (2007)
			+	Feng <i>et al.</i> (2018)
<i>T. ovatum</i>			+	Dreier (1987)
<i>T. smallii</i>			+	Imai <i>et al.</i> (1969c)
			+	Imai <i>et al.</i> (1969d)
			+	Matsuoka <i>et al.</i> (1969)
<i>T. tschonoskii</i>			+	Matsuoka <i>et al.</i> (1969)
			+	Imai <i>et al.</i> (1969c)
			+	Imai <i>et al.</i> (1969d)
			+	Zhang <i>et al.</i> (2013)
			+	Zhang <i>et al.</i> (2014)
<i>Tripleurospermum perforatum</i>	Compositae (D) [Asteraceae (E)]		-	Volodin <i>et al.</i> (2002)
<i>Tripogandra amplexocaulis</i>	Commelinaceae (M)	(+)		Crouzet <i>et al.</i> (2009)
<i>T. cumanensis</i>	[Commelinaceae (M)]	-/(+)		Crouzet <i>et al.</i> (2009)
<i>Trisetum flavescens</i>	Gramineae (M) [Poaceae (M)]		+	Sarker <i>et al.</i> (1998a)
<i>Triticum aestivum</i>	Gramineae (M) [Poaceae (M)]		-	Dinan (1995a)
			-	Dinan <i>et al.</i> (2020a)
			(+)	Janeczko <i>et al.</i> (2021)
<i>T. durum</i>			-	Dinan (1995a)
<i>Trollius acaulis</i>	Ranunculaceae (D)		-	Dinan <i>et al.</i> (2002a)
<i>T. asiaticus</i>	[Ranunculaceae (E)]		-	Dinan <i>et al.</i> (2002a)
<i>T. chinensis</i>			-	Dinan <i>et al.</i> (2002a)
			-	Dinan <i>et al.</i> (2020b)
<i>T. europaeus</i>			(+)	Dinan <i>et al.</i> (2002a)
			-	Volodin <i>et al.</i> (2002)
			-	Dinan <i>et al.</i> (2020b)

<i>T. hondoensis</i>		-	Dinan <i>et al.</i> (2002a)
<i>T. hybridus</i>		-	Dinan <i>et al.</i> (2002a)
<i>T. ircuticus</i>		-	Dinan <i>et al.</i> (2020b)
<i>T. laxus</i>		-	Dinan <i>et al.</i> (2002a)
		-	Dinan <i>et al.</i> (2020b)
<i>T. ledebourii</i>		-	Dinan <i>et al.</i> (2002a)
<i>T. pumilus</i>		(+)/-	Dinan <i>et al.</i> (2002a)
		-	Dinan <i>et al.</i> (2020b)
<i>T. vaginatus</i>		-	Dinan <i>et al.</i> (2020b)
<i>T. yunnanensis</i>		-	Dinan <i>et al.</i> (2020b)
<i>Tropaeolum majus</i>	Tropaeolaceae (D)	-	Dinan <i>et al.</i> (2020b)
<i>T. minus</i>	[Tropaeolaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>T. polyphyllum</i>		-	Dinan <i>et al.</i> (2020b)
<i>Tulipa sprengeri</i>	Liliaceae (M)	-	Dinan <i>et al.</i> (2001c)
<i>T. turkestanica</i>	[Liliaceae (M)]	-	Dinan <i>et al.</i> (2001c)
<i>T. urumiensis</i>		-	Dinan <i>et al.</i> (2001c)
<i>Tussilago farfara</i>	Compositae (D) [Asteraceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Typha latifolia</i>	Typhaceae (M) [Typhaceae (M)]	-	Volodin <i>et al.</i> (2002)
U			
<i>Ulmus procera</i>	Ulmaceae (D) [Ulmaceae (E)]	-	Blackford & Dinan (1997b)
<i>Umbilicus rupestris</i>	Crassulaceae (D) [Crassulaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Urena lobata</i>	Malvaceae (D) [Malvaceae (E)]	-	Volodin <i>et al.</i> (2018)
<i>Urtica dioica</i>	Urticaceae (D) [Urticaceae (E)]	-	Blackford & Dinan (1997a)
		-	Blackford & Dinan (1997b)
		-	Blackford & Dinan (1997c)
<i>U. sondenii</i>		-	Volodin <i>et al.</i> (2002)
<i>U. urens</i>		-	Blackford & Dinan (1997a)
		-	Blackford & Dinan (1997c)
		-	Volodin <i>et al.</i> (2002)
V			
<i>Vaccaria elisabethae</i>	Caryophyllaceae (D)	-	Zibareva <i>et al.</i> (2003)
<i>V. pyramidalis</i>	[Caryophyllaceae (E)]	-	Zibareva <i>et al.</i> (2003)
<i>Vaccinium macrocarpon</i>	Ericaceae (D)	-	Dinan <i>et al.</i> (2020a)
<i>V. myrtillus</i>	[Ericaceae (E)]	-	Volodin <i>et al.</i> (2002)
		-	Dinan <i>et al.</i> (2020a)
<i>V. uliginosum</i>		-	Volodin <i>et al.</i> (2002)
<i>V. vitis-idaea</i>		-	Volodin <i>et al.</i> (2002)
<i>Valeriana wolgensis</i>	Valerianaceae (D) [Caprifoliaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>Vandenboschia (Crepidomanes) auriculata</i>	Hymenophyllaceae (F) [Hymenophyllaceae (F)]	-	Hikino <i>et al.</i> (1973)
		-	Yen <i>et al.</i> (1974)

<i>V. radicans</i>		-	Hikino <i>et al.</i> (1973)
<i>V. radicans</i> var. <i>naseana</i>		-	Hikino <i>et al.</i> (1973)
<i>Veratrum grandiflorum</i>	Melanthiaceae (M)	+	Matsuoka <i>et al.</i> (1969)
<i>V. lobelianum</i>	[Melanthiaceae (M)]	-	Volodin <i>et al.</i> (2002)
<i>Verbascum blattaria</i>	Scrophulariaceae (D)	-	Blackford & Dinan (1997c)
<i>V. lychnitis</i>	[Scrophulariaceae (E)]	-	Blackford & Dinan (1997c)
<i>V. nigrum</i>		-	Blackford & Dinan (1997c)
		-	Volodin <i>et al.</i> (2002)
<i>V. thapsus</i>		-	Blackford & Dinan (1997c)
		-	Volodin <i>et al.</i> (2002)
<i>Verbena officinalis</i>	Verbenaceae (D)	3.4	Wong <i>et al.</i> (1979)
	[Verbenaceae (E)]		
<i>Vernonia cinerea</i>	Compositae (D)	-	Sreejit & Nelshi (2019)
	[Asteraceae (E)]		
<i>Veronica gentianoides</i>	Scrophulariaceae (D)	(+)	Dinan <i>et al.</i> (2001d)
<i>V. longifolia</i>	[Plantaginaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>V. officinalis</i>		-	Volodin <i>et al.</i> (2002)
<i>V. spicata</i>		-	Dinan <i>et al.</i> (2020b)
<i>Vestia foetida</i>	Solanaceae (D)	-(+)	Savchenko <i>et al.</i> (2000)
<i>V. lycioides</i>	[Solanaceae (E)]	-(+)	Savchenko <i>et al.</i> (2000)
<i>Viburnum opulus</i>	Caprifoliaceae (D)	-	Volodin <i>et al.</i> (2002)
	[Adoxaceae (E)]		
<i>Vicia gramineae</i>	Leguminosae-P. (D)	-	Dinan <i>et al.</i> (2001d)
<i>V. orobus</i>	[Fabaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>V. sylvatica</i>		-	Volodin <i>et al.</i> (2002)
<i>Vigna radiata</i>	Leguminosae-P. (D)	-	Dinan <i>et al.</i> (2020a)
	[Fabaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Viola biflora</i>	Violaceae (D)	-	Volodin <i>et al.</i> (2002)
<i>V. collina</i>	[Violaceae (E)]	-	Volodin <i>et al.</i> (2002)
<i>V. tricolor</i>		-	Volodin <i>et al.</i> (2002)
<i>Viscaria alpina</i>	Caryophyllaceae (D)	-	Zibareva <i>et al.</i> (2003)
	[Caryophyllaceae (E)]		
<i>Vitex agnus-castus</i>	Verbenaceae (D)	2.5	Wong <i>et al.</i> (1979)
	[Lamiaceae (E)]	+	Ramazanov (2004)
<i>V. cannabifolia</i>		2.0	Wong <i>et al.</i> (1979)
<i>V. canescens</i>		+	Suksamrarn <i>et al.</i> (1995)
		+	Suksamrarn <i>et al.</i> (1997)
		+	Suksamrarn <i>et al.</i> (1998)
		+	Suksamrarn <i>et al.</i> (2000)
		+	Laosooksathit <i>et al.</i> (2003)
		+	Zhang <i>et al.</i> (2009)
<i>V. cienkowskii</i>		+	Stark <i>et al.</i> (2020)
<i>V. cymosa</i>		+	dos Santos <i>et al.</i> (2001)
<i>V. doniana</i>		+	Ochieng <i>et al.</i> (2013)
		+	Ishola <i>et al.</i> (2014)
		+	Tijjani <i>et al.</i> (2017)
		+	Bunu <i>et al.</i> (2021)
<i>V. fisherii</i>		+	Kubo <i>et al.</i> (1990)

<i>V. gardneriana</i>		+	Kubo & Nakatsu (1991)
<i>V. glabrata</i>		+	de Sá Barreto <i>et al.</i> (2005)
		+	Werawattanametin <i>et al.</i> (1986)
		+	Suksamrarn <i>et al.</i> (1998)
		+	Suksamrarn <i>et al.</i> (1999)
		+	Sinlapraya <i>et al.</i> (2007a&b)
		+	Thanokeo <i>et al.</i> (2011)
<i>V. leptobotrys</i>		+	Thuy <i>et al.</i> (1998)
		+	Volodin <i>et al.</i> (2018)
<i>V. madiensis</i>		+	Kubo <i>et al.</i> (1984)
<i>V. megapotamica</i>		+	Rimpler & Schultz (1967)
		+	Rimpler (1967)
		+	Rimpler (1969)
		+	Rimpler (1972b)
<i>V. negundo</i>		-	Sreejit (2014)
<i>V. pinnata</i> (syn. <i>V. pubescens</i>)		+	Suksamrarn & Sommechai (1993)
		+	Suksamrarn <i>et al.</i> (1998)
<i>V. polygama</i>		+	dos Santos <i>et al.</i> (2001)
		+	Gallo <i>et al.</i> (2006)
		+	Martins de Jesus <i>et al.</i> (2020)
<i>V. pseudo-negundo</i>		-	Rimpler (1972a)
<i>V. quinata</i>		+	Cheng <i>et al.</i> (2007)
		+	Lu <i>et al.</i> (2009)
		+	Lu <i>et al.</i> (2017)
<i>V. quintata</i>		+	Volodin <i>et al.</i> (2018)
<i>V. rehmanni</i>		+	Rimpler (1972a)
<i>V. scabra</i>		+	Suksamrarn <i>et al.</i> (2002)
<i>V. sereti</i>		+	Rimpler (1972a)
<i>V. strickeri</i>		+	Zhang <i>et al.</i> (1992)
		+	Kubo (1992)
<i>V. stylosa</i>		+	Volodin <i>et al.</i> (2018)
<i>V. thyrsoiflora</i>		+	Kubo <i>et al.</i> (1985)
<i>V. trifolia</i>		+	Nguyen <i>et al.</i> (2018)
<i>V. trifolia</i> var. <i>simplicifolia</i>		0	Wong <i>et al.</i> (1979)
<i>Vitis vinifera</i>	Vitaceae (D) [Vitaceae (E)]	- -	Blackford & Dinan (1997c) Dinan <i>et al.</i> (2020a)
<i>Vittaria anguste-elongata</i>	Vittariaceae (F)	-	Yen <i>et al.</i> (1974)
<i>V. flexuosa</i>	[Pteridaceae (F)]	+/-	Hikino <i>et al.</i> (1973)
<i>V. zosterifolia</i>		+	Hikino <i>et al.</i> (1973)
W			
<i>Weigela florida</i>	Caprifoliaceae (D) [Caprifoliaceae (E)]	-	Dinan <i>et al.</i> (2020b)
<i>Wigandia caracasana</i>	Hydrophyllaceae (D) [Boraginaceae (E)]	-	Dinan <i>et al.</i> (2001d)
<i>Wilcoxia viperina</i>	Cactaceae (D) [Cactaceae (E)]	+/?	Djerassi <i>et al.</i> (1964)
<i>Withania frutescens</i>	Solanaceae (D) [Solanaceae (E)]	+ + -/(+)	Imai <i>et al.</i> (1969d) Matsuoka <i>et al.</i> (1969) Savchenko <i>et al.</i> (2000)
<i>W. somnifera</i>		-/(+)	Savchenko <i>et al.</i> (2000)
<i>Woodsia polystichoides</i>	Woodsiaceae (F) [Woodsiaceae (F)]	-	Hikino <i>et al.</i> (1973)

<i>Woodwardia harlandii</i> var. <i>takeoi</i>	Blechnaceae (F)	+	Hikino <i>et al.</i> (1973)
<i>W. japonica</i>	[Blechnaceae (F)]	+	Hikino <i>et al.</i> (1973)
		+	Yen <i>et al.</i> (1974)
		+	Liu & Gao (2011)
<i>W. orientalis</i>		+	Yen <i>et al.</i> (1974)
		-	Takemoto <i>et al.</i> (1967c)
		+	Hikino <i>et al.</i> (1973)
<i>W. unigemmata</i>		+	Hikino <i>et al.</i> (1973)
<i>Wyethia angustifolia</i>	Compositae (D) [Asteraceae (E)]	-	Dinan <i>et al.</i> (2020b)
X			
<i>Xanthium strumarium</i>	Compositae (D) [Asteraceae (E)]	0.4	Wong <i>et al.</i> (1979)
<i>Xerophyllum tenax</i>	Melanthiaceae (M) [Melanthiaceae (M)]	+	Alison <i>et al.</i> (1997)
Z			
<i>Zanthoxylum avicennae</i>	Rutaceae (D)	+	Volodin <i>et al.</i> (2018)
<i>Z. piperitum</i>	[Rutaceae (E)]	+	Matsuoka <i>et al.</i> (1969)
<i>Z. scabrum</i>		+	Volodin <i>et al.</i> (2018)
<i>Zea japonica</i>	Gramineae (M)	-	Dinan (1995a)
<i>Zea mays</i>	[Poaceae (M)]	-	Devarenne <i>et al.</i> (1995)
		-	Blackford <i>et al.</i> (1996)
<i>Zebrina (Tradescantia) pendula</i>	Commelinaceae (M) [Commelinaceae (M)]	+	Yang <i>et al.</i> (1996)
		-	Dreier (1987)
<i>Zingiber officinale</i>	Zingiberaceae (M) [Zingiberaceae (M)]	-	Dinan <i>et al.</i> (2020a)
<i>Zostera marina</i>	Zosteraceae (M) [Zosteraceae (M)]	-	Volodin <i>et al.</i> (2002)

1b. References for the occurrence of phytoecdysteroids

Where possible, Abstracts are included, especially for publications which are difficult to obtain.

Abdukadirov I.T., Yakubova M.R., Nuriddinov K.R., Mamatkhanov A.U. and Turakhozhaev M.T. (2005) Ecdysterone and turkesterone in *Ajuga turkestanica* determined by HPLC. *Chemistry of Natural Compounds* 41(4), 475-476 [in English]/*Khimiya Prirodnikh Soedinenii* (4), 386-387 (2005) [in Russian].

Abstract: shoots of *Ajuga turkestanica* (Rgl.) Brig. (Labiatae, Mint) are a source of highly effective biologically active compounds such as phytoecdysteroids and natural glycosides [1, 2]. Ecdysterone, cyasterone, ajugalactone, ajugasterone, α -ecdysone, and turkesterone have been isolated previously from *A. turkestanica*. Of these, the main components are ecdysterone and turkesterone, which have very high anabolic activities [1]. The ecdysteroid content, in particular, that of turkesterone, in the raw material was determined by gravimetry [3-5] or UV spectrophotometry [6]. The turkesterone content varied from 0.025 to 0.17, which means that *A. turkestanica* is not an industrial raw material. HPLC is known to determine with high accuracy the content of ecdysteroids in certain plant sources [7-9]. We investigated the contents of turkesterone and ecdysterone in the aerial part of *A. turkestanica* by HPLC on a Zorbax Eclipse XDB-C18 column, 3×150 mm, 3.5 μ m (Agilent Technologies) using an Agilent LC 1100 chromatograph with a four-eluent pump, deaerator, and variable wavelength UV detector. The chromatograph was controlled and the results were processed using Agilent ChemStation software for the liquid chromatograph. UV detection of separate ecdysteroids was carried out at working wavelength 247 nm.

- Abiramasundari G., Mohan Gowda C.M. and Sreepriya M. (2017) Selective estrogen receptor modulator (SERM) and prostimulatory effects of phytoestrogen β -ecdysone in *Tinospora cordifolia* on osteoblast cells. *Journal of Ayurveda and Integrative Medicine* (<http://dx.doi.org/10.1016/j.JAIM.2017.04.003>).
- Abubakirov N.K. (1980) New phytoecdysones. In: *Frontiers in Bioorganic Chemistry and Molecular Biology; Proceedings of an International Conference, 1978*, pp. 257-259.
- Abubakirov N.K. (1982) Ecdysteroids of flowering plants (Angiospermae). *Proceedings of the Indian National Science Academy* **48A** (supplement 1), 122-138.
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- Agabekova A.G. et al. (2010) Guided search of new promising sources of ecdysterone among Kazakhstan plants. *Vestia Karfandinskoro Universiteta* **60**(4), 64-67 [in Russian, with an abstract in English].
- Agzamova M.H., Isaev I.M.O., Mamathanov A.U., Isaev M.I.O. and Ibragimov T.F. (2014) Phytoecdysteroids from *Silene praemixta*. *Advances in Biological Chemistry* **4**, 1-4.
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- Akin S. and Anil H. (2007) A furostanol saponin and phytoecdysteroid from roots of *Helleborus orientalis*. *Chemistry of Natural Compounds* **43**(1), 90-92 [in English]/*Khimiya Prirodnykh Soedinenii* (1), 75-77 (2007) [in Russian].
- Abstract:** A furostanol saponin mixture and a known phytoecdysteroid were isolated from the roots of *Helleborus orientalis* Lam. Their structures were established as 26-[(β -D-glucopyranosyl)oxy]-22 α -hydroxyfurosta-5,25(27)-dien-1 β ,3 β ,11 α -triol (1a), 26-[(β -D-glucopyranosyl)oxy]-22 α -methoxyfurosta-5,25(27)-dien-1 β ,3 β ,11 α -triol (1b), and 20-hydroxy- β -ecdysone-3-O- β -D-glycoside (2). Acid hydrolysis of 1a,b gave (1 β ,3 β ,11 α ,22 α)-22,26-dimethoxyfurosta-5,25(27)-dien-1,3,11-triol (aglycone 1) and of 2 gave 20-hydroxy- β -ecdysone (aglycone 2). Their structures were elucidated by spectral analysis.
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- Akinwumi I.A., Sonibare M.A., Yeye E.O. (2020) Bioassay-guided isolation and identification of anti-ulcer ecdysteroids from the seeds of *Sphenoentrum jollyanum* (Menispermaceae). *Steroids* **159**, 108636.
- Alekseeva L.I. (2004) Ecdysone 20-monooxygenase activity of cytochrome P-450 in *Ajuga reptans* L. plants and cell culture. *Applied Biochemistry and Microbiology* **40**(2), 135-139 [in English]/*Prikladnaya Biokhimiya i Mikrobiologiya* **40**(2), 159-164 [in Russian].
- Abstract:** The concentration of cytochrome P450 and ecdysone 20-monooxygenase activity in plants and callus cell culture of the bugleweed *Ajuga reptans* L. were determined. The maximal ecdysone 20-monooxygenase activity of cytochrome P450 was found in vegetative rosettes of intact plants. During the stage of flowering, the ecdysone 20-monooxygenase activity of cytochrome P450 in plant leaves was higher than in other organs. It was demonstrated that the content of ecdysteroids in callus cell culture is higher than in the intact plant with concurrent retention of a high ecdysone-20-monooxygenase activity.

Alekseeva L.I., Lafont R., Volodin V.V. and Luksha V.G. (1998a) Ecdysteroids from *Ajuga reptans*. Russian Journal of Plant Physiology **45**(3), 316-321 [in English]/Fiziologiya Rastenii **45**(3) 327-377 [in Russian].

Abstract: Ecdysteroids from *Ajuga reptans* plants, grown at the northern limit of the middle taiga subzone of the northeastern region of the European part of Russia, have been identified. For the first time, ajugasteron B was isolated, along with other ecdysteroids characteristic of this species: polypodine B, 20-hydroxyecdysone, 29-norcysterone, 29-norsengosterone, sengosterone, and ajugalactone.

Alexeeva LI, Teter'uk LV, Volodin VV and Kolegova NA (1998b) The dynamics of ecdysteroid content in *Ajuga reptans* L. on the northern border of the Komi region. Rastitelny Resursy (4), 56-61 [in Russian].

Alexeeva L.I., Volodin V.V. and Luksha V.G. (2000) Methods of concentration of hydrophobic ecdysteroids with usage of frontal chromatography. Rastitelny Resursy (4), 122-127 [in Russian, with English abstract].

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Abstract: Several phytoecdysteroids were detected in *Serratula coronata* plants. Among them were 20-hydroxyecdysone, inokosterone, makisterone A, and ecdysone. The highest level of ecdysteroids was found in young, actively growing leaves during the vegetative stage, as well as in leaves and flower stalks during the stage of flower bud formation. The content of ecdysteroids was compared in field-grown plants, plants grown in vitro, and in callus cultures derived from the latter. 20-Hydroxyecdysone, inokosterone, and ecdysone were present in callus cultures; makisterone A was not detected; and an unidentified metabolite, absent from intact plants, was found as a predominant component. The total content of ecdysteroids in the callus tissues, about 0.1-0.2% of dry wt, was higher than that in plants grown in vitro, but lower than in field-grown plants. It was demonstrated that the biosynthetic ability of these callus tissues did not depend on the epigenetic pattern of explants, but was determined by their genotypes.

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Abstract: With the aid of high-performance liquid chromatography, six phytoecdysteroids have been detected in the butanolic fraction of extractive substances from the epigeal part of *Melandrium nutans* L. Preparative separation has yielded ecdysterone and polypodine B.
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Abstract: In the search for plants containing ecdysteroids, we have investigated the East Asian species *Lychnis fulgens* Fisch. from the family Caryophyllaceae, which grows in Primor'e, Priamur'e, in the north of the Korean peninsula, in north-eastern China, and in Japan (islands of Honshu and Hokkaido). Samples of the flowering plants were collected in Primorskii Krai [Maritime Territory] in the environs of the village of Shkotovo in the valley of the R. Shkotovka in July, 1984.
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Abstract: From the roots with rhizomes of the plant *Rhaponticum carthamoides* Willd) Iljin Compositae), in addition to integristerone A, ecdysterone, polypodin B, 2-deoxyecdysterone, and 24(28)-dehydromakisterone A, we have isolated the new compounds ecdysterone 3-2,3-monoacetone (I), ecdysterone 20,22-monoacetone (II) and rhapisterone (III): I — C₃₀H₄₈O₇, mp 232–233° (ethyl acetate-methanol) [α]_D²⁰ +56.4±2° (c 0.0; methanol); II — C₃₀H₄₈O₇, mp 227–229° (ethyl acetate-methanol), [α]_D²⁰ +60.1±2° (c 1.3; methanol); III — C₂₉H₄₈O₇, mp, 241–242° (ethyl acetate-methanol), [α]_D²⁰ +30±2° (c 0.1; dioxane). The structure of (III) was established on the basis of spectral characteristics as 2 β , 3 β , 14 α , 20R, 22R, 23 ζ -5 β -stigmast-7-en-6-one. Details of the PMR, mass, and IR spectra of all the compounds and of the CD of rhapisterone are given.
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Abstract: A new ecdysteroid, which has proved to be ecdysterone 20-O-benzoate, has been isolated from the whole plant *Silene tatarica* (L.) Pers.
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Abstract: A quantitative method has been developed for determination of the 20-hydroxyecdysone content of *Serratula tinctoria* (L.). Thin-layer chromatography on silica was performed in two consecutive steps; the first was with chloroform - ethanol and the separation was completed with chloroform - methanol - benzene. Plant samples of different phenophases were collected in 1994, ecdysteroids were extracted, and, after a clean-up procedure, the 20-hydroxyecdysone content was determined by TLC - densitometry.
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Abstract: Six minor new ecdysteroid components have been isolated from *Silene otites* (L.) Wib. by a combination of chromatographic methods. Three of them (2-deoxy-20-hydroxyecdysone 3,22-diacetate, 5 alpha-2-deoxy-20-hydroxyecdysone 3-acetate, and 2-deoxy-20-hydroxyecdysone 3-crotonate) are new natural products.
- Báthori M., Máthé I., Girault J.-P., Kalász H. and Lafont R. (1998a) Isolation and structural elucidation of two plant ecdysteroids, gerardiasterone and 22-*epi*-20-hydroxyecdysone. *Journal of Natural Products* **61**(3), 415-417.
Abstract: Two minor plant ecdysteroids, 22-*epi*-20-hydroxyecdysone (**1**) and gerardiasterone (**2**), were isolated from *Serratula tinctoria* L. (Compositae). The first compound, a new natural product, was characterized by an unusual stereochemistry at C-22 (i.e., 22*S*). The second compound was identified as (20*R*,23*S*)-20,23-dihydroxyecdysone, a compound previously isolated from the Zooanthid *Gerardia savaglia*.
- Báthori M, Máthé I and Guttman A (1998b) Determination of 20-hydroxyecdysone content by thin-layer chromatography and micellar electrokinetic chromatography. *Chromatographia* **48**(1/2), 145-148.
Abstract: Seasonal dependence of 20-hydroxyecdysone content of *Serratula tinctoria* and *Serratula wolffii* (Asteraceae) was investigated by micellar electrokinetic chromatography (MEKC). Samples were collected each month through the vegetation period. The leaves were dried, milled and extracted with methanol. Clean-up of the extracts was by solid-phase extraction using a polyamide micro-column to remove flavonoids and other plant phenolics which can interfere with the analysis. This work deals with the separation of 20-hydroxyecdysone from

polypodine B and the seasonal variation of 20-hydroxyecdysone concentration. Determinations have been performed by both thin-layer chromatography and capillary electrophoresis using micellar electrokinetic chromatography.

Báthori M., Girault J.-P., Kalász H., Máthé I., Dinan L.N. and Lafont R. (1999) Complex phytoecdysteroid cocktail of *Silene otites* (Caryophyllaceae). Archives of Insect Biochemistry and Physiology 41, 1-8.

Abstract: Several new minor ecdysteroids from *Silene otites* (Caryophyllaceae) have been purified and identified. This plant species had previously been shown to contain a complex ecdysteroid cocktail, with 20-hydroxyecdysone as the major component, and significant amounts of 2-deoxyecdysone, 2-deoxy-20-hydroxyecdysone, and 20-hydroxyecdysone 22-acetate, and a set of minor ecdysteroids. The use of powerful techniques for purification and spectroscopic analyses has now allowed the isolation and identification of more than 30 different molecules including 21-hydroxylated ecdysteroids, thus adding a new position of the carbon skeleton that can be modified in ecdysteroids. Thus, in *S. otites*, a complex array of individual reactions can be used in various combinations leading both to major and minor components. Deciding whether this represents a random process without biological consequences or if (some of) the various minor components may play a specific role in, e.g., insect-plant relationships, will require the extensive use of appropriate in vitro and in vivo bioassays.

Báthori M., Kalász H., Csikkel-Szolnoki A and Máthé I. (1999b) Components of *Serratula* species; screening for ecdysteroid and inorganic constituents of some *Serratula* plants. Acta Pharmaceutica Hungarica 69, 72-76 [in Hungarian, with English abstract].

Abstract: Ecdysteroid and inorganic components were analyzed from several plant species belonging to Caryophyllaceae family, such as in the cases of *Serratula tinctoria*, *Serratula wolffii*, *Serratula coronata* (20-hydroxyecdysone and inorganic components) and *Jurinea mollis*, *Serratula gmelinii* and *Leuzea carthamoides* (inorganic components, only). The 20-hydroxyecdysone content was determined using thin-layer chromatography after a simple clean up that had been performed by solid-phase extraction. Inorganic constituents were determined using either ICP or flame photometry. Vegetation dependence of both 20-hydroxyecdysone and inorganic elements was studied. *Serratula coronata* shows remarkable high 20-hydroxyecdysone (2.3%, in April) and the studied *Serratula* plants gave a minimum of 20-hydroxyecdysone in June. Favorable period for harvesting is suggested as July and August, through the blossoming of these plants. Potassium and 20-hydroxyecdysone content gave a similar tendency considering their vegetation dependence, while the magnesium content moved toward opposite direction. The calcium contents of *Serratula tinctoria*, *Serratula wolffii* and *Serratula coronata* were found between 2.2% and 3.8%, which values are high relative to the other medicinal plants. At the same time, the Cu content of the ecdysteroid producing (and screened) Caryophyllaceae plants is low. The Fe, Mn and Mg contents of *Serratula coronata* are high, even higher than that of the *Leuzea carthamoides*. Our results have suggested the importance of analysis and control of inorganic constituents of crude plant extracts used for medicinal and recreational purposes.

Báthori M, Gergely A, Kalász H, Nagy G, Dobos A, Máthé I (2000a) Liquid chromatographic monitoring of phytoecdysteroid production of *Serratula wolffii*. Journal of Liquid Chromatography & Related Technologies 23(2), 281-294.

Abstract: Seasonal dependence of 20-hydroxyecdysone content of *Serratula wolffii* was monitored. Thin-layer chromatography was used after a single clean-up using polyamide. High performance liquid chromatography was employed for the analysis, after removal of the high excess of flavonoids and other impurities using a clean-up on both C18 and polyamide.

Parallel analyses using both thin layer chromatography with densitometry and high performance liquid chromatography gave similar results at the determination of vegetation dependence of ecdysteroids. Results indicated two maxima of ecdysteroid content (w/w), such as beginning of vegetation (in April and May) and during blossoming (in August).

The two ecdysteroids, 20-hydroxyecdysone and polypodine B were also isolated from *Serratula wolffii*, their chemical structures were identified by spectroscopic methods.

Báthori M., Girault J.-P., Máthé I. and Lafont R. (2000b) Isolation of 5 α - and 5 β -dihydroxirubrosterone from *Silene otites* L. (Wib.). Biomedical Chromatography 14(7), 464-467.

Abstract: 5 alpha-Dihydroxirubrosterone (2 beta, 3 beta, 14 alpha, 17 beta-tetrahydroxy-5 alpha-androst-7-ene-6-one), a new 19-carbon 5 alpha-ecdysteroid, was isolated together with its 5 beta counterpart from the aerial parts of *Silene otites* L. (Wib.) (Caryophyllaceae) by a combination of solvent partition, low-pressure column chromatography, thin-layer chromatography (normal-phase and reversed-phase) and finally HPLC. Mass spectrometry and nuclear magnetic resonance spectroscopic procedures were used for compound characterization.

Báthori M., Blunden G. and Kalász H. (2000c) Two-dimensional thin-layer chromatography of plant ecdysteroids. Chromatographia 52(11/12), 815-817.

Abstract: Two-dimensional thin-layer chromatography has been used to screen plant samples for ecdysteroids. Satisfactory separations were obtained even in the presence of substantial impurities and for mixtures containing many ecdysteroids. Normal-phase silica and octadecyl silica plates were used to differentiate apolar and polar ecdysteroids, respectively. The major advantages of thin-layer chromatography are multiple detection, which enables specific detection of the ecdysteroids. Use of a small amount of water in the mobile phase in the first dimension, the a water-free mobile phase in the second dimension, can afford favorable separations.

Báthori M., Lafont R., Girault J.-P. and Máthé I. (2001) Structural diversity of ecdysteroids of *Lychnis flos-cuculi*. *Acta Pharmaceutica Hungarica* 71, 157-167 [in Hungarian, with an English abstract].

Abstract: Eleven ecdysteroids have been isolated from *Lychnis flos-cuculi*; we are the first who report eight ecdysteroids of the eleven compounds in this plant. Two of these ecdysteroids, dihydrorubrosterone and 20-hydroxyecdysone 3-acetate are newly discovered natural products. The success of isolation of these new ecdysteroids has been based on the use of separation methods in a proper order; these separation procedures were completing each others. At the beginning steps of isolation simple separation methods were used, such a solvent-solvent distribution and fractionated precipitation. Two third of the contaminants were removed thereby. High capacity low resolution methods were used then, such as classical adsorption column chromatography and preparative thin-layer chromatography. The major component (20-hydroxyecdysone) and certain minor ecdysteroids (polypodine B and rubrosterone) were isolated in pure form here. Purification of the further minor components (poststerone, 2-deoxy-20-hydroxyecdysone, vitikosterone E, dihydrorubrosterone, makisterone A, taxisterone, 20-hydroxyecdysone 2-acetate, 20-hydroxyecdysone 3-acetate) required HPLC and other absorption chromatographic methods. Our recent separation scheme means a generally applicable guiding principle for isolation of any plant ecdysteroid, major and minor alike. Structural identification of the known ecdysteroids was based on their spectral data and that of their literature information. Structural elucidation of 20-hydroxyecdysone 3-acetate was done by the help of a standard component prepared by acetylation of 20-hydroxyecdysone. From the mixture of seven acetates the corresponding compound (20-hydroxyecdysone 3-acetate) was isolated, and used for identification. Structural diversity of ecdysteroids of *Lychnis flos-cuculi* is evaluated, and a tentative explanation is introduced for the formation and biosynthesis of the versatility of phytoecdysteroids.

Báthori M., Pongrácz Z., Tóth G., Simon A., Kandra L., Kele Z. and Ohnmacht R. (2002a) Isolation of a new member of the ecdysteroid glycoside family: 2-deoxy-20-hydroxyecdysone 22-O- β -D-glucopyranoside. *Journal of Chromatographic Science* 40(7), 409-415.

Abstract: A new ecdysteroid glycoside, 2-deoxy-20-hydroxyecdysone 22-O- β -D-glucopyranoside, is isolated from the herb *Silene italica* ssp. *nemoralis* (Waldst. and Kit.) Nyman. The compound is purified with multistep chromatography, such as classical column chromatography on alumina and droplet countercurrent distribution. Also, it is expanded using twice low-pressure reversed-phase liquid column chromatography. Chromatography in four steps results in the purified 2-deoxy-20-hydroxyecdysone 22-O- β -D-glucopyranoside. Two other ecdysteroids have also been separated, including the formerly identified integristerone A and 24(28)-dehydromakisterone A.

Báthori M., Kalász H., Pongrácz Z., Máthé I., Kálman A. and Argay G. (2002b) 5 α - and 5 β -2-deoxyintegristerone A, a 5 α and 5 β isomer pair of ecdysteroids isolated from the *Silene* genus. *Biomedical Chromatography* 16(6), 373-378.

Abstract: 5- α -2-deoxyintegristerone A and 5- β -2-deoxyintegristerone A were isolated from the aerial parts of *Silene italica* ssp. *nemoralis* (Waldst. and Kit.) Nyman using a specific combination of absorption column chromatography, preparative thin-layer chromatography and preparative HPLC. Both normal-phase and reversed-phase modes of HPLC were employed for isolation. Structural elucidation of 5- α -2-deoxyintegristerone A was completed by X-ray diffraction. Both 5- α -2-deoxyintegristerone A and 5- β -2-deoxyintegristerone A were firstly isolated from this plant. We propose that 5- α -2-deoxyintegristerone A is not an artifact but an integral part of the ecdysteroid spectrum of *Silene italica* ssp. *nemoralis* (Waldst. and Kit.) Nyman.

Báthori M., Kalász H., Janicsák G., Pongrácz Z. and Vámos J. (2003) Thin-layer chromatography of phytoecdysteroids. *Journal of Liquid Chromatography and Related Technologies* 26(16), 2629-2649.

Abstract: Planar chromatography of ecdysteroids is reviewed. Separation of various ecdysteroids is detailed using both straight-phase and reversed-phase thin-layer chromatography (RP-TLC). The generally used special techniques, such as two-dimensional TLC (2D-TLC), forced-flow TLC (FF-TLC), displacement mode of development, etc., are also specified. The particular behavior of certain ecdysteroids is discussed.

Báthori M., Pongrácz Z., Omacht R. and Máthé I. (2004) Preparative scale purification of shidasterone, 2-deoxy-polypodine B and 9 α ,20-dihydroxyecdysone from *Silene italica* ssp. *nemoralis*. *Journal of Chromatographic Science* 42(5), 275-279.

Abstract: A suitable combination of preparative scale separation methods results in effective clean-up of the ecdysteroids of *Silene italica* ssp. *nemoralis* (Waldst. and Kit.) Nyman. The isolation of minor ecdysteroids from the partially purified extract is based on the use of both droplet counter-current chromatography and low-pressure reversed-phase liquid chromatography. The purification is completed by preparative thin-layer chromatography and preparative high-performance liquid chromatography to obtain the minor ecdysteroids, such as 2-deoxy-20-hydroxyecdysone, shidasterone, 2-deoxy-polypodine B, makisterone C, and 9 α ,20-dihydroxyecdysone.

Báthory M., Tóth I., Szendrei K. and Reisch J. (1982) Ecdysteroids in *Spinacia oleraceae* and *Chenopodium bonus-henricus*. *Phytochemistry* 21, 236-238.

Abstract: The roots of *Chenopodium bonus-henricus* and the seeds of *Spinacia oleracea* contain 20-hydroxyecdysone and polypodine B. The seeds of *S. oleracea* also contain a compound with properties similar to those of 24(28)-dehydromakisterone-A and may contain small amounts of ecdysone.

Báthory M., Tóth I., Szendrei K., Rattai M., Minker E and Blazsó G. (1984) Determination and isolation of ecdysteroids in native goosefoot species. *Herba Hungarica* 23, 131-145 [in Hungarian, with an English abstract].

Báthory M., Szendrei K. and Herke I. (1986) The ecdysteroids of *Silene otites* L. *Wib. Herba Hungarica* 25, 105-117.

Beisler J.A., Tsay Y.-H., Silverton J.V., Beisler J.A. and Sato Y. (1970) The structure of carpesterol. *Journal of the American Chemical Society* 92, 7005-7006 [this letter concerns an ecdysteroid-related compound; no Abstract].

Ben Nejma A., Znati M., Nguir A., Daich A., Othman M., Lawson A.M. and Ben Jannet H. (2017) Phytochemical and biological studies of *Atriplex inflata* f. Muell.: isolation of secondary bioactive metabolites. *Journal of Pharmacy and Pharmacology* 69, 1064-1074.

Abstract:

Objectives: This work describes the phytochemical and biological investigation of the Tunisian *Atriplex inflata* F. Muell (Chenopodiaceae).

Methods: Their chemical structures were elucidated on the basis of extensive spectroscopic methods, including 1D NMR and 2D NMR, ESI-HRMS and comparison with available literature data. The isolates were evaluated for their antioxidant activity by the DPPH[•], ABTS^{•+}, Fe³⁺ and catalase assays and also for their antibacterial and anticholinesterase activity.

Key findings: The chemical study of *Atriplex inflata* F. Muell led to the isolation of two fatty acids (9E)-methyl-8,11,12-trihydroxyoctadec-9-enoate 1 and (9E)-8,11,12-trihydroxyoctadecenoic acid 2 together with (Z)-litchiol B 3 and 20-hydroxyecdysone 4. Three of which are reported here for the first time in *Atriplex* genus. Based on the biosynthesis of hydroxylated arachidonic acid and derivatives, a plausible biogenesis pathway of the two fatty acids (1 and 2) was proposed. (Z)-litchiol B (3) was found to be the most active against *Staphylococcus aureus*. According to the literature, this is the first time that compounds 1, 2 and 3 were tested for their eventual biological activity.

Conclusions: In the results of the present work, we have proposed the biogenesis pathway of unsaturated fatty acid and described the structure-activity relationship.

Beredze M., Kalandia A., Japaridze I., Vanidze M., Varshanidze N., Turmanidze N., Dolidze K., Diasamidze I. and Jakeli E. (2020) Phytochemical study of endemic species *Helleborus caucasicus* and *Helleborus abchasicus*. *HighTech and Innovation Journal* 1(1), pp5 (doi: 10.28991/IIJ-2020-01-01-04).

Bergamasco R. and Horn D.H.S. (1983) Distribution and role of insect hormones in plants. In: *Endocrinology of Insects*, A.R. Liss, New York, pp. 627-654.

Berkenov A.K., Datkhayev U.M. and Mombekov S.E. (2016) Emphasizing and purification 2B,3B,14A,20R,22R,25 hexahydroxy 5B(N) cholest-7-en-6-OH *Serratula cardunculus* (Pall.) Schischk (sic). *Vestnik: Pharmacy & Pharmacology* (2), 369-373.

Berkenov A.K. and Datkhayev U.M. (2017) New technology acquisition 3 α ,14 α ,22R,25-tetrahydroxy-5 α (H)-cholest-7-en-6-one and biological research anti-inflammatory activities; *International Journal of Green Pharmacy* 11(3), 149-153.

Abstract:

Aim: Design a new pharmaceutical technology for isolation novel ecdysteroid 3 α ,14 α ,22R, 25-tetrahydroxy-5 α (H)-cholest-7-en-6-one from the epigeal organs of *ComastomatennellaRottb.* **Research NMR spectroscopy and biological activity.** **Material and Methods:** The isolation procedure involved extraction with aqueous ethanol, partitioning(petroleum ether–ethyl acetate) of the obtained extract to remove non-polar impurities, extraction of the water layer with isobutyl alcohol, and subsequent purification by aluminum oxide column chromatography.

According to NMR spectroscopy, mass spectrometry, and microanalysis, the structure of 3 α ,14 α ,22R,25-tetrahydroxy-5 α (H)-cholest-7-en-6-one (1). Results and Discussion: During the study of plants *Comastomatena* Rottb was isolated new phytoecdysteroid. According to NMR spectroscopy, mass spectrometry, and microanalysis, the structure of 3 α ,14 α ,22R,25-tetrahydroxy-5 α (H)-cholest-7-en-6-one. Isolated object was examined for anti-inflammatory activity of the compound. Thus, the present study established high anti-inflammatory activity of 3 α ,14 α ,22R,25-tetrahydroxy-5 α (H)-cholest-7-en-6-one in the dose of 50 mg/kg in an experimental model of an acute exudative reaction. Conclusion: From the results of the study on the anti-inflammatory activity, it became clear that object potently and significantly reduced the number of abdominal writhings as compared to the control animals.

Bespayeva A.M., Tuleuov B.I., Habdolda G., Turmuhkambetov A.A., Tuleuova B.K., Isaiynova L.A., Kuatbayev O.U. and Adekenov S.M. (2012) The spread of 20-hydroxyecdysone and its analogues in plants. *Chemistry* 2(66) 13-16 [in Russian, with English abstract].

Bhakuni D.S., Bittner M., Sammes P.G. and Silva M. (1974) The chemistry of *Podocarpus* species of Chile. *Rev. Latinoamer. Quím.* 5, 163-168.

Bittner M. and Silva M. (1992) Estudio Químico de las especies de la Familia Podocarpaceae en Chile [Chemical study of the species in the Family Podocarpaceae in Chile]. In: *Química de la flora de Chile* (Ed. Munoz O.). Universidad de Chile, pp. 243-261 [in Spanish].

Blackford M, Clarke B and Dinan L (1996) Tolerance of the Egyptian Cotton Leafworm *Spodoptera littoralis* (Lepidoptera: Noctuidae) to ingested phytoecdysteroids. *Journal of Insect Physiology* 42(10), 931-936. Abstract: The Egyptian cotton leafworm *Spodoptera littoralis* is remarkably resistant to exogenous ecdysteroids, and is able to tolerate both prolonged exposure to artificial diets containing up to 100 ppm 20-hydroxyecdysone (20E) and single dose administration of up to 10 μ g ecdysone or 20E without any adverse effects on growth and development. In addition, it feeds readily on *Chenopodium album*, a plant which has been shown to contain levels of ecdysteroid reaching 175 ppm. The significance of these results in relation to the phytoecdysteroid content of the larval host plants of *S. littoralis* is discussed.

Blackford MJP and Dinan L (1997a) The effects of ingested 20-hydroxyecdysone on the larvae of *Aglais urticae*, *Inachis io*, *Cynthia cardui* (Lepidoptera: Nymphalidae) and *Tyria jacobaeae* (Lepidoptera: Arctiidae). *Journal of Insect Physiology* 43(4), 315-327.

Abstract: A comparative survey was carried out to investigate the effects, distribution and metabolism of ingested 20-hydroxyecdysone in four species of lepidopteran larvae in relation to the phytoecdysteroid content of the insect's host plants. Analysis of the leaves of the host plants of each of the species revealed a strong relationship between the levels of phytoecdysteroids and the relative tolerance of the larvae to ingested 20-hydroxyecdysone. Monophagous or oligophagous species (*Aglais urticae*, *Inachis io*) feeding on ecdysteroid-negative host plants were either deterred from feeding or showed marked abnormalities in growth and development after incorporation of 20-hydroxyecdysone in their diets. Oligophagous or polyphagous species (*Tyria jacobaeae*, *Cynthia cardui*) which feed on host plants from families which are known to contain phytoecdysteroid-positive species, were able to tolerate low levels of 20-hydroxyecdysone in their diets, but exhibited developmental defects at high concentrations. These species were termed semi-tolerant. In each of the species, ingested [3 H]20-hydroxyecdysone appeared to follow the same fate as injected [3 H]20-hydroxyecdysone. The data are compared to those obtained in previous studies, where truly polyphagous species were shown to tolerate very high concentrations of 20-hydroxyecdysone in their diets by the production of ecdysteroid 22-fatty acyl esters.

Blackford M and Dinan L (1997b) The tomato moth *Lacanobia oleracea* (Lepidoptera: Noctuidae) detoxifies ingested 20-hydroxyecdysone, but is susceptible to the ecdysteroid agonists RH-5849 and RH-5992. *Insect Biochemistry and Molecular Biology* 27(2), 167-177.

Abstract: Larvae of *Lacanobia oleracea* (Tomato moth) are able to feed on a diet containing 400 ppm 20-hydroxyecdysone (20E) without any adverse effects on growth and development. This can be attributed to the fact that ingested [3 H]20E is rapidly metabolised by conjugation with common long-chain fatty acids, followed by rapid excretion. The polar metabolites 20,26-dihydroxyecdysone and 20-hydroxyecdysoneic acid have been identified after injection of [3 H]20E into the larval haemolymph. This presumably represents the metabolic fate of endogenous ecdysteroids. In contrast, ingestion of >1 ppm RH-5992 and >10 ppm RH-5849 induces a premature, abnormal and lethal larval moult. The effects of lower concentrations of the ecdysteroid agonists are also investigated.

Blackford M and Dinan L (1997c) The effects of ingested ecdysteroid agonists (20-hydroxyecdysone, RH5849 and RH5992) and an ecdysteroid antagonist (cucurbitacin B) on larval development of two polyphagous lepidopterans (*Acherontia atropos* and *Lacanobia oleracea*). *Entomologia Experimentalis et Applicata* **83**, 263-276.

Abstract: Larvae of two polyphagous lepidopteran species, *Lacanobia oleracea* (Tomato moth) and *Acherontia atropos* (Death's head hawkmoth), have been treated with representatives of three classes of compounds which are known to interact with the ligand binding site of insect steroid hormone receptors: a steroidal ecdysteroid (20-hydroxyecdysone; 20E; agonist), two non-steroidal dibenzoylhydrazines (RH5849 and RH5992; agonists) and the triterpenoid cucurbitacin B (cucB; antagonist). Both species are unaffected by dietary 20E at concentrations up to 400 ppm. *L. oleracea* metabolises ingested [³H]20E to a mixture of C-22 fatty acyl esters, while *A. atropos* excretes [³H]20E unmetabolised. Both species are susceptible to the dibenzoylhydrazines when these are incorporated into the diet, with RH5992 proving lethal at 1 ppm and RH5849 proving lethal at 10 ppm. Thus, the metabolic/excretion mechanisms which are so effective against ingested 20E do not recognise the non-steroidal agonists. The two species showed varying degrees of tolerance to cucB; *L. oleracea* was unaffected by 400 ppm in the diet, while *A. atropos* showed effects at 400 ppm, but not at 40 ppm. Feeding cucB in conjunction with 20E or injecting cucB when the ecdysteroid titre was low or rising also had no effect in *L. oleracea*. It is suggested that tolerance to the two classes of natural products (ecdysteroids and cucurbitacins) is associated with the occurrence of these compounds in some of the host plants of these species; evidence is presented for the presence of phytoecdysteroids in the host plants.

Blunt J.W., Lane G.A., Munro M.H.G. and Russell G.B. (1979) The absolute configuration at C24 of the ecdysteroids dacrysterone, pterosterone and ponasterone C. *Australian Journal of Chemistry* **32**, 779-782.

Abstract: The shielding values in ¹³C N.M.R. spectra of the ecdysteroids dacrysterone, pterosterone, and ponasterone C have been assigned, together with those for the 20,22- and 22,24-benzylidene acetal derivatives of the latter. From the N.M.R. data the C24 configuration of dacrysterone is assigned as 24R, while the 24s configuration is assigned to pterosterone and ponasterone C.

Bokov D.O., Sidorova Y.S., Mazo V.K. and Bessonov V.V. (2020) Prospects for the use of spinach (*Spinacia oleracea* L.) containing phytoecdysteroids and polyphenols. *Pharmacognosy Journal* **12**(2), 246-250.

Abstract:

Background: Phytoadaptogens, biologically active compounds increasing the nonspecific resistance of the human organism, are well known for the prevention and correction of stressful conditions. Phytoadaptogens group includes phytoecdysteroids and polyphenols, that are characterized by the multiplicity of pharmacological effects in combination with the low toxicity. According to literature data, spinach (*Spinacia oleracea* L.) is a promising source of these compounds. This work aims to systematize data on the chemical composition of biologically active compounds of spinach, that determine its adaptogenic properties and concentration methods in the spinach processing for use in specialized foods and dietary supplements. Materials and Methods: Manifold electronic search engines, electronic databases, and libraries such as Google, Google scholar, Crossref, Indian Science Abstracts, Emerging Sources Citation Index, e-Library, Scopus, Web of Science, Pubmed, Chemical Abstracts, Index Copernicus, scientific literature had been searched and data obtained. Results: Botanical characteristics of spinach, main cultivation conditions, the latest data on the chemical composition of raw spinach material cultivars and extracts based on it are presented in this study. Schemes for the obtaining of products enriched with polyphenols and ecdysteroids are considered, and ways of proper purification are mentioned. Conclusion: It is recommended to introduce spinach into the diet of healthy people in order to increase the functional reserves of a person during periods of hypovitaminosis, overwork, intense physical exertion, and also to compensate for the adverse effects of external factors. Spinach extracts containing phytoecdysteroids (20-hydroxyecdysone) and polyphenols (flavonoids) can be used as a prophylactic to overcome the negative effects of stress, accelerate recovery after strong physical and mental stress, particularly for people with extreme occupations, athletes, and those who are engaged in hard physical labor.

Boo K.-h., Cho S.-k., Chae H.-b., Jin S.-b., Lee D.-s., Kim D.-w., Cho M.-j. and Riu K.-z. (2001) Isolation of genes involved in ecdysteroid biosynthesis in *Achyranthes japonica*. *Han'guk Nonghwa Hakhoe chi (Agricultural Chemistry and Biotechnology)* **44**(3), 153-161.

Abstract: In order to isolate genes involved in ecdysteroids biosynthesis in plants, total RNA was isolated from *Achyranthes japonica* Nakai, and RT-PCR was performed using degenerate primers selected based on the results of multi-alignment of four cytochrome P450 genes from plants and a putative ecdysone 20-hydroxylase gene from an insect. Fourteen partial cDNA clones showing unique base sequences were obtained, out of which six showed homologies at the levels of nucleotide and amino acid sequences to the other cytochrome P450 genes known to be involved in the ecdysteroid biosynthesis. Of the six clones, four showed relatively high homologies to a putative ecdysone 20-hydroxylase gene isolated from an insect.

Boo K.H., Lee D., Jeon G.L., Ko S.H., Cho S.K., Kim J.H., Park S.P., Hong Q., Lee S-H., Lee D-S. and Riu K.Z. (2010) Distribution and biosynthesis of 20-hydroxyecdysone in plants of *Achyranthes japonica* Nakai. *Biosciences, Biotechnology and Biochemistry* **74**(11), 2226-2231.

Abstract: There is increasing interest in phytoecdysteroids (PEs) because of their potential role in plant defense against insects. To understand the mechanism regulating their levels in plants, the fluctuation, distribution, and biosynthesis of PE 20-hydroxyecdysone (20E) examined in *Achyranthes japonica*. The total amount of 20E per individual plant initially remained at a constant level, and increased markedly after the first leaf pair (LP) stage, while the concentration of 20E in a given plant decreased rapidly during vegetative growth. In addition, the incorporation of [2-¹⁴C]-mevalonic acid into 20E did not differ significantly depending on plant organs and developmental stages, suggesting that biosynthesis of 20E is not restricted to particular organs or growth stages.

Boo K.H., Lee D., Nguyen Q.V., Jin S.B., Kang S., Viet C.D., Park S.P., Lee D-S. and Riu K.Z. (2013) Fluctuation of 20-hydroxyecdysone in individual organs of *Achyranthes japonica* during reproductive growth stage and its accumulation into seed. *Journal of the Korean Society of Applied Biological Chemistry* **56**, 335-338.

Abstract: To better understand 20-hydroxyecdysone (20E) fluctuation and accumulation in perennial plant, 20E concentration in individual organs of *Achyranthes japonica* during reproductive growth stage were analyzed by high performance liquid chromatography (HPLC). Concentrations of 20E in root and floral part were much higher than those in stem and leaf during reproductive growth stage and rapidly increased from flowering stage in August to seed-setting stage in October, and thereafter decreased at the stage of seed maturing in November. In contrast, the 20E concentrations in stem and leaf gradually decreased during reproductive growth. In the analysis of detailed fluctuation of 20E in floral part, the 20E concentration was highest in the seed at the early stage of seed development, compared to flower, peduncle, seed coat, and/or seed in other growth stages, and decreased during seed maturation. The accumulation of 20E in reproductive organs, especially seed and root, suggests that 20E has a defensive role for protection of especially newly developing organs against phytophagous insects.

Borovikova E.B. and Baltaev U.A. (1999) Lesterone, a new phytoecdysteroid from the seeds of *Leuzea carthamoides*. *Khimiya Prirodnykh Soedinenii* (2), 205-206 [in Russian]/*Chemistry of Natural Compounds* **35**(2), 182-183 [in English].

Abstract: A new phytoecdysteroid, lesterone has been isolated from the seeds of *Leuzea (Rhaponticum) carthamoides*. It has been unambiguously identified as 5 β -cholest-7-en-2 α ,3 α ,11 β ,14 α ,20R,22R,25-heptahydroxy-6-one with the aid of NMR and mass spectroscopies.

Borovikova E.B., Shangaraeva G.S. and Baltaev U.A. (1999) Rhapisterone D 20-acetate from the seeds of *Leuzea carthamoides*. *Khimiya Prirodnykh Soedinenii* (2), 207-208 [in Russian]/ *Chemistry of Natural Compounds* **35**(2), 184-185 [in English].

Abstract: In addition to makisterone A, the new ecdysteroid rhapisterone D 20-acetate has been isolated from the seeds of *Leuzea carthamoides*.

Bratoeff E., Pérez-amador M.C. and Ramírez E. (1996) Ecdysteroids and other secondary metabolites from *Amaranthus indica* Mill (Amaranthaceae). *Phyton - International Journal of Experimental Botany* **58**(1-2), 119-123.

Abstract: A general screening for secondary metabolites was carried out with the methanolic extract from the leaves and stems of *Amaranthus indica* Mill (Amaranthaceae). Three ecdysteroids: amasterol, ecdysterone, pterosterone, one sesquiterpene lactone, iresin, and an isoflavone, tlatlancuayin were isolated.

Brillatz T., Jacmin M., Vougianniopoulou K., Petrakis E.A., Kalpoutzakis E., Houriet J., Pellissier L. Rutz A., Marcourt L., Queiroz E.M., Crawford A.D., Skaltsounis A-L. and Wolfender J-L. (2020) Antiseizure potential of the ancient Greek medicinal plant *Helleborus odoratus* subsp. *cyclophyllus* and identification of its main active principles. *Journal of Ethnopharmacology* **259**, article 112954, pp14.

Abstract:

Ethnopharmacological relevance

Ethnopharmacological data and ancient texts support the use of black hellebore (*Helleborus odoratus* subsp. *cyclophyllus*, Ranunculaceae) for the management and treatment of epilepsy in ancient Greece.

Aim of the study

A pharmacological investigation of the root methanolic extract (RME) was conducted using the zebrafish epilepsy model to isolate and identify the compounds responsible for a potential antiseizure activity and to provide evidence of its historical use. In addition, a comprehensive metabolite profiling of this studied species was proposed.

Materials and methods

The roots were extracted by solvents of increasing polarity and root decoction (RDE) was also prepared. The extracts were evaluated for antiseizure activity using a larval zebrafish epilepsy model with pentylentetrazole (PTZ)-induced seizures. The RME exhibited the highest antiseizure activity and was therefore selected for bioactivity-guided

fractionation. Isolated compounds were fully characterized by NMR and high-resolution tandem mass spectrometry (HRMS/MS). The UHPLC-HRMS/MS analyses of the RME and RDE were used for dereplication and metabolite profiling.

Results

The RME showed 80% inhibition of PTZ-induced locomotor activity (300 µg/ml). This extract was fractionated and resulted in the isolation of a new glucopyranosyl-deoxyribonolactone (**1**) and a new furostanol saponin derivative (**2**), as well as of 20-hydroxyecdysone (**3**), hellebrin (**4**), a spirostanol glycoside derivative (**5**) and deglucohellebrin (**6**). The antiseizure activity of RME was found to be mainly due to the new furostanol saponin (**2**) and hellebrin (**4**), which reduced 45% and 60% of PTZ-induced seizures (135 µM, respectively). Besides, the aglycone of hellebrin, hellebrigenin (**S34**), was also active (45% at 7 µM). To further characterize the chemical composition of both RME and RDE, 30 compounds (**A7-33**, **A35-37**) were annotated based on UHPLC-HRMS/MS metabolite profiling. This revealed the presence of additional bufadienolides, furostanols, and evidenced alkaloids.

Conclusions

This study is the first to identify the molecular basis of the ethnopharmacological use of black hellebore for the treatment of epilepsy. This was achieved using a microscale zebrafish epilepsy model to rapidly quantify *in vivo* antiseizure activity. The UHPLC-HRMS/MS profiling revealed the chemical diversity of the extracts and the presence of numerous bufadienolides, furostanols and ecdysteroids, also present in the decoction.

Budešinsky M., Vokac K., Harmatha J. and Cvacka J. (2008) Additional minor ecdysteroid components of *Leuzea carthamoides*. *Steroids* **73** 502-514.

Abstract: Seventeen additional minor ecdysteroid compounds were isolated and identified from the roots of *Leuzea carthamoides* (Wild.) DC. Eight of them are new phytoecdysteroids: carthamoleusterone (**13**) is a new side-chain cyclo-ether with five-membered ring; 14-epi-ponasterone A 22-glucoside (**12**) is a rare and unusual natural 14 beta-OH epimer; 15-hydroxyponasterone A (**11**) is also new and rare with its C-15 substituted position, as well as 22-deoxy-28-hydroxymakisterone C (**18**) possessing secondary hydroxyl in position C-28 and 26-hydroxymakisterone C (**20**) with hydroxy groups in positions 25 and 26. New are also 1 beta-hydroxymakisterone C (**21**) and 20,22-acetonides of inokosterone (**8**) and integristerone A (**10**). Series of already known ecdysteroids: ecdysone (**1**), 20-hydroxyecdysone 2- and 3-acetates (**3** and **4**), turkesterone (**6**), inokosterone (**7**), 24-epi-makisterone A (**14**), and amarasterone A (**22**) are reported here as new constituents of *L. carthamoides*. Seven earlier reported *Leuzea* ecdysteroids: 20-hydroxyecdysone (**2**), ajugasterone C (**5**), integristerone A (**9**), 24(28)-dehydromakisterone A (**15**), 24(28)-dehydroamarasterone B (**16**), (24Z)-29-hydroxy-24(28)-dehydromakisterone C (**17**) and makisterone C (**19**) are also included because they are now better characterized.

Bunu M.I., Ndinteh D.T., Macdonald J.R., Langat M.K., Isyaka S.M., Sadgrove N.J., Melnikovova I. and Fernandez-Cusimamani E. (2021) Ecdysteroids from the stem bark of *Vitex doniana* Sweet (Lamiaceae; ex. Verbenaceae): a geographically variable African medicinal species. *Antibiotics* **10**, article 937 (doi: 10.3390/antibiotics10080937).

Abstract: *Vitex doniana* Sweet is an African medicinal species that is prescribed as an aqueous bark extract to be applied topically or orally to achieve anti-infective outcomes. In select regions it is also taken orally as an antimalarial agent. The aim of the current study was to explore the biological properties of *V. doniana* and isolated compounds in the context of pathogenic bacteria and the protozoan parasite *Plasmodium falciparum*. Three compounds were isolated and assigned by nuclear magnetic resonance spectroscopy as ecdysteroids: (1) 20-hydroxyecdysone, (2) turkesterone, and (3) ajugasterone C. Interestingly, two of these compounds had not previously been identified in *V. doniana*, providing evidence of chemical variability between regions. The bark extract and three ecdysteroids were screened for activity against a panel of pathogenic bacteria associated with skin, stomach and urinary tract infections, and the protozoan parasite *P. falciparum*. The crude extract of the bark inhibited all bacterial strains with MIC values of 125–250 µg.mL⁻¹. The three isolated compounds demonstrated less activity with MIC values of 500–1000 µg.mL⁻¹. Furthermore, no activity was observed against *P. falciparum* at the screening concentration of 4.8 µg.mL⁻¹. Nevertheless, we present a hypothesis for the possible mechanism for symptomatic relief of malarial fever, which may involve reduction of prostaglandin E(1) & E(2) activity in the hypothalamus via modulation of the monoaminergic system. While further studies are required to identify all antimicrobial agents within this plant species and to determine the cytotoxicity of each of these compounds, these data suggest that the traditional application of this species as an antiseptic is valid.

Bürecy K. (1977) Nachweis von Ecdysonen in Gametophyten und Sporophyten von *Anemia phyllitidis* (Schizaeaceen) (Evidence of ecdysones in the gametophyte and sporophyte of *Anemia phyllitidis* (Schizaeaceae)). *Zeitschrift für Pflanzenphysiologie* **81**, 466-469 [in German, with an English abstract].

Summary: The occurrence of ecdysterone and a more unpolar ecdysone — probably ponasterone A — could be demonstrated as well in the sporophyte as in the culture medium of the gametophyte of *Anemia phyllitidis*.

Burns B.G. and Gilgan M.W. (1977) The isolation and identification of makisterone A from the yew, *Taxus cuspidata*. Canadian Journal of Chemistry 55, 1129-1134.

Abstract: The ecdysteroids were extracted from yew, *Taxus cuspidata*, needles and twigs and recovered from the aqueous extract by reversed phase adsorption. Purification of the ecdysteroids was achieved by a single solvent partition and a dry column chromatogram followed by fractionation on an adsorptive Porasil A (60) column. Makisterone A was a minor component identified by melting point, liquid chromatography, gas chromatography of the trimethylsilyl ether, mass spectrometry, and nuclear magnetic resonance spectrometry. A second minor component was isolated but not identified.

Calcagno M.P., Camps F., Coll J., Melé E., Messeguer J. and Tomás J. (1994) Sengosterone, and ecdysteroid present in *Ajuga reptans* L. Anales de Química de la Sociedad Española de Química 90(7/8), 483-486.

Calcagno M.P., Camps F., Coll J. and Melé E. (1995a) Acetylated ecdysteroids from *Ajuga reptans* var. *atropurpurea* (Lamiales: Lamiaceae). European Journal of Entomology 92, 287-294.

Abstract: The structural elucidation of acetylated ecdysteroids isolated from dried roots of *Ajuga reptans* var. *atropurpurea*, based on spectroscopic procedures, is reported. Among them 2-O-acetyl- and 3-O-acetyl-20-hydroxyecdysone (20E2Ac, 20E3Ac) and 3-O-acetyl-29-norcyasterone (29NCY3Ac) had previously been only partially described and 3-O-acetylcysterone (CY3Ac) was heretofore unreported.

Calcagno M.-P., Camps F., Coll J., Melé E. and Sánchez-Baeza F. (1995b) A new family of phytoecdysteroids isolated from aerial part of *Ajuga reptans* var. *atropurpurea*. Tetrahedron 51(44), 12119-12126.

Abstract: A new family of four phytoecdysteroids have been isolated from aerial part of *Ajuga reptans* var. *atropurpurea* harvested at the beginning of autumn. These compounds show a novel 22-oxo-12 β -hydroxy functionalization of the C-28 and C-29 γ -lactonic ecdysteroids. The structure of those compounds was inferred from the corresponding IR, ¹H and ¹³C-NMR and HPLC-MS(TSP) spectral data.

Calcagno M.P., Camps F., Coll J., Melé E. and Sánchez-Baeza F. (1996) New phytoecdysteroids from roots of *Ajuga reptans* varieties. Tetrahedron 52(30), 10137-10146.

Abstract: Reptansterone (**8**), a new C-29 phytoecdysteroid with a δ -lactone in the side chain, was isolated from roots of a green variety of *Ajuga reptans*. Likewise, other unprecedented members of this polyhydroxysteroid family namely 28-*epi*-sengosterone (**9**), 5,29-dihydroxycapitasterone (**10**) 2- and 3-dehydroajugalactone (**11** and **12**) were isolated from *A. reptans* var. *atropurpurea*. The structures of all these new compounds were inferred from the corresponding ¹H and ¹³C-NMR homo- and heterocorrelations and IR and HPLC-MS(TSP) spectral data.

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Calderón A.I., Chung K.S. and Gupta M.P. (2009) Ecdysteroids from *Dichorisandra hexandra* (Commelinaceae). Biochemical Systematics and Ecology 37 693–695

Camps F., Coll J. and Cortel A. (1981) Allelochemicals on insects isolated from *Ajuga plants* (Labiatae). Rev. Latinoam. Quim. 12, 81-88.

Camps F., Coll J. and Cortel A. (1982) 29-Norsengosterone and 29-norcyasterone, new C-28 phytoecdysteroids from *Ajuga reptans* (Labiatae). Chemistry Letters 1313-1316.

Abstract: Ajugalactone (**1**), cyasterone (**3**), polypodine B (**5**), β -ecdysone (**6**) and two new C-28 phytoecdysteroids, 29-norsengosterone (**2**) and 29-norcyasterone (**4**), have been isolated from *Ajuga reptans* and characterized by spectral means and X-ray diffraction analysis of 2,3,20,22-diacetonide of **4**.

Camps F., Coll J. and Dargallo O. (1984) Phytoecdysteroids from *Ajuga chamaepitys*. Anales de Química de la Sociedad Española de Química 81, 74-75.

Camps F., Coll J., Cortel A., Molins E. and Miravittles C. (1985) 2-Acetyl and 3-acetyl-29-norcyasterone, new minor phytoecdysteroids from *Ajuga reptans* (Labiatae). Journal of Chemical Research (S) 14-15, (M) 0241-0266.

Camps F., Coll J., Marco M.P. and Tomás J. (1990a) Efficient determination of phytoecdysteroids from *Ajuga* species and *Polypodium vulgare* by high-performance liquid chromatography. Journal of Chromatography 514, 199-207.

Abstract: Efficient chromatographic conditions were established for the simultaneous determination of eight phytoecdysteroids by high-performance liquid chromatography. Spherisorb ODS-2 columns, ultraviolet detection, isopropanol-water as the mobile phase and temperature control were used. Ecdysteroids were obtained by purification of methanol plant extracts with Sep-Pak C18 cartridges. The results compared well with those obtained by other chromatographic methods in terms of resolution, selectivity and efficiency.

Camps F., Coll J., Marco M.P., Tomás J., Melé E., Messeguer J. and Claveria E. (1990b) On the production of ecdysteroids by plant cell and tissue cultures. *Invertebrate Reproduction and Development* **18**, 107.

Camps F., Claveria E., Coll J., Marco M-P., Messeguer J. and Melé E. (1990c) Ecdysteroid production in tissue cultures of *Polypodium vulgare*. *Phytochemistry* **29**(12), 3819-3821.

Abstract: The haploid phase of *Polypodium vulgare* obtained by *in vitro* culture has been shown to produce a greater amount of phytoecdysteroids than the corresponding diploid plant. Comparative studies on the dependence of this content on the geographical origin of the parent plant and culture conditions have also been carried out.

Canonica L., Danieli B. and Weisz-Vincze I. (1972) Structure of muristerone A, a new phytoecdysone. *Journal of the Chemical Society, Chemical Communications* 1060-1061.

Abstract: Muristerone A, a new phytoecdysone isolated from *I pomoea calonyction*(Choisy) Hallier f. sp. nova, has been identified as 2 β ,3 β ,5 β ,11 α ,14 α ,20R,22R-hepta-hydroxycholest-7-en-6-one.

Canonica L., Danieli B., Ferrari G., Haimova M.A. and Krepinsky J. (1973a) The structure of a new phytoecdysone kaladasterone: an application of ¹³C magnetic resonance spectroscopy to structural problems. *Experientia* **29**(9), 1062-1063.

Zusammenfassung: Isolierung und Strukturaufklärung von Kaladasteron (C₂₇H₄₂O₇), eines neuen Phytoecdysons, werden beschrieben.

Canonica L., Danieli B., Ferrari G., Krepinsky J. and Rainoldi G. (1973b) Structure of calonysterone, an unusually modified phytoecdysone. *Journal of the Chemical Society, Chemical Communications* 737-738.

Abstract: The structure of 2 β ,3 β ,6,20R, 22R,25-hexahydroxycholesta-5,8(9),14-trien-7-one (I) has been assigned to calonysterone, a phytoecdysone isolated from kaladana (*I pomoea* sp.), by ¹³C and ¹H n.m.r. spectroscopy.

Canonica L., Danielli B., Ferrari G., Krepinsky J. and Weisz-Vincze I. (1975) A novel method of isolation of phytoecdysones from kaladana seeds. *Phytochemistry* **14**, 525-527.

Abstract: A new simplified technique is described for the isolation and separation of phytoecdysones from kaladana seeds. Using this method ecdysone, crustecdysone, muristerone A, kaladasterone, calonysterone and makisterone A were obtained. In addition methyl 3,4-dihydroxycinnamate was found in the seed.

Canonica L., Danielli B., Ferrari G., Krepinsky J. and Haimova M. (1977a) New phytoecdysones from kaladana. I. Structure of muristerone A and kaladasterone. *Gazzetta Chimica Italiana* **107**, 123-130.

Zusammenfassung: Aufgrund massen-, UV-, IR- und NMR-spektroskopischer Untersuchungen und chemischer Reaktionen der Titelverbindungen sowie einiger ihrer Derivate werden den beiden aus dem Samen von Kaladana, einer zur Familie Convolvulaceae gehörenden Pflanze *Ipomoea calonyction* (Choisy), isolierten Phytoecdysonen die Strukturen (I) und (II) zuerkannt.

Canonica L., Danielli B., Palmisano G., Ferrari G. and Krepinsky J. (1977b) New phytoecdysones from kaladana II. Structure of calonysterone. *Gazzetta Chimica Italiana* **107**, 229-235.

Cao V.D., Riu K-Y. and Boo K.H. (2018) Biosynthesis and accumulation of 20-hydroxyecdysone in individual male and female spinach plants during the reproductive stage. *Plant Physiology and Biochemistry* **129**, 394-399.

Abstract: The steroid 20-hydroxyecdysone (20E) is a major component of phytoecdysteroid in plants and may play a defensive role against insect pests in higher plants. In spinach, the biosynthesis and accumulation of 20E have been investigated during the vegetative stage; however, these processes have not been clearly studied during the reproductive stage, particularly in male and female individuals. In this study, we analyzed the level and distribution of 20E in individual male and female spinach plants during the reproductive stage via high performance liquid chromatography (HPLC). We found that 20E biosynthesis and accumulation were markedly different between male and female spinach during the late flowering stage. Compared with the male plant, biosynthesis of 20E in the leaves was more active and its accumulation in the floral parts was higher in female plants during the late flowering stage. These results indicate that the female reproductive organs at least in PE-positive plants could be effectively protected against harmful insects via active biosynthesis and accumulation of PE during the late flowering stage to protect floral parts from harmful insects for seed formation and store the available 20E in seeds for the next generation.

Cao Y., Gu C., Zhao F., Tang Y., Cui X. Shi L., Xu L. and Yin L. (2017) Therapeutic effects of *Cyathula officinalis* Kuan and its active fraction on acute blood stasis rat model and identification constituents by HPLC-QTOF/MS/MS. *Pharmacognosy Magazine* **13**(52), 693-701.

Abstract:

Background: *Cyathula officinalis* Kuan is widely used in the clinics for the treatment of blood stasis in China.

Objective: To evaluate the improving blood rheology and anti-inflammatory properties of *C. officinalis* Kuan extract (CO) and its active fraction (ACO) on acute blood stasis model Wistar rats and characterize the correlative constituents.

Materials and methods: CO at 0.26, 0.53, and 1.04 g/kg and ACO at 0.38, 0.75, and 1.5 g/kg were administered to acute blood stasis model Wistar rats for 3 days. Whole blood viscosity, plasma viscosity, and the levels of interleukin-6 (IL-6), nitric oxide (NO), tumor necrosis factor alpha (TNF- α), and cyclooxygenase-2 (COX-2) in the plasma were measured. HPLC-QTOF/MS/MS method was used to identify the major constituents of ACO; the properties of two representative components (cyasterone and chikusetsusaponin IV) from ACO on thrombin-induced human umbilical vein endothelial cells damage model were also assessed by the levels of thromboxane A2 (TXA2), endothelin (ET), malondialdehyde (MDA), COX-2, endothelial nitric oxide synthase (eNOS), and superoxide dismutase (SOD).

Results: CO and ACO significantly reduced whole blood viscosity, plasma viscosity, and levels of IL-6, NO, TNF- α , and COX-2 *in vivo*. Forty compounds were identified from ACO, mainly as phytoecdysteroids and saponins. Cyasterone and chikusetsusaponin IV could significantly inhibit levels of TXA2, ET, MDA, and COX-2 and promote the activities of eNOS and SOD *in vitro*.

Conclusion: CO and ACO possessed significant improving blood rheology and anti-inflammatory effects on acute blood stasis model rats and the representative components Cyasterone and chikusetsusaponin IV showed significant anti-inflammatory, antioxidant, and anticoagulant effects *in vitro*.

Summary: *Cyathula officinalis* Kuan is widely used in the clinic for the treatment of blood stasis in China. The *C. officinalis* Kuan extract and the active fraction of *C. officinalis* Kuan (ACO) possessed significant improving blood rheology and anti-inflammatory effects on acute blood stasis model rats. Forty compounds were identified from ACO, mainly as phytoecdysteroids and saponins.

Cao Z. and Zhou S. (2013) *Cyanotis arachnoidea* extract rich in β -ecdysone and preparation method thereof. Chinese Patent CN102872167 A 2013-01-16 [in Chinese].

Abstract: The title *Cyanotis arachnoidea* extract is prepared by the steps of: selecting *Cyanotis arachnoidea*, removing impurities, pulverizing dried *Cyanotis arachnoidea* roots, adding water 4-10 times, performing water extraction for 1.5-2.5 h, centrifugally separating, adsorbing with a macroporous resin column, eluting with ethanol, recovering ethanol, crystallizing, adding ethyl acetate, performing recrystallization, and drying. The *Cyanotis arachnoidea* extract has high β -ecdysone content, good bioavailability of β -ecdysone, and high quality.

Castro A., Coll J., Tandrón Y.A., Pant A.K. and Mathela C.S. (2008) Phytoecdysteroids from *Ajuga macrosperma* var. *breviflora* roots. *Journal of Natural Products* **71**(7), 1294–1296.

Abstract: Three new phytoecdysteroids, ajugacetalsterones C (**1**) and D (**3**) and breviflorasterone (**2**), were isolated from the roots of *Ajuga macrosperma* var. *breviflora* along with five known compounds, namely, 20-hydroxyecdysone, cyasterone, makisterone A, 20-hydroxyecdysone 3-acetate, and 20-hydroxyecdysone 2-acetate. The structures of **1–3** were elucidated on the basis of extensive 1D and 2D NMR spectroscopic studies. The new compounds possess acetal oxygen bridges between C-26 and C-20/C-22, or C-26/C-23, or a lactone bridge between C-26 and C-23.

Chari A., Ben Jannet H. and Mighri Z. (2000) Isolement et élucidation structurale d'un nouveau phytoecdystéroïde antibactérien et de deux dérivés de l'acide cinnamique des feuilles de la plante *Ajuga pseudo-iva*. [Isolation and structural determination of a novel antibacterial phytoecdysteroid and two cinnamic acid derivatives based on *Ajuga pseudo-iva* plant leaves]. *Journal de la Société Chimique de Tunisie* **4**(8), 789-799 [in French, with an English abstract].

Summary: During the search for bioactive metabolites from *A. pseudoiva* leaves, Ben and co-workers isolated five novel monoglycerides 334-338, two novel cinnamic acids 318, 319 and one new steroid 63, along with five known compounds 61, 62, 331-333. Three compounds 331-333 show significant antifeedant activity, which might be associated with the presence of two β -hydroxyalkanoic moieties in each compound [38,48,50,132]. A phytochemical investigation on *A. parviflora* resulted in the isolation of quinols 306-312 and pyrrolizidine alkaloids 343-345. Derivatives 306-308 are new compounds.

Chadin I., Volodin V., Whiting P., Shirshova T., Kolegova N. and Dinan L. (2003) Ecdysteroid content and distribution in plants of *Potamogeton*. *Biochemical Systematics and Ecology* **31**(4), 407-415.

Abstract: Species of the genus *Potamogeton* L. growing in European North-East Russia were studied for the presence of ecdysteroids. Coupled HPLC/RIA showed that all *Potamogeton* species studied (*P. alpinus* Balb., *P. berchtoldii* Fieb., *P. compressus* L., *P. gramineus* L., *P. lucens* L., *P. natans* L., *P. pectinatus* L. and *P. perfoliatus* L.) contain 20-hydroxyecdysone, ecdysone and some unidentified minor compounds in the concentration range 0.2–4.0 µg ecdysone equivalents/g dry wt, depending on the species, growth stage and plant part studied. Detailed study of 20-hydroxyecdysone distribution in individual plants of *P. pectinatus* L. revealed that highest content of ecdysteroids is associated with points of active growth (170–250 µg/g dry wt in apical leaves of main and side-shoots). The presence of ecdysteroids in pondweeds and the pattern of their distribution within individual plants indicate a possible role in the interaction between plants and herbivorous arthropods in aquatic ecosystems.

Chae H-B., Boo K-H., Jin S-B., Lee D-S., Kim D-W., Cho M-J. and Riu K-Z. (2001) Effects of light and some plant growth regulators on ecdysteroid content in *Polypodium vulgare* L. and *Achyranthes japonica* Nakai. Applied Biological Chemistry 44(3), 162-166 [in Korean].

Abstract: Effects of light, methyl jasmonate(MJ), 6-benzylaminopurine(BA), thidiazouron(TDZ), and 2,4-dichlorophenoxyacetic acid(2,4-D) on the contents of ecdysteroids (β -ecdysone+polypodine B) in *Polypodium vulgare* L. and *Achyranthes japonica* Nakai were studied. When the plants of *P. vulgare* were cultured under light control, the ecdysteroids contents in both leaves and rhizomes decreased with increasing light intensity. The ecdysteroids contents in *A. japonica* were also lower when cultured under light than under dark. Among the tested plant growth regulators, MJ, BA, and TDZ increased the ecdysteroids contents in both *P. vulgare* and *A. japonica*.

Chakraborty V. and Bordoloi M. (2003) Pentahydroxycholestenone derivative from Eastern Himalayan yew. Indian Journal of Chemistry, Section B: Organic Chemistry, including Medicinal Chemistry 42, 944-945.

Abstract: Chemical investigation of the Eastern Himalayan yew (*Taxus baccata* L.) found in Arunachal Pradesh, India resulted in the isolation of a phytosteroid and its structure was established as 2,3,9,20,21-pentahydroxy-cholest-7-en-6-one through extensive use of FABMS, ID and 2D NMR spectroscopy.

Chamnipa N., Thanonkeo S. and Thanonkeo P. (2012) Enhance production [sic] of 20-hydroxyecdysone in cell suspension cultures of *Vitex glabrata* R.Br. by elicitor feeding. Journal of Medicinal Plants Research 6(7), 3317-3323.

Abstract: The effects of chitosan and methyl jasmonate on growth and 20-hydroxyecdysone production in cell suspension cultures of *Vitex glabrata*, a medicinal plant in Thailand, were investigated. Elicitation with chitosan at 50 mg/L resulted in 17.16 g/L biomass and 377.09 mg/100 g dry weight (DW) 20hydroxyecdysone, which were 1.62 and 8.33 times higher than the control cultures, respectively. Likewise, addition of methyl jasmonate at 100 µM also enhanced growth and production of 20hydroxyecdysone. The highest growth and 20-hydroxyecdysone production reached 14.44 g/L and 621.76 mg/100 g DW, which were 1.35 and 14.54 times higher in comparison to the control cultures, respectively. This is the first report to indicate that elicitation with chitosan and methyl jasmonate enhanced the production of 20-hydroxyecdysone in cell suspension cultures of *V. glabrata*.

Chan Y.-Y., Wu T.-S., Kuoh C.S. and Damu A.G. (2005) A new phytoecdysteroid from *Ajuga taiwanensis*. Chemical and Pharmaceutical Bulletin 57(7), 836-838.

Abstract: A new phytoecdysteroid, namely ajugalide-E (1), together with twenty five known compounds were isolated from *Ajuga taiwanensis* collected from Tainan, Taiwan. Their structures were determined by spectral analysis including high resolution one- and two-dimensional NMR spectroscopy.

Chen H., Tang B-Q., Chen L., Liang J-Y. and Sun J-B. (2018) Neo-clerodane diterpenes and phytoecdysteroids from *Ajuga decumbens* Thunb. And evaluation of their effects on cytotoxic, superoxide anion generation and elastase release in vitro. Fitoterapia (doi: 10.1016/j.fitote.2018.06.004).

Chen H., Wang C., Qi M., Ge L., Tian Z., Li J., Zhang M., Wang M., Huang L. and Tang X. (2017) Anti-tumor effect of *Rhaponticum uniflorum* ethyl acetate extract by regulation of peroxiredoxin 1 and epithelial-to-mesenchymal transition in oral cancer. Frontiers in Pharmacology 8, 870 (doi: 10.3389/fphar.2017.00870).

Abstract:

Objective: To explore whether *Rhaponticum uniflorum* (*R. uniflorum*) had anti-tumor effects in oral cancer and investigate the molecular mechanisms involved in these anti-tumor effects.

Methods: Chemical compositions of *R. uniflorum* ethyl acetate (RUEA) extracts were detected by ultra-performance liquid chromatography-Q/time-of-flight mass spectrometry (UPLC-Q/TOF-MS), followed by pharmacology-based network prediction analysis. The effects of RUEA extracts on proliferation, apoptosis, migration, and invasion ability of human oral squamous cell carcinoma (OSCC) cell line SCC15 were evaluated by CCK8 assay, Annexin V-fluorescein isothiocyanate/propidium iodide staining, wound healing assay, and Matrigel invasion assay, respectively. The mRNA and protein expression of peroxiredoxin1 (Prx1), the epithelial-to-mesenchymal transition

(EMT) marker E-cadherin, vimentin, and Snail were determined by quantitative real-time reverse transcription polymerase chain reaction and western blotting. A mouse xenograft model of SCC15 cells was established to further evaluate the effect of RUEA extracts *in vivo*. Immunohistochemical assessment of Ki67 and terminal deoxynucleotidyl transferase dUTP nick end labeling staining of apoptotic cells were performed on the tumor tissues to assess the effects of RUEA extracts on proliferation and apoptosis.

Results: Fourteen compounds were identified from RUEA extracts by UPLC-Q/TOF-MS. The pharmacology-based network prediction analysis showed that Prx1 could be a potential binder of RUEA extracts. In SCC15 cells, RUEA extracts inhibited cell viability, induced apoptosis, and suppressed cell invasion and migration in a concentration-dependent manner. After treatment with RUEA extracts, the mRNA and protein expression of E-cadherin increased, whereas those of Prx1, vimentin, and Snail decreased. RUEA extracts also affected the EMT program and suppressed cell invasion and migration in Prx1 knockdown SCC15 cells. In an OSCC mouse xenograft model, RUEA extracts (25 and 250 mg/kg) significantly inhibited the growth of tumors. Compared with the control group, Ki67 expression was reduced and apoptosis rates were elevated in the transplanted tumors treated with RUEA extracts. RUEA extracts increased the expression of E-cadherin and decreased the expression of Prx1, vimentin, and Snail *in vivo*.

Conclusion: RUEA extracts inhibited tumor growth and invasion by reducing Prx1 expression and suppressing the EMT process in OSCC. RUEA extracts may be a potential candidate for OSCC treatment.

Chen J. and Wei Y. (1989) Isolation and identification of ecdysterone in *Serratula chinensis* root. *Zhongcaoyao* 20(7), 296 [in Chinese].

Chen L. and Ding X. (1997) Chemical constituent of uniflower swisscentaury (*Rhaponticum uniflorum*). *Zhongcaoyao* 28(11), 648-650 [in Chinese].

Abstract: 15 Compounds were isolated from root of *Rhaponticum uniflorum* and 11 of them were identified as β -sitosterol, arctic acid, palmitic acid, arctinal, n-tetracosanoic acid, daucosterol, ecdysterone, sucrose, rhapontisterone, turkesterone and maltose.

Chen L., Wang Qi, Wu B., Li Z-f., Feng Y-l., Zhong G-y. and Yang S-l. (2018) Isolation and identification of chemical constituents from *Disporum cantoniense* (II). *Chinese Traditional and Herbal Drugs* (20), 4803-4807 [in Chinese].

Abstract: Objective: To investigate the chemical constituents from *Disporum cantoniense*. Methods: Compounds were isolated and purified by macroporus resin, ODS, Sephadex LH-20, MCI-gel CHP20 resin column chromatography and preparative HPLC. The structures were identified by spectral analysis. Results: Twenty compounds were isolated and identified as 2-hydroxybenzyl alcohol (1), p-hydroxybenzaldehyde (2), 4-hydroxyacetophenone (3), 3,5-dimethoxy-4-hydroxy-benzaldehyde (4), 4-(4-hydroxyphenyl)-2-butanone (5), neoliquiritin (6), 2,3,5,4'-tetrahydroxy stilbene-2-O- β -D-glucoside (7), (E)-1-(4'-hydroxyphenyl)-but-1-en-3-one (8), isoquercitrin (9), 3-hydroxy-1-(4-hydroxy-3,5-dimethoxy-phenyl)-1-propanone (10), icariol A2 (11), ecdysterone (12), glansreginic acid (13), hesperidin (14), ononin (15), quercetin (16), isorhamnetin-3-O- β -D-glucoside (17), (E)-4-(4-hydroxy-3-methoxyphenyl) but-3-en-2-one (18), (-)-secoisolaricresinol (19), and luteolin(20). Conclusion: Compounds 1,3—8,10—15, and 17—19 are isolated from the genus of *Disporum* for the first time.

Chen T, Diao Q-Y., Yu H-Z., Jiao C-L. and Ruan J. (2017) Phytochemical, cytotoxic and chemotaxonomic study ofn *Ajuga forrestii* Diels (Labiateae). *Natural Product Research* (doi: 10.1080/14786419.2017.1371161).

Abstract: A phytochemical investigation of *Ajuga forrestii* Diels led to the isolation of 14 compounds, including eight neo-clerodane diterpenes (1-8), two phytoecdysteroids (9, 11), one stigmastane sterol (10) and three iridoid glycosides (12-14). The structures of these compounds were identified by spectroscopic methods and a comparison of their data with those reported in the literature. This is the first report of compounds 1-14 from *A. forrestii*. The cytotoxic activities of the aqueous extract of *A. forrestii* and several compounds have been studied and the chemotaxonomic significance of isolated compounds has also been summarised.

Chen Y-S. and Feng Y-Q. (2021) Rapid determination of endogenous 20-hydroxyecdysone in plants on MALDI-TOF/TOF mass spectrometry via chemical labelling based on boronate affinity. *Journal of Analysis and Testing* (doi: 10.1007/s41664-021-001-00179-8).

Abstract: 20-Hydroxyecdysone (20E) derived from plants has a wide range of physiological and pharmacological effects on animals and humans, and rapid and sensitive methods for screening of the endogenous 20E in plants are thus required. Herein, a matrix-assisted laser desorption/ionization time-of-flight tandem mass spectrometry (MALDI-TOF/TOF MS) method is described for rapid and sensitive determination of endogenous 20E in plants. It is based on the use of the (3-(acridin-9-ylamino) phenyl) boronic acid (AYPBA) as the mass tag to assist the MS and tandem MS (MSⁿ) analysis of 20E on MALDI-TOF/TOF MS. Good linearity was obtained with a determination coefficient (R^2) larger than 0.99 in the range of 0.025–2.5 μ M. The limit of detection (LOD) was 2.4 fmol.

Acceptable precision and accuracy were gained by intra- and inter-day analysis with relative standard deviations less than 19.5% and relative recoveries ranging from 85.7 to 105.2%. In addition, the AYPBA labeled 20E produced abundant characteristic fragment ions under the high energy collision-induced dissociation, which facilitated the identification of 20E by MS² analysis on MALDI-TOF/TOF MS. Using the method, we enabled the identification and quantification of endogenous 20E in four herbs including *Cyanotis arachoides*, *Achyranthes bidentata*, *Spinacia oleracea* and *Chenopodium quinoa* Willd., demonstrating the feasibility of the proposed method for screening of the endogenous 20E in plants.

Cheng C-m., Dai Y., Huang X-z., Zhu Y., Dai J-h., Liu X-f. And Wang J. (2010) Chemical constituents from roots of *Tinospora sagittata* var. *yunnanensis*. *Chinese Traditional and Herbal Drugs* (5), 689-692 [in Chinese].

Abstract: Chem. constituents from roots of *Tinospora sagittata* var. *yunnanensis* were studied. Various chromatog. techniques were used to isolate and purify the constituents. The structures were elucidated by chem. evidence and spectral methods. Eleven compounds were isolated and identified as columbin (1), palmatoside C (2), fibleucin (3), palmatine (4), jatrorrhizine (5), columbamine (6), 20-hydroxyecdysone (7), abutasterone (8), 2-deoxy-20-hydroxyecdysone 3-O- β -D-glucopyranoside (9), (+)-lyoniresinol-2 α -O- β -D-glucopyranoside (10) and α -D-glucopyranosyl-(2 \rightarrow 1)- α -D-glucopyranoside (11). All compounds are isolated from *T. sagittata* var. *yunnanensis* for the first time.

Cheng D.M., Yousef G.G., Grace M.H., Rogers R.B., Gorelick-Feldman J., Raskin I. and Lila M.A. (2008) In vitro production of metabolism-enhancing phytoecdysteroids from *Ajuga turkestanica*. *Plant Cell Tissue Organ Culture* 93, 73-83.

Abstract: In order to develop a sustainable source of metabolism-enhancing phytoecdysteroids, cell suspension and hairy root cultures were established from shoot cultures of wild-harvested *Ajuga turkestanica*, a medicinal plant indigenous to Uzbekistan. Precursors of phytoecdysteroids (acetate, mevalonic acid cholesterol) or methyl jasmonate (an elicitor) were added to subculture media to increase phytoecdysteroid accumulation. In cell suspension cultures, 20-hydroxyecdysone (20E) content increased 3- or 2-fold with the addition of 125 or 250 μ M methyl jasmonate, respectively, compared to unelicited cultures. Precursor addition, however, did not provoke phytoecdysteroid accumulation. In hairy root cultures, addition of sodium acetate, mevalonic acid, and methyl jasmonate, but not cholesterol, increased phytoecdysteroid content compared to unelicited cultures. Hairy root cultures treated with 150 mg l⁻¹ sodium acetate, or 15 or 150 mg l⁻¹ mevalonic acid, increased 20E content approximately 2-fold to 19.9, 20.4 or 21.7 μ g mg⁻¹, respectively, compared to control (10.5 μ g mg⁻¹). Older hairy root cultures, extracted after the seventh subculture cycle, also showed increases in 20E content (24.8 μ g mg⁻¹), turkesterone (0.9 μ g mg⁻¹) and cyasterone (8.1 μ g mg⁻¹) compared to control cultures maintained for a shorter duration of four subculture cycles. Doses of 10 or 20 μ g ml⁻¹ hairy root extract increased protein synthesis by 25.7% or 31.1%, respectively, in a C2C12 mouse skeletal cell line. These results suggest that sustainable production of metabolically active phytoecdysteroid can be achieved through hairy root culture systems.

Cheng D.M., Yousef G.G. and Lila M.A. (2010) Variation in phytoecdysteroid accumulation in seeds and shoots of *Spinacia oleracea* L. accessions. *HortScience* 45(11), 1634-1638.

Abstract: Spinach (*Spinacea oleracea* L.) is a valuable agricultural crop that accumulates phytoecdysteroids, polyhydroxylated triterpenoids, which may play a role in plant defense and have purported health benefits for human consumers. In this study, phytoecdysteroid accumulation was measured in seeds and shoots of 15 spinach accessions to determine whether phytoecdysteroid levels vary between spinach varieties and whether seed content could reliably predict relative levels in the edible foliage. Additionally, phytosterols, precursors to phytoecdysteroids, were examined to determine potential points of regulation of spinach phytoecdysteroid biosynthesis. Significant variations in phytoecdysteroid levels between accessions were observed ($P < 0.05$), suggesting the potential for genetic manipulation through traditional breeding or genetic engineering to increase phytoecdysteroid levels in spinach. However, results suggest that estimation of phytoecdysteroid levels in shoots may not be achieved by measuring levels in the seeds. Levels of phytoecdysteroids in spinach ranged from 19.9 to 44.1 μ g per shoot, 0.7 to 1.2 μ g·mg⁻¹ dry mass shoot, 3.2 to 9.6 μ g per seed, and 0.5 to 1.1 μ g·mg⁻¹ seed. Several phytosterols connected to the phytoecdysteroid biosynthetic pathway were identified by gas chromatography–mass spectroscopy, predominantly spinasterol, 5-dihydroergosterol, and 22-dihydrospinasterol, which comprised 79.8%, 6.3%, and 4.6% of the total phytosterol content, respectively. Detection of the phytosterols cycloartenol and lanosterol in spinach suggests that spinach may also have dual biosynthetic pathways to phytosterols that contribute to the production of phytoecdysteroids.

Cheng J.K., Zhang Y.H., Zhang Z.Y., Cheng D.L. and Zhang G.L. (2002) Studies on structure of ecdysterones from *Rhaponticum uniflorum*. *Chemical Journal of Chinese Universities* 23(11), 2084-2088 [in Chinese, with an abstract in English].

Abstract: Seven phytoecdysteroids were isolated from the root of *Rhaponticum uniflorum*. Their structures, by means of spectroscopic techniques and chemical properties, were identified as ajugasterone C-2, 3;20,22-diacetonide(1); 25-deoxy-9(11)-dehydro-20-hydroxyecdysone-20,22-amonocetonide(2); ajugasterone C-20,22-monoacetonide (3); ajugasterone C (4); rhapontisterone C (5); ecdysterone(6); 11 α -hydroxyecdysterone ecdysterone (7). Compounds 1 and 2' were new compounds. Compound 3 was obtained from *Rhaponticum uniflorum* for the first time.

Cheng W-x., Chen H-y., Zhang Y-p., Qin X-l. and Gu K. (2007) Chemical constituents of *Vitex quinata*. *Natural Product Research and Development* (2), 244-246 [in Chinese, with an English abstract].

Cheng Y.-X., Zhou J., Tan N.-H. and Ding Z.-T. (2001) Phytoecdysterones from *Cucubalus baccifer* (Caryophyllaceae). *Acta Botanica Sinica* 43(3), 316-318.

Abstract: Six phytoecdysterones have been isolated from the n-BuOH portion of *Cucubalus baccifer* L., a Chinese folk medicinal plant. Their structures were elucidated as ecdysterone (1), 24(28)-ecdysterone (2) [?], 22-deoxyecdysterone (3), 25-hydroxypanuosterone (4), rubrosterone (5) and 2,22-dideoxyecdysterone 3 β -O- β -D-glucopyranoside (6) on the basis of spectroscopic and chemical methods. Among them, compound 6 was a new phytoecdysterone glycoside and 1-5 were obtained from this plant for the first time.

Chi D-f., Sun M-x. and Xia W-f. (2002) Pesticidal character of phytoecdysteroids from *Ajuga multiflora* Bunge (Labiatae) on larvae of *Cryptorrhynchus lapathi* L. (Coleoptera; Curculionidae). *Journal of Forestry Research* 13, 177-182.

Abstract: Eight kinds of phytoecdysteroids extracted from different parts of *Ajuga multiflora* Bunge (Labiatae) that were collected from different places at different time were tested for killing effects on the 2-instar larvae of *Cryptorrhynchus lapathi* L by adding them to the artificial diet of larvae. The experimental results indicated that adding 1–3-mL phytoecdysteroids to the artificial diet could lead 58%–100% of 2-instar larvae of *C. lapathi* to death within 24 days. The phytoecdysteroids extracted from the whole plant of *A. multiflora* which was collected before flowering time were much more effective than those extracted from the plants collected at flowering and after flowering periods, and the modified mortality rate of larvae reached 65.22%, 85.07%, and 98.11% at the dosage level of 1-mL, 2-mL, and 3-mL extracts, respectively. The extract made from root of *A. multiflora* plant was more effective in killing efficiency than those from stem and leaves, and the average death rates of larvae were up to 100%, 98.20% and 98.32% at dosage levels of 1-mL, 2-mL, and 3-mL extracts, respectively. The killing speed of the extracted phytoecdysteroids was slower than that of triflumuron, hexaflumuron or deltamethrin emulsifiable concentrate. The mortality rate of larvae is closely related to the feeding duration on the diets containing phytoecdysteroids. Feeding on the diets with addition of phytoecdysteroids for 16 days, more than 80% of treated 2-instar larvae of *C. lapathi* were led to death. The killing effect of the extracts was little affected by the growth areas of *A. multiflora* plant and the adding way to artificial diet.

Chiang H. C., Wang J. J. and Wu R. T. (1992) Immunomodulating effects of the hydrolysis products of formosamin C and β -ecdysone from *Paris formosana* Hayata. *Anticancer Research* 12(5), 1475-1478.

Abstract: Formosanin C (PF-3, I), a diosgenin glycoside with four sugars isolated from *Paris formosana* Hayata as main constituent, significantly showed immunomodulating effects on the proliferative response of mouse lymphocytes to concanavalin A (Con A). The partial hydrolysis products of formosanin C, dioscin (II) and prosapogenin A of dioscin (III), also increased 3H-thymidine incorporation of Con A-stimulated lymphocytes maximally at 0.01 micrograms/ml, whereas formosanin C did so at 0.0001 micrograms/ml. However, trillin (IV) and diosgenin (V) obtained from the partial hydrolysis of formosanin C had no effects on these immune responses. Evidently, the immunomodulating activities increased in the order of increasing polarity. Probably the solubility in water was a factor. This demonstrated that the sugar moiety in the structure of formosanin C (I) displays a very important pattern for the effect on the proliferative response of mouse lymphocytes to Con A. On the other hand, these hydrolysis products at higher concentrations of 1 and 10 micrograms/ml reduced the cytotoxic effects on spleen cells as compared with formosanin C. The other constituent, beta-ecdysone (VI) isolated from the stems of *Paris formosana* Hayata also increased 3H-thymidine incorporation of Con A-stimulated splenocytes. At the concentration of 0.001 micrograms/ml, the stimulation index of beta-ecdysone (2) is higher than that of formosanin C (1.65), and at the concentration of 100 micrograms/ml, beta-ecdysone had no cytotoxicity for normal spleen cells whereas formosanin C at the lower concentration of 10 micrograms/ml showed cytotoxicity. Based on this study, beta-ecdysone (VI) is therefore a better immunomodulator than formosanin C.

Choi Y.H., Kim J. and Choi Y.-H. (1999) A steroidal glycoside from *Lepisorus ussuriensis*. *Phytochemistry* 51, 453-456.

Abstract: A new steroidal glycoside, 2 α ,3 β -(22R)-trihydroxycholestan-6-one-22-O- β -D-glucopyranosyl-(1 \rightarrow 2)- α -L-arabinopyranoside, was isolated from the whole plants of *Lepisorus ussuriensis*, together with α -ecdysone and ecdysterone. Their structures were determined by means of spectroscopic and chemical methods.

Chong Y.K., Galbraith M.N. and Horn D.H.S. (1970) Isolation of deoxycrustecdysone, deoxyecdysone and α -ecdysone from the fern *Blechnum minus*. Journal of the Chemical Society, Chemical Communications 1217-1218.

Abstract: The structure of deoxycrustecdysone (I) has been confirmed following its isolation from the fern *Blechnum minus*, together with α -ecdysone (III) and the new compound deoxyecdysone (II).

Chou W-S. and Lou H-S. (1980) Growth regulation and silk production in *Bombyx mori* L. from phytoenous ecdysteroids. In: Progress in Ecdysone Research (Ed. Hoffmann J.A.), Elsevier/North Holland, pp. 281-297.

Chua M. T., Santos A. C., Abela C. and Wagner U. (1982) Isolation of steroid CV-3 (C₁₉H₂₆O₅) from *Cyanotis vaga* (Lour.) Schultes. Philippine Journal of Science 111, 1-15.

Abstract: The acetone extract of *Cyanotis vaga* whole plant yielded a crystalline compound (C₁₉H₂₆O₅) m.p. 248-251 C. Spectral data (IR, UV, NMR, MS and CD) are presented, showing the substance are an ecdysone, identical with the recently elucidated rubrosterone. This represents the second plant family reported to contain a steroid having an etiocholane skeleton

Clément C.Y. and Dinan L. (1991) Development of an assay for ecdysteroid-like and anti-ecdysteroid activities in plants. In: Insect Chemical Ecology (ed. Hrdý I.), Academia Prague, pp. 221-226.

Coll J. (2007) New minor ecdysteroids from *Ajuga iva* (Labiatae) and complete ¹H-NMR assignment of cyasterone. *Affinidad* 64 242-250.

Abstract: 24-Hydroxycyasterone and ajugasterone B (newly found in this source) have been isolated from *Ajuga iva* whole plant as minor components. As already reported, the three major compounds, cyasterone, 20-hydroxyecdysone and makisterone A, were present in large amounts together with 24,25- didehydroprecyasterone, a third minor compound. All the structures have been identified from spectral analysis and the ¹H-NMR data for cyasterone are revised and completed.

Coll J., Reixach N., Sánchez-Baeza F., Casas J. and Camps F. (1994) New ecdysteroids from *Polypodium vulgare*. *Tetrahedron* 50(24), 7247-7252.

Abstract: Five phytoecdysteroids not previously reported in *Polypodium vulgare* have been isolated from methanol extracts of *in vitro* cultures of prothalli of this species. Among them, inokosterone, pterosterone and abutasterone had already been identified in other plants, whereas 24-hydroxyecdysone and 5-hydroxyabutasterone are described for the first time. The structure of these compounds was inferred from the corresponding ¹H and ¹³C NMR and thermospray-HPLC-MS spectral data. The complete description of the structures includes the stereochemical assignment at C-24, which was 24*S*, for all compounds. This assignment was carried out by ¹³C NMR spectroscopic studies of 22,24-benzylidene acetal derivatives.

Coll J., Tandrón Y.A. and Zeng X. (2007) New phytoecdysteroids from cultured plants of *Ajuga nipponensis* Makino. *Steroids* 72(3), 270-277.

Abstract: An extract from aerial parts of *Ajuga nipponensis* Makino was examined by high performance liquid chromatography for minor ecdysteroids. Along with the compounds already reported, namely cyasterone, ajugasterone C, cyasterone 22-acetate and 22-dehydrocyasterone, the presence of three additional bands with the expected ecdysteroid-like UV absorption was observed. The structures of the isolates were unambiguously elucidated based on extensive NMR spectral studies (one and two-dimensional experiments) and pointed out three new phytoecdysteroids. One of the new compounds, 22-dehydrocyasterone 2-glucoside is just the second example of a C-2 glucosyl derivative. The other two compounds displayed hemiacetal functions in the side chain, one unprecedented, and were named ajugacetalsterone A and B.

Coll Toledano J. (1998) Old and new ecdysteroids in *Ajuga* species: an overview. *Russian Journal of Plant Physiology* 45(3), 310-315 [in English]/*Fisiologiya Rastenii* 45(3) 365-371 [in Russian].

Colombo, M. L., & Tomè, F. (1993) Ecdysteroid production in *Helleborus odoratus* ssp. *laxus*: response to different environments. *International Journal of Pharmacognosy* 31(4), 311-315.

Abstract: Two ecdysteroids, 5 β -ecdysone and 5P-hydroxyecdysone, were separated from hypogeous organs of five populations of *Helleborus odoratus* ssp. *laxus* (Host) Merxm. & Podl. (Ranunculaceae). The ecdysteroid content is higher during the vegetative growth (summer) AND lower during flowering (spring) AND quiescency (autumn-

winter). Plants growing in short grass plains have an ecdysteroid content higher (30-40%) than plants from oak AND chestnut woods.

Colombo M.L., Tomè F., Servettaz O. and Bugatti C. (1990) Phytochemical evaluation of *Helleborus* species growing in Northern Italy. *International Journal of Crude Drug Research* **28**(3), 219-223.

Abstract: Hellebrin, desglucohellebrin, hellebrigenin, bufatetraenolide and traces of spirostane-types steroids have been isolated from roots and rhizomes of *Helleborus odorus* ssp. *laxus*, *Helleborus viridis* ssp. *viridis* and *Helleborus "viridis"*. In addition, β -ecdysterone and 5B-hydroxyecdysterone have been isolated for the first time from these species.

Cook IF, Lloyd-Jones JG, Rees HH and Goodwin TW (1973) The stereochemistry of hydrogen elimination from C-7 during biosynthesis of ecdysones in insects and plants. *Biochemical Journal* **136**, 135-145.

Abstract: 1. [7 α -(3)H(1)]- and [7 β -(3)H(1)]-Cholesterol were synthesized by a modified method. 2. The stereochemistry of Delta(7)-bond formation during ecdysone and ecdysterone biosynthesis in the insect, *Calliphora erythrocephala* and the plants, *Taxus baccata* and *Polypodium vulgare* was investigated by using [4-(14)C,7 α -(3)H(1)]cholesterol and [4-(14)C,7 β -(3)H(1)]cholesterol. 3. In each case, the 7 β hydrogen was stereospecifically eliminated. 4. The possible significance of the results is discussed in relation to double-bond formation in other systems and the stage at which the Delta(7) bond is introduced during ecdysone biosynthesis.

Corio-Costet M.F., Chapuis L., Mouillet J.F. and Delbecque J.P. (1993a) Sterol and ecdysteroid profiles of *Serratula tinctoria* (L): plant and cell cultures producing steroids. *Insect Biochemistry and Molecular Biology* **23**, 175-180.

Abstract: Cell suspension cultures have been obtained from *Serratula tinctoria*, a plant producing ecdysteroids. Sterol profiles and ecdysteroid contents have been analysed and compared in plants and cell cultures. In particular, the composition of free and esterified sterols was investigated using appropriate analytical techniques.

In plants, esterified sterols were more abundant (50–70% of the total sterol) than in cell cultures (13–36%). A selectivity for sterol esterification was noted: in plants, the triterpenes (as amyryns) were esterified, whereas it was the 4-desmethylsterols (sitosterol and cholesterol) in cell cultures.

Ecdysteroids were present in higher quantities in plant (0.1–1.2% dry wt) than in cell cultures (0.01–0.03%).

Analysis confirmed the presence of 20-hydroxyecdysone, 20-hydroxyecdysone-3-acetate and polypodine B as the main compounds. Roots were not only the richest organ in ecdysteroids, but also in cholesterol, lathosterol and 24-methylene-cholesterol. The results obtained suggest interesting relationships between free or esterified sterols and ecdysteroid contents.

Corio-Costet M.-F., Chapuis L., Scalla R. and Delbecque J.-P. (1993b) Analysis of sterols in plants and cell cultures producing ecdysteroids: I. *Chenopodium album*. *Plant Science* **91**, 23-33.

Abstract: Cell cultures have been obtained in vitro from a plant producing ecdysteroids, *Chenopodium album*. The composition of free and esterified sterols was compared in plant and cell cultures after purification by TLC and identification by GC/MS. Free sterols were the most abundant. A selectivity of sterol esterification was noted for 4,4-dimethylsterols (except in roots) and Δ 7-sterols. The 4-desmethylsterols from leaves, roots and cell cultures of *Chenopodium album* were a mixture of Δ 7-sterols (>61%) and Δ 5-sterols (>36%). The Δ 7-sterols were 24-ethylcholesta-7,22-dien-3 β -ol and 24-ethylcholest-7-en-3 β -ol. The isolated Δ 5-sterols were sitosterol and stigmasterol. Traces of cholesterol and Δ 5,7-sterols were also detected. Our attention was also given to ecdysteroid contents, which showed much lower quantity in cell cultures than in plants, confirming however the presence of 20-hydroxyecdysone.

Corio-Costet M.-F., Chapuis L. and Delbecque J.-P. (1996) *Serratula tinctoria* (Dyer's savory): *in vitro* culture and the production of ecdysteroids and other secondary metabolites. *Biotechnology in Agriculture and Forestry* **37**: Medicinal and Aromatic Plants IX (Ed. Bajaj Y.P.S.) Springer Verlag, pp. 384-401.

Corio-Costet M.F., Chapuis L. and Delbecque J.-P. (1998) *Chenopodium album* L. (Fat Hen): *in vitro* cell culture, and production of secondary metabolites (phytosterols and ecdysteroids). *Biotechnology in Agriculture and Forestry* **41**: Medicinal and Aromatic Plants X (Ed. Bajaj Y.P.S.) Springer Verlag, pp. 97-112.

Corio-Costet M.-F., Chapuis L., Delbecque J.-P. and Ustache K. (1999) Genetic transformation of *Serratula tinctoria* (Dyer's Savory) for ecdysteroid production. *Biotechnology in Agriculture and Forestry* **45**: Transgenic Medicinal Plants (Ed. Bajaj YPS), Springer-Verlag, Berlin/Heidelberg, pp.284-297.

Correa J.P.O., Vital C.E., Pinheiro M.V.M., Batista D.S., Saldanha C.W., Ferreira da Cruz A.C., Notini M.M., Freitas D.M.S., DaMatta F.M. and Otoni W.C. (2016) Induced polyploidization increases 20-hydroxyecdysone content, in

vitro photoautotrophic growth, and ex vitro biomass accumulation in *Pfaffia glomerata* (Spreng.) Pedersen. In *Vitro Cellular Development and Biology* **52**, 45-55.

Abstract: The present study aimed to verify the effects of induced polyploidization on *Pfaffia glomerata* regarding its 20-hydroxyecdysone (20E) production both *in vitro* and under greenhouse conditions, its *in vitro* photoautotrophic potential, and its *ex vitro* biomass accumulation and photosynthetic performance. Synthetic polyploidization efficiently produced individuals with increased *in vitro* photoautotrophic potential and *ex vitro* biomass accumulation, although photosynthetic rates per leaf area did not vary between diploids and tetraploids. Among the five tetraploids tested (P28, P60, P68, P74, and P75), P28 showed significantly increased biomass both *in vitro* and *ex vitro* when compared with diploid plants, whereas the other tetraploids did not differ significantly from the diploids in terms of biomass accumulation. Although photosynthetic rates per unit leaf area remained constant among all the plants tested, P28 showed a significantly greater total leaf area, which may have resulted in an increase in net photosynthesis on a whole-plant basis. Under greenhouse conditions, the 20E content in the tetraploid P28 was 31% higher than that in diploid plants, and the final 20E mass per plant produced by P28 *ex vitro* was approximately twice that produced by diploid plants. Accumulation of 20E *in vitro* did not follow the same pattern observed among the plants *ex vitro*; instead, greater accumulation was observed in diploid plants. The induction of polyploidy in *P. glomerata* appears to be a promising strategy for producing plants with higher biomass accumulation and 20E production *ex vitro*, in addition to its higher *in vitro* photoautotrophic potential.

Crouzet S., Maria A., Dinan L., Lafont R. and Girault J.-P. (2009) Ecdysteroids from *Cyanotis longifolia* Benth. (Commelinaceae). *Archives of Insect Biochemistry and Physiology* **72**(4), 194-209.

Abstract: *Cyanotis longifolia* Benth. (Comelinaceae) contains ecdysteroids, which are highly concentrated in the roots and flowers, whereas leaves contain only very low amounts and stems intermediate amounts. 20-Hydroxyecdysone is the major component found in all tissues, but roots also contain large amounts of 20-hydroxyecdysone 3-acetate and ajugasterone C. A preparative experiment has shown that roots contain a complex ecdysteroid mixture, and the analysis of minor components has allowed the isolation of several already known ecdysteroids (polypodine B, 2-deoxy-20,26-dihydroxyecdysone, isovitexirone, poststerone) together with five new (ajugasterone C 3-acetate, 5beta-hydroxy-poststerone, poststerone 2-acetate, 14(15)-dehydro-poststerone 2-acetate, 24-epi-atrosterone A [=24-methyl-ajugasterone C]) ecdysteroids that have been fully characterized. A preliminary investigation of 55 species belonging to 15 different genera of the Commelinaceae has shown that several of them contain significant concentrations of ecdysteroids, among which some previously uninvestigated ones appear to be very promising sources of ecdysteroids.

Cui S.Y., Chen X.G. and Hu Z. (2003) Identification and determination of ecdysone and phenylpropanoid glucoside and flavonoids in *Lamium maculatum* by capillary zone electrophoresis. *Biomedical Chromatography* **17**(7), 477-482.

Abstract: The simultaneous determination of 20-hydroxy ecdysone (1), 3,7-dimethoxy-quercetin (2), acteoside (3) and rutin (4) in the mixture of leaf and stem, and the flower of *Lamium maculatum* has been investigated by capillary zone electrophoresis for the first time. With an electrolyte containing 30 mmol/L borate, at pH 9.47 and 20 kV applied voltage, the four active compounds were completely separated within 5 min with satisfactory results. The effects of concentration of borate and electrolyte pH on electrophoretic behavior and separation were studied. Regression equations revealed linear relationships (correlation coefficients 0.9998-0.9999) between the peak area of each analyte and the concentration. The levels of analytes in the different parts of *Lamium maculatum* were easily determined with recoveries ranging from 98.3 to 105.0%.

Dai H., Liu Y., Deng S., Tan N. and Zhou J. (2002) Study on the chemical constituents of *Lychnis coronaria*. *Natural Product Research and Development* (1), 9-12 [in Chinese].

Abstract; Eleven known compounds, tricin 7-O-glucopyranoside, (+)-isoscoparin, epoxyactinidionoside, 1 α ,20R-hydroxyecdysone, ecdysterone, polypodine B, ecdysterone 22-O- β -D-glucopyranoside, stigmast-5-ene-3-one, taraxerol, α -tocopherol and dehydrodiconiferyl alc. 4-O- β -D-glucopyranoside were isolated from the ethanolic extract of *Lychnis coronaria* (L.) Desr. Their structures were elucidated by spectral evidence (IR, NMR, MS, etc.).

Dai J.Q., Cai Y.J., Shi Y.P., Zhang Y.H., Liu Z.L., Yang L. and Li Y. (2002) Antioxidant activity of ecdysteroids from *Serratula strangulata*. *Chinese Journal of Chemistry* **20**(5), 497-501.

Abstract: One new ecdysteroid, (24R)-24-(2-hydroxyethyl)-20-hydroxyecdysone (3), as well as three known ecdysteroids, has been isolated from Chinese herb *Serratula strangulata* and these compounds 1-4 showed effective antioxidant activity on AAPH-induced hemolysis of human RBC and Fe²⁺ + cysteine-induced lipid peroxidation of liver microsomes.

Dai J.Q., Zhu Q.X., Zhao C.Y., Yang L. and Li Y. (2001) Glyceroglycolipids from *Serratula strangulata*. *Phytochemistry* **58**(8), 1305-1309.

Abstract: The rhizomes of *Serratula strangulata* yielded three glyceroglycolipids, i.e. 1,2-di-*O*-(9*Z*,12*Z*,15*Z*-octadecatrienoyl)-3-*O*-(6-amine-6-deoxy- α -D-glucosyl)-glycerol, 1,2-di-*O*-(9*Z*,12*Z*,15*Z*-octadecatrienoyl)-3-*O*-(6-*p*-hydroxy-phenyl-propionamido-6-deoxy- α -D-glucosyl)-glycerol and 1,2-di-*O*-(9*Z*,12*Z*,15*Z*-octadecatrienoyl)-3-*O*-[α -D-glucose(1-6)- β -D-allose]-glycerol, as well as a known sesquiterpene lactone and three known phytoecdysones. Their structures were elucidated on the basis of spectral data, especially by 2D NMR spectroscopic methods and chemical conversion. These compounds exhibited significant antibacterial and antitumor activities. Three compounds, 1,2-di-*O*-(9*Z*,12*Z*,15*Z*-octadecatrienoyl)-3-*O*-(6-amine-6-deoxy- α -D-glucosyl)-glycerol (**1**), 1,2-di-*O*-(9*Z*,12*Z*,15*Z*-octadecatrienoyl)-3-*O*-(6-*p*-hydroxy-phenyl-propionamido-6-deoxy- α -D-glucosyl)-glycerol (**2**) and 1,2-di-*O*-(9*Z*,12*Z*,15*Z*-octadecatrienoyl)-3-*O*-[α -D-glucose(1-6)- β -D-allose]-glycerol (**3**) were isolated from the rhizome of *Serratula strangulata*. Their antibacterial and antitumor activities were evaluated.

Dai J.R., Chai H., Pezzuto J.M., Kinghorn A.D., Tsauri S. and Padmawinata K. (1993) Studies on Indonesian plants. V. Cytotoxic constituents of the roots of the Indonesian medicinal plant *Fibraurea chloroleuca*. *Phytotherapy Research* **7**(4) 290-294.

Abstract: Three protoberberine alkaloids, berberine chloride, berberrubine chloride and thalifendine chloride were isolated from the roots of *Fibraurea chloroleuca* Miers, and found to show significant cytotoxic activity with one or more human cancer cell-lines and cultured P-388 cells. A phytoecdysteroid, 20-hydroxyecdysone, was also isolated as a noncytotoxic compound. Based on comparison with previous investigations, different collections of *F. chloroleuca* roots appear to exhibit considerable variation in their alkaloidal profiles.

Daniel M. and Mammen D. (2014) Ecdysterone and antioxidants in purslane (*Portulaca oleracea* Linn.). *Asian Journal of Chemical and Pharmaceutical Research* **2**(2) 137-139.

Abstract: Purslane (*Portulaca oleracea* Linn.), a common weed of great importance as a nutraceutical, was studied for its steroids and phenolics. The methanolic extract is found to contain up to 0.532 mg (in 100g of dry powder) of ecdysterone, in addition to β -sitosterol. Quercetin was present in traces and the phenolic acids present were vanillic and syringic acids. Both betacyanins and betaxanthins also were located. The discovery of appreciable amount of the anabolic steroid ecdysterone having a number of pharmacologically significant properties in elevating the status of purslane as a nutraceutical of great potential is discussed.

Darmogray V.N., Erofeeva N.S., Darmogray S.V., Filippova A.S., Morozova V.A. and Dubodelova G.V. (2015) Qualitative and quantitative determination of polyphenolic and ecdysteroid compounds from earflaps (*Otites parviflorus* Grossh.). *Advances in Current Natural Sciences* (12) 21-25 [in Russian, with an abstract in English].

Abstract: The chemical composition of the earflaps (*Otites parviflorus* Grossh.) polyphenolic and steroid compounds was studied. As a result of examination of the sample by high performance liquid chromatography (HPLC) identified 14 compounds of polyphenol nature and 2 ecdysteroid substances (ecdysterone, polipodin B). Actoprotective effect of ecdysterone and aqueous-alcoholic extract of the herb *Otites parviflorus* was studied in the article, and their actoprotective effect were close in value.

Darvas B. (1991) Phytoecdysteroids of *Ajuga* spp., as insect growth regulator-type botanical insecticides. *Novenyvedelem* **27**(11/12), 481-498 [in Hungarian, with an English abstract].

da Rosa H.S., Salgueiro A.C.F., Colpo A.Z.C., Paula F.R., Mendez A.S.L. and Folmer V. (2016) *Sida tuberculata* (Malvaceae): a study based on development of extractive system and *in silico* and *in vitro* properties. *Brasilian Journal of Medical and Biological Research* **49**(8), e5282 (DOI.org/10.1590/1414-431X20165282).

Abstract: *Sida tuberculata* (Malvaceae) is a medicinal plant traditionally used in Brazil as an antimicrobial and anti-inflammatory agent. Here, we aimed to investigate the different extractive techniques on phytochemical parameters, as well as to evaluate the toxicity and antioxidant capacity of *S. tuberculata* extracts using *in silico* and *in vitro* models. Therefore, in order to determine the dry residue content and the main compound 20-hydroxyecdysone (20E) concentration, extracts from leaves and roots were prepared testing ethanol and water in different proportions. Extracts were then assessed by *Artemia salina* lethality test, and toxicity prediction of 20E was estimated. Antioxidant activity was performed by DPPH and ABTS radical scavenger assays, ferric reducing power assay, nitrogen derivative scavenger, deoxyribose degradation, and TBARS assays. HPLC evaluation detected 20E as main compound in leaves and roots. Percolation method showed the highest concentrations of 20E (0.134 and 0.096 mg/mL of extract for leaves and roots, respectively). All crude extracts presented low toxic potential on *A. salina* (LD₅₀ >1000 μ g/mL). The computational evaluation of 20E showed a low toxicity prediction. For *in vitro* antioxidant tests, hydroethanolic extracts of leaves were most effective compared to roots. In addition, hydroethanolic extracts presented a higher IC₅₀ antioxidant than aqueous extracts. TBARS formation was prevented by leaves hydroethanolic extract from 0.015 and 0.03 mg/mL and for roots from 0.03 and 0.3 mg/mL on egg yolk

and rat tissue, respectively ($P < 0.05$). These findings suggest that *S. tuberculata* extracts are a considerable source of ecdysteroids and possesses a significant antioxidant property with low toxic potential.

da Rosa H.S., Coehlo I.S., da Silva M.D., Fernandes M.S., Bertelli P.R., Minetto L., Moura S., de Paula F., Santos A.R., Mendez A.S.L. and Folmer V. (2018a) *Sida tuberculata* extract reduces the nociceptive response by chemical noxious stimuli in mice: implications for mechanism of action, relation to chemical composition and molecular docking. *Phytotherapy Research* pp 10 (doi: 10.1002/ptr.6220).

Abstract: *Sida tuberculata* R.E.Fr. (Malvaceae) is a medicinal plant widely found in Southern Brazil, and popularly used for inflammatory disorders and to pain relief. A phytochemical analysis followed by an investigation about antinociceptive potential and mechanism of action were performed with leaves and roots extracts. Methanolic extracts, designated as *S. tuberculata* leaves extract (STLE) and *S. tuberculata* roots extract, were analyzed both by UHPLC–MS. The in vivo antinociceptive potential of STLE (10–300 mg kg⁻¹) was assessed in mice subjected to the acetic acid-induced abdominal writhes and formalin model. Agonist/antagonist tests and computational docking suggest the involvement of opioid and adenosinergic systems. The main chemical class detected on extracts was the ecdysteroids, and 20-hydroxyecdysone (20HE) was confirmed as the major phytoconstituent. The pretreatment with STLE (100 mg kg⁻¹) reduced more than 70% abdominal contortions induced by acetic acid model and produced significant inhibition on formalin-induced licking response. The mechanism of action study revealed STLE might act through opioid and adenosine systems. Molecular docking suggested kaempferol derivative and 20HE might interacting with μ -opioid receptor. Thus, the results suggest the existence of antinociceptive potential from *S. tuberculata* extracts being in accordance to the traditional use.

da Rosa H.S., Koetz M., Santos M.C., Jandrey E.H.F., Folmer V., Henriques A.T. and Mendez A.S.L. (2018b) Extraction optimization and UHPLC method development for determination of the 20-hydroxyecdysone in *Sida tuberculata* leaves. *Steroids* 132, 33-39.

Abstract: *Sida tuberculata* (ST) is a Malvaceae species widely distributed in Southern Brazil. In traditional medicine, ST has been employed as hypoglycemic, hypocholesterolemic, anti-inflammatory and antimicrobial. Additionally, this species is chemically characterized by flavonoids, alkaloids and phytoecdysteroids mainly. The present work aimed to optimize the extractive technique and to validate an UHPLC method for the determination of 20-hydroxyecdysone (20HE) in the ST leaves. Box-Behnken Design (BBD) was used in method optimization. The extractive methods tested were: static and dynamic maceration, ultrasound, ultra-turrax and reflux. In the Box-Behnken three parameters were evaluated in three levels (-1, 0, +1), particle size, time and plant:solvent ratio. In validation method, the parameters of selectivity, specificity, linearity, limits of detection and quantification (LOD, LOQ), precision, accuracy and robustness were evaluated. The results indicate static maceration as better technique to obtain 20HE peak area in ST extract. The optimal extraction from surface response methodology was achieved with the parameters granulometry of 710 nm, 9 days of maceration and plant:solvent ratio 1:54 (w/v). The UHPLC-PDA analytical developed method showed full viability of performance, proving to be selective, linear, precise, accurate and robust for 20HE detection in ST leaves. The average content of 20HE was 0.56% per dry extract. Thus, the optimization of extractive method in ST leaves increased the concentration of 20HE in crude extract, and a reliable method was successfully developed according to validation requirements and in agreement with current legislation.

da Silva Souza L.D.Z., da Fonseca S.R.A.V., Ferrari A. and Felipe D.F. (2021) β -Ecdysone content and antioxidant capacity in different organs of Brazilian ginseng. *Ciência Rural*, Santa Maria 51(5), e20200618, pp5.

ABSTRACT: Plants that contain antioxidant compounds have attracted increasing interest for their vital role in the attenuation of oxidative damage caused by free radicals and in the treatment of various diseases. The present study investigated the β -ecdysone content and the antioxidant activity of Brazilian ginseng (*Pfaffia glomerata*) extracts obtained from inflorescences, stems, and roots. The *P. glomerata* extracts were tested for antioxidant activity using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging method, β -carotene bleaching test, and phosphomolybdenum method. The β -ecdysone content of *P. glomerata* extracts was measured by high-performance liquid chromatography (HPLC). The *P. glomerata* inflorescences showed the strongest DPPH radical scavenging activity and the strongest antioxidant activity in the β -carotene bleaching assay and phosphomolybdenum test. The roots showed the lowest antioxidant capacity in all of the assays. The concentration of β -ecdysone in the plant organs followed the following decreasing order: inflorescences > stems > roots. The present study showed that *P. glomerata* inflorescence extract had high antioxidant capacity that could be attributed to the presence of β -ecdysone.

Darwish F.M.M. and Reinecke M.G. (2003) Ecdysteroids and other constituents from *Sida spinosa* L. *Phytochemistry* 62(8), 1179-1184.

Abstract: Two compounds (3 and 10) were isolated from the aerial parts of *Sida spinosa* L. Their structures have been established as glyceryl-1-eicosanoate and 20-hydroxy, 24-hydroxymethylecdysone by 1D and 2D-NMR techniques. In addition 12 known compounds (1, 2, 4-9 and 11-14) have been isolated and identified.

Das N., Saha T. and Bhattacharjee S. (2014) A new biologically active ecdysteroid from the aerial parts of *Sida glutinosa*. *Journal of Pharmacognosy and Phytochemistry* **3**(5) 73-78.

Abstract: The aerial parts of *Sida glutinosa* have been analyzed for their chemical constituents, resulting in the isolation of a new ecdysone, named glutinosterone (1). The structure of glutinosterone was determined on the basis of extensive spectroscopic analysis, including 2D NMR data and the results of hydrolytic cleavage. The structure of glutinosterone was elucidated as 1 β -hydroxy 24(28)-dehydromakisterone A. The *in vitro* biochemical analysis of compound 1 showed a significant effect on different enzymes involved in human liver function along with lipid metabolic enzymes. The compound 1 also exhibits moderate to significant anti-bacterial property.

Dat L.D., Ngoc N.T., Diep C.N., Thao N.P., Anh H.L.T., Nam N.H., Cuong N.X., Vy N.T., Kim Y.H., Ban N.K., Kiem P.V. and Minh C.V. (2012) Ecdysteroids from *Achyranthes bidentata*. *Vietnam Journal of Chemistry* **50**(5A), 254-258.

Abstract: Five ecdysteroids, makisterone A (1), 24-epi-makisterone A (2), 24-methylene-shidasterone (3), polypodine B (4), and 20-hydroxy-ecdysone (5), were isolated from the methanol extract of *Achyranthes bidentata* by various chromatographic methods. Their structures were elucidated by spectroscopic methods including nuclear magnetic resonance (NMR) and electrospray ionization mass spectrometry (ESI-MS), and comparison of their spectral data with reported values. Compounds 1-4 were isolated from *A. bidentata* for the first time.

Davies T.G., Lockley W.J.S., Boid R., Rees H.H. and Goodwin T.G. (1980) Mechanism of formation of the A/B *cis* ring junction of ecdysteroids in *Polypodium vulgare*. *Biochemical Journal* **190**, 537-544.

Abstract: 1. The fates of the alpha-, 4 alpha- and 4 beta-hydrogen atoms of cholesterol during formation of the A/B *cis* ring junction of ecdysteroids was investigated by administration of [4-14C, 3 alpha-3H], [4-14C, 4 alpha-3H]- and [4-14C, 4 beta-3H]cholesterol species to the fern, *Polypodium vulgare*, and isolation of the 20-hydroxyecdysone formed in each case. 2. The 3H was retained in the ecdysteroid formed from each substrate. 3. Location of the 3H in the 20-hydroxyecdysone indicated that migration of 3H from the 3 alpha- and 4 beta-positions to C-4 and C-5, respectively, had occurred, whereas the 4 alpha-3H atom was retained at C-4. 4. A possible mechanism for the formation of the A/B *cis* ring junction of ecdysteroids in *P. vulgare* is presented.

Davis P., Lafont R., Large T., Morgan E.D. and Wilson I.D. (1993) Micellar capillary electrophoresis of the ecdysteroids. *Chromatographia* **37**(1/2), 37-42.

Abstract: The technique of micellar capillary electrophoresis has been applied to the separation of ecdysteroids (polyhydroxylated insect hormones) both as pure standards and in the extracts of plants and insect eggs. Successful separations of a range of closely related ecdysteroids were obtained, however, the quality of the result was found to be critically dependent on the solvent in which the sample was applied and the degree of purity of the extracts. The technique was found to be suitable for the analysis of purified ecdysteroid-rich extracts of plants (*Silene nutans*, *S. otites*) and the eggs of the desert locust (*Schistocerca gregaria*). The elution order of the analytes was similar to that obtained using reversed-phase HPLC.

Debien I.C.N and Meireles M.A.A. (2014) Supercritical fluid extraction of beta-ecdysone from Brazilian ginseng (*Pfaffia glomerata*) roots. *Food and Public Health* **4**(2), 67-73.

Abstract: Ecdysteroids have been found in a variety of plants and have several valuable biological properties. Beta-ecdysone is the major biologically active ecdysteroid that can be isolated from *Pfaffia glomerata* roots. *Pfaffia glomerata* is a medicinal plant known in Brazil as "Brazilian ginseng," and this plant has been used as a substitute for "Asian" ginseng due their similar morphologies and bioactive properties. In this work, supercritical fluid extraction (SFE) using carbon dioxide (CO₂) was used to obtain beta-ecdysone-rich extracts from Brazilian ginseng roots. The effects of pressure (20 and 30 MPa) and cosolvent amount (10, 15, 75 and 90% of Ethanol, EtOH) on the behavior of the overall extraction curve (OEC) and beta-ecdysone content were studied. Larger amounts of beta-ecdysone were obtained in shorter processing times using CO₂: EtOH (85:15, v/v) as the extracting solvent at 20 MPa. Higher amounts of EtOH in the solvent mixture leads to increased extraction yield as well as increased beta-ecdysone content.

Delbecq J-P., Beydon P., Chapuis L. and Corio-Costet M-F. (1995) *In vitro* incorporation of radiolabelled cholesterol and mevalonic acid into ecdysteroid by hairy root cultures of a plant, *Serratula tinctoria*. *European Journal of Entomology* **92**, 301-307.

Abstract: After the transformation of stems with *Agrobacterium rhizogenes*, hairy root cultures have been obtained from *Serratula tinctoria* (Asteraceae), a plant containing very high amounts of 20-hydroxyecdysone (20E) and numerous other phytoecdysteroids. These cultures were found to grow regularly *in vitro*, either in liquid or solid media, and to maintain a high ecdysteroid production (c.a. 0.1-0.2% dry weight). 20E was the predominant ecdysteroid, together with the corresponding 3-acetate (20E3Ac). Ecdysteroids were not secreted in the medium but

remained in root tissues. Moreover, a concentration gradient was observed in transformed hairy roots, as in plant roots, characterized by a higher ecdysteroid content in the meristematic zone. In vitro incubations with two radiolabelled precursors, [³H]-cholesterol and [¹⁴C]-mevalonic acid, have been performed, showing an efficient incorporation of these molecules into ecdysteroids: with labelled cholesterol, 20E indeed reached 0.25 to 0.67% of the radioactivity recovered in hairy roots after several days of culture and, with labelled mevalonate, 0.4 to 2.7%. Thus, hairy roots of *S. tinctoria* undoubtedly constitute a very promising tool for the analysis of ecdysteroid biosynthesis and functions in plants.

DellaGreca M., D'Abrosca B., Fiorentino A., Previtera L. and Zarrelli A. (2005) Structure elucidation and phytotoxicity of ecdysteroids from *Chenopodium album*. *Chemistry & Biodiversity* **2**(4), 457-462.

Abstract: The leaves of *Chenopodium album* were infused in H₂O/MeOH. The extract treated with cold acetone gave heavy precipitation, which was removed by centrifugation. Solid material was fractionated into acidic and neutral fractions. The acidic material was subjected to different silica-gel column chromatographies, and then it was purified by reversed-phase HPLC to afford four known ecdysteroids and the new 3 β ,14 α -dihydroxy-5 β -pregn-7-ene-2,6,20-trione that were characterized by extensive spectroscopic investigation, especially by 1D- and 2D-NMR. Their effects on germination and growth of *Lactuca sativa* L. have been studied. The results are reported as percentage differences of germination, root elongation and shoot elongation, from the control at concentrations ranging from 10⁻⁴ to 10⁻⁷ M.

Deng Y., He L., Li W. and Wang H. (2003) Studies on chemical constituents in herb of *Lamium maculatum* L. var. *kansuense*. *China Journal of Chinese Materia Medica* (8), 730-732 [in Chinese].

Abstract: The chem. constituents from *Lamium maculatum* var. *kansuense* were studied. The chem. constituents were isolated and repeatedly purified by silica gel column chromatog. and the structures were elucidated by NMR spectra and physicochem. properties. Ten compounds were obtained and they were identified as D-mannitol, β -sitosterol, stigmaterol, rutin, 3'-methylquercetin-3-O-rutinoside, N-butyl- β -D-fructopyranoside, daucosterol, acteoside, 20-hydroxyecdysone, and allantoin. All the compounds were obtained from *L. maculatum* var. *kansuense* for the first time.

Deng Y., Ding L., Wu S. and Wang H. (2005) Studies on chemical constituents in herb of *Lamium maculatum* var. *kansuense* (II). *Zhongguo Zhongyao Zazhi* **30**(4), 272-274 [in Chinese].

Abstract: The chem. constituents from *Lamium maculatum* var. *kansuense* were studied. The chem. constituents were isolated and repeatedly purified on silica gel column and the structures were elucidated by the NMR spectra and physico-chem. properties. Six compounds were obtained and identified as polypodine B (I), 5-hydroxy-8-epiloganin (II), shanzhiside Me ester (III), liriodendrin (IV), quercitroside (V), uridine (VI). Compound IV was found from genus *Lamium* for the first time and the rest of the compounds were found from *Lamium maculatum* var. *kansuense* for the first time.

de Sá Barreto L.C.L., Xavier H.S., Barbosa-Filho J.M. and Braz-Filho R. (2005) Ecdisteróidi e iridóide glicosilado de *Vitex gardneriana* Schauer (Verbenaceae) [Ecdysteroids and iridoid glycosides from *Vitex gardneriana* Schauer (Verbenaceae)]. *Revista Brasileira de Farmacognosia* **15**(1), 51-54 [in Portuguese, with and English abstract].

Abstract: The methanolic extract of the stem bark of *Vitex gardneriana* Schauer afforded the ecdysteroid, 20-hydroxy-ecdysone and the iridoid glycoside, aucubin. Their structures were established on the basis of chemical and spectroscopic studies.

De Souza N.J., Ghisalberti E.L., Rees H.H. and Goodwin T.W. (1969) Studies on insect moulting hormones: biosynthesis of ponasterone A and ecdysterone from [2-¹⁴C]mevalonate in *Taxus baccata*. *Biochemical Journal* **114**, 895-896.

No Abstract

De Souza N.J., Ghisalberti E.L., Rees H.H. and Goodwin T.W. (1970) Studies on insect moulting hormones: biosynthesis of ecdysone, ecdysterone and 5 β -hydroxyecdysterone in *Polypodium vulgare*. *Phytochemistry* **9**, 1247-1252.

Abstract: The incorporation of [2-¹⁴C]-mevalonic acid into ecdysterone, and of [4-¹⁴C]-cholesterol into ecdysone, ecdysterone and 5 β -hydroxyecdysterone in *Polypodium vulgare* has been demonstrated.

Devarenne T.P., Sen-Michael B. and Adler J.H. (1995) Biosynthesis of ecdysteroids in *Zea mays*. *Phytochemistry* **40**(4), 1125-1131.

Abstract: The incubation of several radiolabelled substrates with *Zea mays* plants demonstrated the incorporation of radiolabels into ecdysteroid conjugates. Radiolabelled [¹⁴C]ecdysone and [¹⁴C]20-hydroxyecdysone, were biosynthesized from [2-¹⁴C]mevalonic acid (MVA) in *Z. mays*. These ecdysteroids can be released from their

conjugates by treatment with wheat germ acid phosphatase or mild acid hydrolysis. The C-24 reduced side chain sterols, lathosterol and cholesterol were endogenously biosynthesized from [2-¹⁴C]MVA and were identified by ¹H NMR. Lathosterol accounts for *ca* 0.5% of the total sterol composition, whereas cholesterol is present at 2.5%. However, lathosterol was consistently found to have a specific activity three to six times that of cholesterol. Incubation of [4-¹⁴C]cholesterol with *Z. mays* leaves also demonstrated incorporation into a 20-hydroxyecdysone conjugate. Incubation of [22,23-³H]ecdysone with *Z. mays* demonstrated incorporation into radiolabelled ecdysone diphosphate and ecdysone polyphosphate, which were cleaved upon acid phosphatase treatment. *Z. mays* biosynthesizes primarily ecdysteroid conjugates and does not appear to produce detectable levels of non-conjugated ecdysteroids.

Dinan L. (1992a) The association of phytoecdysteroids with flowering in fat hen, *Chenopodium album*, and other members of the Chenopodiaceae. *Experientia* **48**, 305-308.

Abstract: Very high concentrations of ecdysteroid are associated with flowering in *Chenopodium album*. Highest concentrations are found in anthers, but significant levels are also found in the stamens, carpels and sepals. In contrast, pollen contains only low levels. The ecdysteroid profile is the same in anthers as in whole-plant extracts, with 20-hydroxyecdysone and polypodine B predominating. The results for flowers of *C. album* are compared with the patterns determined for other chenopods (*C. capitatum*, *C. polyspermum*, *C. anthelminticum*, *C. giganteum*, *C. quinoa* and *C. foliosum*). The significance of these findings for plant-insect interactions and the relationship to the mode of plant pollination are discussed.

Dinan L. (1992b) The analysis of phytoecdysteroids in single (preflowering stage) specimens of fat hen, *Chenopodium album*. *Phytochemical Analysis* **3**, 132-138.

Abstract: A simple microextraction procedure together with an ecdysteroid-specific radioimmunoassay (RIA) have been used to quantify the levels of phytoecdysteroid in small (<50 mg dry wt.) samples of plant tissues during the development of *Chenopodium album*. The existence of a distinct concentration gradient of phytoecdysteroids within the aerial portions has been confirmed by the detailed analysis of ecdysteroid titres throughout single plants. Ecdysteroid concentration increased steadily up the main stem of the plant. This pattern was reflected in the main leaves and in the petioles associated with the main leaves. The ecdysteroid concentration in the growing tip was higher than those at the top of the stem or in the uppermost leaves. High concentrations were also present in the roots. The distinct concentration gradient characteristic of the main stem, associated leaves and growing tip recurred in each of the side-shoots on a mature plant. The identity of the RIA-positive material is essentially identical in most parts of the plant, being mainly 20-hydroxyecdysone (20OHE) and polypodine B (PolB; 5 β ,20-dihydroxyecdysone) with much smaller amounts of more polar and less polar unidentified compounds. In the oldest leaves, the contribution of 20OHE and PolB is reduced and new polar and apolar compounds are found, indicating that catabolism of phytoecdysteroids occurs as leaves begin to senesce.

Dinan L. (1992c) Phytoecdysteroids and insect-plant relationships in the Chenopodiaceae. *Proceedings of the 8th International Symposium on Insect-Plant Relationships* (Eds. Menken S.B.J., Visser J.H. and Harrewijn P.), Kluwer Academic Publishers, pp. 86-88.

Dinan L. (1994) Phytoecdysteroids in *Kochia scoparia* (burning bush). *Journal of Chromatography A* **658**, 69-76.

Dinan L. (1995a) A strategy for the identification of ecdysteroid receptor agonists and antagonists from plants. *European Journal of Entomology* **92**, 271-283.

Abstract: A strategy is described for receptor-based phytochemical screening of plant extracts for ecdysteroid agonists and antagonists. Milligram amounts of seed are methanol extracted. Lipids and pigments are removed by hexane partitioning. Agonist and antagonist activities are detected with a microplate-based specific bioassay using the *Drosophila melanogaster* ecdysteroid-responsive BII cell line. Extracts are also screened with ecdysteroid-specific RIAs to identify extracts containing phytoecdysteroids. Over 1,700 species of plant have been screened in this way so far. Extracts are being sought which (i) contain large amounts of phytoecdysteroid, (ii) which contain novel phytoecdysteroids, (iii) which contain non-steroidal agonists and (iv) which contain antagonists. The aim of this paper is to describe the rationale behind the strategy, to describe its operation and to present, as an example, the results obtained with members of the Poaceae and of one genus, *Briza*, in particular. It is hoped that this approach will result in the identification of new sources of phytoecdysteroids, provide new phytoecdysteroid structures for structure/activity relationship studies, throw light on the phylogenetic distribution of phytoecdysteroids in the plant kingdom and provide useful agonists and antagonists for the investigation of ecdysteroid receptor function and as potential lead compounds for new classes of insect control agents.

Dinan L. (1995b) Distribution and levels of phytoecdysteroids within individual plants of species of the Chenopodiaceae. *European Journal of Entomology* **92**, 295-300.

Abstract: Radioimmunoassay was used to quantify the levels of ecdysteroids in extracts of portions of single plants of several members of the Chenopodiaceae. Species were chosen because previous studies had shown that seeds of these species were ecdysteroid-containing and because they represent several tribes within the Chenopodiaceae. Total ecdysteroid levels varied considerably between species and between different parts of the same plant. Phytoecdysteroid-containing members of the genus *Chenopodium* appear to possess the same distribution as previously found for *C. album*, as does *Spinacia oleracea*. *Rhagodia candolleana* is characterized by very high levels of ecdysteroids, with the highest levels being associated with newly developing side-shoots. Preliminary data for *Beta patellaris* reveal high levels associated with the lower portions of the plant and rapidly increasing levels associated with the reproductive tissues during flowering and fruiting, to give the high levels previously found to be associated with the seed. Taken together these data tend to support a role for phytoecdysteroids in insect deterrence, but the situation is complex, probably reflecting the subtle interplay between the plant and detrimental/beneficial insects which has occurred during evolution.

Dinan L., Riseborough S., Brading M., Clément C.Y., Wits D.J., Smith J., Colombé S., Pettitt V., Wheeler D.A. and Greenwood D.R. (1990) Identity and distribution of ecdysteroids in fat hen, *Chenopodium album*. *Invertebrate Reproduction and Development* **18**, 111.

Dinan L., Riseborough S., Brading M., Clément C.Y., Wits D.J., Smith J., Colombé S., Pettitt V., Wheeler D.A. and Greenwood D.R. (1991) Phytoecdysteroids in the Chenopodiaceae (Goosefoots). In: *Insect Chemical Ecology* (Ed. Hrdý I.), Academia Prague, pp. 215-220.

Dinan L., Whiting P. and Scott A.J. (1998) Taxonomic distribution of phytoecdysteroids in seeds of members of the Chenopodiaceae. *Biochemical Systematics and Ecology* **26**, 553-576.

Abstract: Extracts of seeds of ca. 200 species in the Family Chenopodiaceae have been assessed for the presence of insect steroid hormone agonists and antagonists by means of a bioassay based on the ecdysteroid-induced responses of the *Drosophila melanogaster* B_{II} cell line and ecdysteroid-specific radioimmunoassay (using the DBL-1 antiserum). Phytoecdysteroids (ecdysteroid agonists) were found in ca. 35 % of the species, but no antagonistic extracts were detected. Some samples, especially in the genus *Bassia* sect. *Kochia* where phytoecdysteroid levels are low, were cytotoxic to *D. melanogaster* B_{II} cells when assessed as a neat extract. Phytoecdysteroid levels varied considerably with highest levels detected in seeds of *Halimione portulacoides* (3.6 mg ecdysone equivalents g⁻¹), *Atriplex isatidea* (3.7 mg ecdysone equivalents g⁻¹) and *Rhagodia candolleana*. (8.9 mg ecdysone equivalents g⁻¹). The presence/absence of phytoecdysteroids has been related to a taxonomic classification of the Family. Phytoecdysteroids are almost absent from the sub-family Salsoloideae. Within the sub-family Chenopodioideae, ecdysteroids have been detected in all tribes except the Salicornieae and possibly the Camphorosmeae, but are most commonly present in the Atripliceae and Chenopodieae. Within the genus *Chenopodium*, all tested species of the sub-genus *Ambrosia* are ecdysteroid-negative, while most of those of the sub-genus *Chenopodium* are ecdysteroid-positive. Within the genus *Atriplex*, the situation is not so clear-cut, but most ecdysteroid-positive species occur within the Sections *Semibaccata*, *Dialysex* and *Deserticola*. Thus, the distribution of ecdysteroid-containing species within the Chenopodiaceae is not random, but it is rather related to the position of the species within a modern classification of the Family.

Dinan L., Sarker S.D., Bourne P., Whiting P., Šik V. and Rees H.H. (1999) Phytoecdysteroids in seeds and plants of *Rhagodia baccata* (Labill.) Moq. (Chenopodiaceae). *Archives of Insect Biochemistry and Physiology* **41**, 18-23.

Abstract: Seeds of *Rhagodia baccata* afforded, in addition to 20-hydroxy-ecdysone and polypodine B, a novel phytoecdysteroid, (20R)-22-deoxy-20,21-dihydroxyecdysone, the structure of which was elucidated unequivocally by UV, LSIMS and NMR techniques. This compound possessed agonistic activity in the *Drosophila melanogaster* B_{II} cell bioassay, with an ED₅₀ value of 2.0 x 10⁽⁻⁷⁾M (ED₅₀ value for 20-hydroxyecdysone = 7.5 x 10⁽⁻⁹⁾M). The distribution of ecdysteroids in plants of *R. baccata* has been determined. Highest levels are associated with the youngest aerial tissues and with the roots. Ecdysteroid profiles are qualitatively very similar throughout the plant, with 20-hydroxyecdysone and polypodine B predominating in all plant parts tested.

Dinan L., Savchenko T. and Whiting P. (2001a) Phytoecdysteroids in the genus *Asparagus* (Asparagaceae). *Phytochemistry* **56**(6), 569-576.

Abstract: Phytoecdysteroids, plant steroids which are analogues of invertebrate steroid hormones, probably contribute to the deterrence of phytophagous invertebrate predators. They also seem to possess antimicrobial activity and several pharmaceutical and medicinal benefits have been ascribed to them. Here, we present a survey of seeds of 16 species of the genus *Asparagus* (Asparagaceae), including the crop species *A. officinalis*, for ecdysteroid agonists (including phytoecdysteroids) and antagonists. Seven species were found to contain ecdysteroids with levels ranging from just detectable (*A. racemosus* and *A. sarmentosus*) to relatively high (*A. laricinus*). RP-HPLC/RIA/bioassay has been used to separate positive extracts of four species (*A. falcatus*, *A. laricinus*, *A. ramosissimus* and *A. scandens*)

and analyse the ecdysteroid profiles. The identities of the major ecdysteroids were confirmed by NP-HPLC. Seeds of *A. officinalis* do not contain detectable levels of ecdysteroids, but leaves, stems and roots contain low levels (detectable by RIA). This indicates that *A. officinalis* retains the genetic capacity to synthesise ecdysteroids and that future strategies could be developed for enhanced protection of asparagus spears through elevated ecdysteroid levels.

Dinan L., Bourne P. and Whiting P. (2001b) Phytoecdysteroid profiles in seeds of *Sida* spp. (Malvaceae). *Phytochemical Analysis* 12, 110-119.

Abstract: Procedures are presented for the assessment of the phytoecdysteroid profiles in small plant samples (ca. 25 mg), using seeds of *Sida* spp. as an example. The procedures are suitable for the analysis of minute or valuable samples and provide copious information for chemotaxonomic purposes. Methanolic extracts of the plant material, after partitioning against hexane, were separated by reversed-phase gradient HPLC monitored by PAD, RIA and bioassay. Aliquots of the fractions were also treated with *Helix pomatia* hydrolases, followed by RIA and bioassay, in order to assess the presence of hydrolysable ecdysteroid conjugates. Further information could also be obtained by separation of samples using normal-phase gradient HPLC. Among 11 species of *Sida* examined, seed extracts of *S. acuta* (= *S. carpinifolia*) and *S. rhombifolia* were found to contain significant amounts of ecdysteroids, seed extracts of *S. filicaulis* contained only moderate levels, whilst the remaining species showed no detectable levels of ecdysteroids. The ecdysteroid profiles of the extracts of the three positive species were significantly different, demonstrating that phytoecdysteroids have chemotaxonomic value in this genus.

Dinan L., Whiting P. and Savchenko T. (2001c) Phytoecdysteroids in seeds of *Lloydia serotina* (Liliaceae). *Biochemical Systematics and Ecology* 29(9), 923-928.

Abstract: Seeds of a number of species in the Liliaceae (sensu Brummitt, 1992) were examined for the presence of ecdysteroid agonist and antagonist activities. No species were antagonistic to 20-hydroxyecdysone action on the ecdysteroid-responsive *Drosophila melanogaster* B(II) cell line and only one extract, that of *Lloydia serotina*, was agonistic. This activity is attributable to the presence of phytoecdysteroids as detected by ecdysteroid-specific radioimmunoassay and the agonist version of the B(II) bioassay. HPLC in conjunction with radioimmunoassay and bioassay have been used to determine the ecdysteroid profile. The major ecdysteroids present are identified as 20-hydroxyecdysone and polygodine B (5beta,20-dihydroxyecdysone).

Dinan L., Savchenko T. and Whiting P. (2001d) On the distribution of phytoecdysteroids in plants. *Cellular and Molecular Life Sciences* 58, 1121-1132.

Abstract: The occurrence and levels of phytoecdysteroids in the seeds and other parts of plants grown from the seeds of 180 randomly selected plant species were assessed and compared. Ecdysteroids are frequently detectable in leaves and flowers, but less so in stems, roots and seeds. The seeds of 290 species were assessed for the presence of hydrolysable ecdysteroid conjugates. Low levels of conjugates were detected in a significant number of species, large amounts being present only when levels of free ecdysteroids were high. Individual plants of *Arabidopsis thaliana* were assessed for the presence of phytoecdysteroids. While plants of this species are generally ecdysteroid negative, individual plants in the population accumulate low levels of ecdysteroids. Extracts of seeds of 50 "ecdysteroid-negative" species were concentrated and partially purified to determine if they possess ecdysteroids at levels below the usual detection levels. Ecdysteroids were detectable by radioimmunoassay in almost all of these concentrated samples. Thus, all lines of evidence point to the conclusion that all species of plants have the capacity to produce at least low levels of phytoecdysteroids. This has important implications for the protection of crop species through enhancing ecdysteroid levels by breeding/genetic modification strategies.

Dinan L., Savchenko T. and Whiting P. (2002a) Chemotaxonomic significance of ecdysteroid agonists and antagonists in the Ranunculaceae: phytoecdysteroids in the genera *Helleborus* and *Hepatica*. *Biochemical Systematics and Ecology* 30, 171-182.

Abstract: We present here a survey of ca. 100 species within 16 genera of the family Ranunculaceae for the presence of ecdysteroid agonist and antagonist activities in methanolic seed extracts. The levels of phytoecdysteroids (agonists) have been quantified by radioimmunoassay and bioassay. A few samples possess weak antagonistic activity. Phytoecdysteroids are most prominently associated with the genus *Helleborus*. In this genus, species fall into two distinct classes: those with low or undetectable ecdysteroid levels and those with high ecdysteroid levels. The relationship between ecdysteroid levels and the biology of the plants in this genus is discussed. Additionally, the extract of *Hepatica triloba* Chaix seeds contains a significant level of phytoecdysteroids. Several other species contain low levels of phytoecdysteroids, as detected by radioimmunoassay. Together with our previous data on the genera *Anemone* and *Pulsatilla*, this survey allows us to present an overview of the distribution of ecdysteroids in this family.

Dinan L., Mamadalieva N.Z. and Lafont R. (2020a) Dietary phytoecdysteroids. In: *Handbook of Dietary Phytochemicals* (Eds Xiao J. et al.) pp 54 (doi: org/10.1007/978-981-13-1745-3_35-1).

Dinan L., Balducci C., Guibout L. and Lafont R. (2020b) Small-scale analysis of phytoecdysteroids in seeds by HPLC/DAD/MS for the identification and quantification of specific analogues, dereplication and chemotaxonomy. *Phytochemical Analysis* (<https://doi.org/10.1002/pca.2930>).

Abstract:

Introduction

Phytoecdysteroids are analogues of arthropod steroids occurring in plants. They contribute to invertebrate deterrence. A wide diversity of ecdysteroids occurs in phytoecdysteroid-containing plant species, sometimes in high amounts. Ecdysteroids demonstrate potentially useful pharmaceutical actions in mammals.

Objectives

Establish reversed-phase high-performance liquid chromatography with tandem mass spectrometry (RP-HPLC-MS/MS) and RP-HPLC-DAD-MS (diode array detector mass spectrometry) methods for the separation, identification and quantification of ecdysteroids to screen for species containing significant amounts of 20-hydroxyecdysone (20E) and other useful ecdysteroids.

Materials and methods

Micro-extracts of seed samples (ca. 30 mg) in 50% ethanol were subjected to RP-SPE (solid-phase extraction) purification prior to analysis by RP-HPLC-MS/MS and RP-HPLC-DAD-MS.

The method was initially applied to genera

(*Amaranthus*, *Centaurea*, *Lychnis*, *Ourisia*, *Serratula*, *Silene* and *Trollius*) where high-accumulating species had been previously encountered. Seeds of 160 randomly selected species, many of which have not previously been assessed, were then analysed. HPLC-MS/MS with a short analysis time initially identifies ecdysteroid-positive extracts and quantifies 20E. The positive extracts (20 ng 20E) are then analysed by HPLC-MS/MS with a longer analysis time to identify and quantify 17 common phytoecdysteroids and, finally, HPLC-DAD-MS (0.1–0.25 µg 20E) is used to obtain UV- and MS-spectra to confirm identifications or as a basis for characterisation of partially identified or novel analogues.

Results

Lychnis coronaria, *Silene fimbriata* and *Silene hookeri* ecdysteroids are characterised for the first time and those of *Cucubalus baccifer* and *Ipheion uniflorum* are more extensively characterised.

Conclusions

The procedure provides a rapid/sensitive method for screening small plant samples for the presence, quantification and identification of ecdysteroids. It permits ready dereplication of samples, identifying extracts containing large amounts or novel analogues.

Dini I., Tenore G.C. and Dini A. (2005) Nutritional and antinutritional composition of Kancolla seeds: an interesting and underexploited andine food plant. *Food Chemistry* **92**(1), 125-132.

Abstract: The quality of Kancolla seeds, a sweet variety of quinoa, an underexploited food plant, was determined by measuring proximate composition (carbohydrate, lipid, protein, fibre, essential amino acids and minerals), antinutritional factors (anions) and phytoecdysteroids. The results show that the kancolla seeds are nutritionally interesting and differ from other quinoa varieties, mainly in fibre and mineral contents. Results suggest a major alimentary use of kancolla seeds. They have promising economic value. The challenge is to find ways to incorporate them into existing food products, as well as to create new products from them.

Djerassi C., Knight J.C. and Brockmann H. (1964) Neue Sterine aus dem Kaktus *Wilcoxia viperina* [New steroid from the cactus *Wilcoxia viperina*]. *Chemische Berichte* **97**, 3118-3130 [in German].

Zusammenfassung: Aus den Wurzeln des Kaktus *Wilcoxia viperina* konnten neben Cholesterin, Campesterin, β -Sitosterin und Peniocerol drei neue α,β -ungesättigte Steroidketone isoliert werden. Eines dieser Ketone ist mit synthetischem 3β -Hydroxycholesten-(7)-on-(6) identisch, die beiden anderen, Viperidon und Viperidinon, sind Mono- und Dihydroxyderivate dieser Verbindung. Die Ergebnisse der katalytischen Hydrierung und spektroskopische Daten zeigen, dass sich die zusätzlichen Hydroxygruppen in 9α - bzw. 9α - und 14α -Stellung befinden.

Dong Q., Yan J., Zheng M., Huai H. and Tan J. (2010) Chemical constituents from seeds of *Achyranthes bidentata* Blume. *Redai Yaredai Zhiwu Xuebao* **18**(5), 569-572 | [in Chinese]

Abstract: Eight compounds were isolated from seeds of *Achyranthes bidentata* Blume. On the basis of the spectral data, they were identified as N-trans-feruloyltyramine (1), glycerol 1-O-9Z, 12 Z-octadecadienoate (2), β -ecdysterone (3), polypodine B (4), ergosta-7,22-diene- $3\beta,5\alpha,6\beta$ -triol (5), oleanolic acid 28-O- β -D-glucopyranosyl ester (6), oleanolic acid 3-O- β -D-glucopyranosyl ester (7), and daucosterol (8). Compounds 1, 2, 5 and 7 were obtained from the plant at the first time.

dos Santos T.C., Monache F.D. and Leitão S.G. (2001) Ecdysteroids from two Brazilian *Vitex* species. *Fitoterapia* **72**(3), 215-220.

Abstract: A new ecdysteroid, 26-hydroxypinnatasterone (1), together with 20-hydroxyecdysone, was isolated from the stem barks of *Vitex cymosa*. 20-Hydroxyecdysone, ajugasterone C, ajugasterone C monoacetone and turkesterone were isolated from the branches of *V. polygama*. The structure of 1 was determined by spectroscopic methods.

Dreier S.I. (1987) Occurrence and activity of ecdysterone (insect moulting hormone) in plants. M.Sc. thesis, Dept. Botany, University of British Columbia, Canada, pp. 85.

Du Y., Wang X-q., Bao B-q. and Hang H. (2016) Chemical constituents from flowers of *Rhaponticum uniflorum*. *Chinese Traditional and Herbal Drugs* (16), 2817-2821 [in Chinese].

Abstract: Objective: To investigate the chemical constituents from the flowers of *Rhaponticum uniflorum*. Methods: The chemical constituents were separated and purified by macroporous resin, silica gel, Sephadex LH-20, and MCI column chromatography. Their structures were determined by physicochemical properties and spectral data. Results: Seventeen compounds were isolated from the ethanol extract in the flowers of *R. uniflorum*. Among them, eleven flavones were identified as 5,7,4'-trihydroxy-3'-methoxyflavone (1), quercetin-3'-O-methyl ether (2), apigenin (3), kaempferol (4), luteolin (5), quercetin (6), apigenin-7-O- β -D-glycuronate ethyl ester (7), kaempferol-3-O- α -L-rhamnoside (8), quercetin-3-O- α -L-rhamnoside (9), apigenin-7-O- β -D-glucopyranoside (10), and apigenin-6,8-di- β -D-glucoside (11); Two lignans were hemislin B glucoside (12) and hemislin B (13); Two phytoecdysones were ecdysterone (14) and turkesterone (15); Others were ursolic acid (16) and 3,5-O-dicaffeoyl quinic acid (17). Conclusion: Compounds 1, 2, 7—10, 12 and 13 are isolated from the plants of *Rhaponticum* Cass. for the first time and the compounds 1, 2, and 7—17 are found from the flowers of *R. uniflorum* for the first time.

Du Z., Dong C., Xia W. and Wei S. (2016) Quality assessment for *Achyranthes* with different soil condition. *Modern Chinese Medicine* (9), 1164-1166 [in Chinese, with an English abstract].

Dzhukharova M.K., Saatov Z., Gorovits M.B. and Abubakirov N.K. (1991) Phytoecdysteroids of *Silene* plants. XVIII. 2-Deoxyecdysterone 20,22-monoacetone from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (2), 241-244/*Chemistry of Natural Products* **27**(2), 207-209 [in English].

Abstract: In addition to the known 2-deoxyecdysone, 2-deoxyecdysterone, 2-deoxyecdysone 22-O-acetate, ecdysterone, integristerone A, and sileneoside A, B, and C, the new ecdysteroid 2-deoxyecdysterone 20,20-monoacetone has been isolated from the roots of the plant *Silene brahuica* Boiss.

Dzhukharova M.K., Tashkhodzhaev B., Saatov Z. and Abdullayev N.D. (1993) Phytoecdysteroids from plants of the genus *Silene* XIV, brahuisterone from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (4), 553-558 [in Russian]/*Chemistry of Natural Compounds* **29**(4), 484-489 [in English].

Abstract: A new ecdysteroid with the composition $C_{27}H_{44}O_6$ which has been called brahuisterone has been isolated from the epigeal part of the plant *Silene brahuica* Boiss. Its chemical and complete spatial structures have been established by spectral studies and the x-ray structural method (diffractometer, Cu-K α radiations, 996 reflections, direct method, R=0.119).

Dzhukharova M.K., Saatov Z., Abdullaev N.D. and Abubakirov N.K. (1994a) Phytoecdysteroids of *Silene* plants. XV. sileneoside F - 3-O- β -D-glucopyranoside of brahuisterone from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (6), 734-737 [in Russian]/*Chemistry of Natural Products* **30**(6), 680-683 [in English].

Abstract: A new brahuisterone glycoside -- silenoside F -- has been isolated from the epigeal part of *Silene brahuica* Bois. (am. Caryophyllaceae). Its structure has been established by an analysis of spectral characteristics: 3:3, 5,14ot, 22R, 25-pentahydroxy-5-cholest-6-one 3-O-13-D-glucopyranoside.

Dzhukharova M.K., Saatov Z. and Abdullaev N.D. (1995) Phytoecdysteroids of *Silene* plants. XVI. 5 α -sileneoside E from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (2), 253-256 [in Russian].

El-Shakhawy F.S., Abou-Hussein D.R., El-Kersh D.M. and Sleem A.A. (2012) Anabolic and androgenic effects of certain *Atriplex* species grown in Egypt. *Egyptian Journal of Biomedical Science* **40**, 97-113 [in English with a summary in Arabic].

Abstract: 20-Hydroxyecdysone (20-HE) was detected by TLC in the ethyl acetate fraction of two *Atriplex* species grown in Egypt: *A. lindleyi* subsp. *inflata* and *A. leucoclada*. EIA quantification of 20-HE proved its presence in a concentration of 9.15 and 7.3 μ g/g dried aerial parts in each of the two species, respectively. Significant anabolic and androgenic activities were demonstrated for 20-HE comparing to testosterone; the study also revealed that the total alcohol extract and the ethyl acetate fraction of *A. lindleyi* subsp. *inflata* exhibited greater activities than their

analogues in *A. leuoclada*. Column chromatographic fractionation of the ethyl acetate fraction of the most active species *A. lindleyi* subsp. *inflata* resulted in the isolation of 20-HE (I) in addition to three flavonol glycosides: Isorhamnetin 3-O-galactoside (II), Isoquercitin (III) and Rutin (IV) identified via spectral analysis. DNA were amplified using seven decamer primers as a contribution to the characterization and discrimination between the two *Atriplex* species indicating only one polymorphic band between the two species and displaying a similarity coefficient of 88.88%. 20-hydroxyecdysone (20-HE) has been known by its effectiveness as anabolic and is marketed as an ingredient in nutritional supplements for various sports, particularly body building (Bathori et al., 2008). It is a phytoecdysteroid produced by various plants including *Atriplex* species (Dinan, 1995, Dinan et al., 1998 and Keckeis et al., 2000). TLC screening revealed the presence of 20-HE in *A. lindleyi* subsp. *inflata* and *A. leuoclada* grown in Egypt. Previous reports indicated also its isolation from *A. lindleyi* subsp. *inflata* (Ateya et al., 2005). This study aimed to evaluate-for the first time-the anabolic and androgenic potentialities of *Atriplex* species under investigation in correlation to their content of 20-HE. Moreover, RAPD (Random Amplified Polymorphic DNA) technique was performed for genotyping characterization and discrimination between the two plant species.

Erst A.A., Zheleznichhenko T.V., Badulina A.A., Zibareva L.N. and Kovzunova O.V. (2016) Biosynthesis of phytoecdysteroids in the hairy root culture of *Silene linicola* C.C. Gmelin. *Plant Cell Biotechnology and Molecular Biology* 17(7&8) 326-334.

Abstract: The hairy root cultures of the ecdysteroid-containing species *Silene linicola* were obtained. The resulting cultures were analyzed for ecdysteroid content by means of high-performance liquid chromatography. A number of nonpolar and mid-polar ecdysteroids were detected. It was demonstrated that the presence of sodium acetate (150 mg l⁻¹) in the nutrient medium promotes biosynthesis of a more diverse set of ecdysteroid compounds. Estimation of the levels of the major ecdysteroid 20-hydroxyecdysone showed that its general content in all samples varies within the range 0.13-0.18%. The results provide evidence that the use of hairy root cultures of *S. linicola* allows one to obtain a stable yield of the high levels of phytoecdysteroids and to enhance the level of biosynthesis using precursors of biosynthesis.

Erst A.A., Zibareva L.N. and Filonenko E.S. (2018) Effect of methyl jasmonate on production of 20-hydroxyecdysone and turkesterone in *hairy roots* of *Silene linicola* C.C. Gmelin. *BIO Web of Conferences* 11, 00013 (doi: 10.1051/bioconf/20181100013).

Abstract: Methyl jasmonate (MeJ) affects the regulation of secondary metabolism, and it is considered to be a promising elicitor in the culture of cells, tissues and plant organs. High-performance liquid chromatography method was used to identify the composition of ecdysteroids in *hairy roots* of *Silene linicola*. MeJ was found to have a stimulating effect on ecdysteroid biosynthesis in this culture. Addition of MeJ at a concentration of 100 µM increased the biosynthesis of 20-hydroxyecdysone by 74% (day 3), and that of turkesterone by 35% (day 6). The share of turkesterone in total ecdysteroid content in the investigated samples was up to 60%, and the content of 20-hydroxyecdysone was up to 30%. The study shows that MeJ is a promising stimulator of ecdysteroid biosynthesis in *hairy roots* of *S. linicola*.

Erst A.A., Zibareva L.N., Filonenko E.S. and Zheleznichenko T.V. (2019) Influence of methyl jasmonate on production of ecdysteroids from hairy roots of *Silene linicola* C.C. Gmelin. *Russian Journal of Bioorganic Chemistry* 45(7), 920-926.

Abstract: *Hairy roots* cultures are being considered as a promising system for producing valuable second metabolites. These genetically transformed root cultures are characterized by high growth rate, genetic stability and growth in hormone free media. Using *A. rhizogenes*-mediated transformation method (strain A4), we have obtained *hairy roots* cultures of the ecdysteroid-containing species of *Silene linicola*. HPLC analysis of the sample studied revealed that 20-hydroxyecdysone (20E), turkesterone and polypodin B were synthesized under the specified experimental conditions. Application of methyl jasmonate at a concentration of 100 µM resulted in stimulation of 20E biosynthesis (up 74%) after three days of cultivation and turkesterone up to 35% at six days. It was noted that total ecdysteroid content in sample tested varied: turkesterone from 25 to 60%, and 20E from 8 to 30%. At the same time, the level of 20E biosynthesis decreased from 0.023 to 0.014% in the samples without methyl jasmonate treatment. *Hairy roots* lines of *S. linicola* with different responses to the presence of elicitors in the culture medium can be used to study the pathways of ecdysteroid biosynthesis.

Ezhava S., Kothari K. And Patwari A. (2016) 20-Hydroxyecdysone determination from *Achyranthes aspera* linn by high performance thin layer chromatographic method. *Indian Drugs* 53(4), 54-57.

Abstract: 20-Hydroxyecdysone a phytoecdysteroid having diverse biological activities, commonly occurs in roots of *Achyranthes aspera* L., family Amaranthaceae, which have high commercial, economical and medicinal value. In the present study, a simple method was developed for the isolation of 20-hydroxyecdysterone from roots of *Achyranthes aspera* Linn. A validated HPTLC method has been established for the determination of 20-Hydroxyecdysone in roots and seeds of *Achyranthes aspera*. The methanol extracts of root, seeds its formulation and

standard solution were applied on silica gel F254 HPTLC plates. The plates were developed in twin chamber using mobile phase chloroform: methanol: ammonia (8:2:0.2 V/V) and scanned at 254 using Camag TLC scanner 3 with CATS 4 software. A linear relationship was obtained between response (peak area) and amount of 20-hydroxyecdysone in the range of 600-2100ng/spot; the correlation coefficient was 0.9984 respectively. Sharp, well resolved peak 20-hydroxyecdysone spot resolved at R_f 0.41±0.03 from other components of the sample extracts. The LOD and LOQ were found to be 50 and 152.3ng/spot respectively. The HPTLC method showed good linearity, recovery and high precision of 20-hydroxyecdysone. Amount of 20-hydroxyecdysone in roots and seeds was found to be 0.176% and 0.069% w/w.

Fang L., Li J., Zhou J., Wang X. And Guo L. (2017) Isolation and purification of three ecdysteroids from the stems of *Diploclisia glaucescens* by high-speed countercurrent chromatography and their anti-inflammatory activities in vitro. *Molecules* **22**, 1310 (doi: 10.3390/molecules22081310).

Abstract: High-speed counter-current chromatography was used to separate and purify ecdysteroids for the first time from the stems of *Diploclisia glaucescens* using a two-phase solvent system composed of ethyl acetate-*n*-butanol-ethanol-water (3:0.2:0.8:3, v/v). Three ecdysteroids were obtained from 260 mg of ethyl acetate extract of the residue obtained after evaporation of the crude ethanolic extract of *D. glaucescens* in one-step separation, which were identified as paristerone (I, 30.5 mg), ecdysterone (II, 7.2 mg), and capitasterone (III, 8.1 mg) by electrospray ionization mass spectrometry (ESI-MS) and nuclear magnetic resonance (NMR). Their anti-inflammatory activities were evaluated by measuring the inhibitory ratios of β -glucuronidase release in rat polymorphonuclear leukocytes (PMNs) induced by platelet-activating factor. Compounds I-III showed significant anti-inflammatory activities with IC_{50} -values ranging from 1.51 to 11.68 μ M, respectively.

Fang Y., Zhang S-X., Huo C-H., Sauriol F., Shi Q-W. and Kiyota H. (2010) Two new phytoecdysteroids from the needles of *Taxus canadensis*. *Zeitschrift für Naturforschung* **65b** 1-5.

Abstract: Two new phytoecdysteroids with a 20,22-acetal group were identified for the first time from the needles of the Canadian yew, *Taxus canadensis*. Their structures were characterized as ponasterone A 20,22-p-hydroxybenzylidene acetal (1) and ponasterone A 20,22-acetonide (2) on the basis of 1D, 2D NMR evidence and high-resolution FABMS analysis

Faux A., Galbraith M.N., Horn D.H.S. and Middleton E.J. (1970) The structures of two ecdysone analogues, Cheilanthones A and B, from the fern *Cheilanthus tenuifolia*. *Journal of the Chemical Society, Chemical Communications* 243-244.

Abstract: Cheilanthones A and B are shown to be 7,8-dihydroecdysone (II) and 25-deoxy-7,8-dihydroecdysone (V), respectively.

Fekete G., Polgar L.A., Bathori M., Coll J. and Darvas B. (2004) *Per os* efficacy of *Ajuga* extracts against sucking insects. *Pest Management Science* **60**(11) 1099-1104.

Abstract: We studied the efficacy of water-soluble extracts from four *Ajuga* spp on the post-embryonic development of two exopterygota (sucking insect) species. To allow comparison between different *Ajuga* species, results are expressed in terms of quantity of plant extracted per litre of test solution. Crude methanolic extracts of all *Ajuga* plants tested, with the exception of *A. genevensis*, showed considerable *per os* efficacy against larvae of both *Dysdercus cingulatus* F and *Acyrtosiphon pisum* (Harris) even at 1 g litre⁻¹. In the aphid tests the order of efficacy was *A. bracteosa* Wallich ex Benth > *A. chamaepitys* Schreber > *A. reptans* L > *A. genevensis* L. On *D. cingulatus* the order of efficacy was: *A. reptans* > *A. bracteosa* > *A. chamaepitys* > *A. genevensis*. Extracts were fractionated on SepPak using a range of methanol/water mixtures. Results are expressed in terms of the initial weight of plant extracted. The 100% methanolic fraction of *A. chamaepitys* was highly effective on *A. pisum* (100% mortality at 1 g litre⁻¹) and less effective on *D. cingulatus* (about 60% mortality at 5 g litre⁻¹). The entire 60% methanol + 40% water fraction was effective against test insects but showed different efficacies according to test species and concentration applied. 20-Hydroxyecdysone (20E), cyasterone (Cy) and ajugalactone (AjL) were identified in the fractions from all *Ajuga* species, but the remaining phytoecdysteroid profile was quite different between *Ajuga* species. Capitasterone (Cap) and 28- ϵ -sengosterone (5Cy28') were found only in *A. reptans*, makisterone A (MaA) and 29-norcyasterone (29NCy) were only in *A. chamaepitys*, while 22-acetylcysterone (Cy22A), 3- ϵ -cyasterone (Cy') and 3- ϵ -22-acetylcysterone (Cy'22A) were only in *A. bracteosa*. The total amount of phytoecdysteroids was 2053 mgkg⁻¹ for *A. bracteosa*, 1892 mgkg⁻¹ for *A. reptans* and 95 mg kg⁻¹ for *A. chamaepitys*

Felipe D.F., Brambilla L.Z.S., Porto C., Pilau E.J. and Cortez D.A.G. (2014) Phytochemical analysis of *Pfaffia glomerata* inflorescences by LC-ESI-MS/MS. *Molecules* **19**, 15720-15734.

Abstract: *Pfaffia glomerata* contains high levels of β -ecdysone, which has shown a range of beneficial pharmacological effects. The present study demonstrated that inflorescences of *P. glomerata* contain other important

bioactive compounds in addition to β -ecdysone. The identification of compounds from inflorescences using liquid chromatography coupled with electrospray ionization tandem mass spectrometry (LC-ESI-MS/MS) was performed for the first time. The eight compounds identified were β -ecdysone, flavonoid glycosides such as quercetin-3-O-glucoside, kaempferol-3-O-glucoside and kaempferol-3-O-(6-p-coumaroyl)-glucoside, oleanane-type triterpenoid saponins such as ginsenoside Ro and chikusetsusaponin IV, in addition to oleanonic acid and gluconic acid. This study provided information on the phytochemicals contained in *P. glomerata* inflorescences revealing the potential application of this plant part as raw material for the phytotherapeutic and cosmetic industries.

Felipe S.H.S., Batista D.S., Vital C.E., Chagas K., Silva P.O., Silva T.D., Fortini E.A., de Freitas Correia L.N., Avila R.T., Maldaner J., Festucci-Buselli R.A., DaMatta F.M. and Otoni W.C. (2019) Salinity-induced modifications on growth, physiology and 20-hydroxyecdysone levels in Brazilian-ginseng [*Pfaffia glomerata* (Spreng.)]. *Plant Physiology and Biochemistry* **140**, 43-54.

Abstract: Salinity is a major threat to agriculture. However, depending on the concentration of soluble salts in soil, increased secondary metabolite levels can occur with no major damages to plant growth and development. The phytoecdysteroid (PE) 20-hydroxyecdysone (20E) is a secondary metabolite with biotechnological, medicinal, pharmaceutical and agrochemical applicability. Here, we characterize the responses (growth and physiology) of *Pfaffia glomerata* under different NaCl concentrations and examine the production of 20E as affected by salinity. Forty-day-old plants grown in greenhouse were exposed to 0, 120, 240, 360 or 480 mM of NaCl for 11 days. Moderate salinity (i.e., 120 mM of NaCl) led to increased 20E concentrations in leaves (47%) relative to the control with no significant effect on photosynthesis and biomass accumulation, thus allowing improved 20E contents on a per whole-plant basis. In contrast, plants under high salinity (i.e., 240-480 mM of NaCl) displayed similar 20E concentrations in leaves compared to the control, but with marked impairments to biomass accumulation and photosynthetic performance (coupled with decreased sucrose and starch levels) in parallel to nutritional imbalance. High salinity also strongly increased salicylic acid levels, antioxidant enzyme activities, and osmoregulatory status. Regardless of stress severity, 20E production was accompanied by the upregulation of Spook and Phantom genes. Our findings suggest that *P. glomerata* cultivation in moderate salinity soils can be considered as a suitable agricultural option to increase 20E levels, since metabolic and structural complexity that makes its artificial synthesis very difficult.

Felix A.A. and Domingo A.O. (2008) Antioxidant activity and chemical constituents of *Pupalia lappacea* (L.) Juss. *Research Journal of Biological Sciences* **3**(7), 783-785.

Abstract: Phytochemical analysis of foliage of *Pupalia lappacea* (L.) Juss (Amaranthaceae) afforded 8 compounds, namely; 1-docosanol, stearic acid, stigmaterol, -sitosterol, N-benzoyl-L-phenylalaninol acetate, -sitosterol-3-O- -D-glucopyranoside, stigmaterol-3-O- -D-glucopyranoside and 20-hydroxyecdysone. The ecdysteroid, 20-hydroxyecdysone, constitutes nearly 0.3% of the dry weight and 13% of the dichloromethane extracts of the plant, making *Pupalia lappacea* a probable commercial source for this important ecdysteroid. Both the hexane and dichloromethane extracts showed significant antioxidant activity with the dichloromethane exhibiting an IC₅₀ comparable to that of BHT, a synthetic antioxidant. All the isolated compounds are reported for the first time in the genus.

Feng M-y., Chen Y., Yan H., Ni W., Qin X-j. and Liu H-y. (2018) Non-saponin constituents and their cytotoxicities from *Trillium kamtschaticum*. *Chinese Traditional and Herbal Drugs* (1), 90-94 [in Chinese, with an English abstract].

Abstract: Objective To investigate the chemical constituents and bioactivity of the moieties without hemostatic effects of *Trillium kamtschaticum*. Methods All compounds were isolated and purified by means of silica gel column chromatography, reverse phase C18 column chromatography, Sephadex LH-20, and recrystallization methods. Their structures were determined by physicochemical properties, and spectral data. The cytotoxic activity of selected compounds was also evaluated using the MTT method. Results Ten compounds were obtained from the moieties without hemostatic effects of *T. kamtschaticum* and their structures were identified as β -ecdysone (1), polypodine B (2), integristerone B (3), (9Z,15Z)-11,12,13-trihydroxyoctadeca-9,15-dienoic acid (4), (Z)-11, 12,13-trihydroxyoctadec-9-enoic acid (5), methyl (9Z,15Z)-11,12,13-trihydroxyoctadeca-9,15-dienoate (6), methyl (Z)-11,12,13-trihydroxyoctadec-9-enoate (7), 7,11-dimethyl-3-methylene-1,6-dodecadien-10 β ,11-diol-10-O- β -D-glucopyranoside (8), kaempferol-3-O- α -L-arabinopyranosyl (1 \rightarrow 6)-O- β -D-galacopyranoside (9), and 5-hydroxy-4-hydroxymethyl-2H-pyran-2-one (10), respectively. Compounds 1, 9, and 10 showed cytotoxic activities with IC₅₀ values of (26.6 \pm 1.3) μ mol/L (DU145 cells), (16.2 \pm 6.2) μ mol/L (CEM cells), and (23.7 \pm 1.2) μ mol/L (HeLa cells), respectively. Conclusion Compounds 4—7 and 10 are obtained for the first time from *Trillium* genus, and compounds 1, 9, and 10 showed cytotoxic activities

Feng Q., Chen G. and Pei Y. (2018) Isolation and identification of the chemical constituents from *Cephalotaxus fortunei* Hook. f. *Journal of Shenyang Pharmaceutical University* (2), 89-91 + 97 [in Chinese, with English abstract].

Ferreira P.R.B., Ferreira da Cruz A., Batista D.S., Nery L.A., Andrade I.G., Rocha D.I., Felipe S.H.S., Koehler A.D., Nunes-Nesi A. and Otoni W.C. (2019) CO₂ enrichment and supporting material impact the primary metabolism and 20-hydroxyecdysone levels in Brazilian ginseng grown under photoautotrophy. *Plant Cell, Tissue and Organ Culture* pp13 (doi: org/10.1007/s11240-019-016664-w).

Abstract: In vitro photoautotrophic propagation system has been successfully established for *Pfaffia glomerata*, a medicinal species that produces the phytoecdysteroid 20-hydroxyecdysone (20E) under forced ventilation and CO₂ enrichment. For that, an adequate supporting material with high porosity in place of agar is required. In this study, we investigated metabolic and morpho-anatomical alterations of two accessions (Ac) of *P. glomerata* (Ac 22 and Ac 43) under photoautotrophy conditions, using two supporting materials (agar and Florialite®) and two CO₂ concentrations (360 and 1000 µL CO₂ L⁻¹). High CO₂ concentration and the use of Florialite® as supporting material enhanced the production of 20E, and influenced on the levels of amino acids, sugars, tricarboxylic acid cycle intermediates, stress related metabolites (aromatic amines and shikimate), and osmotic adjustment-related compounds (hydroxyproline, aspartate and myo-inositol). Interestingly, Ac 22 displayed less tolerance to stress caused by low photoautotrophy compared to Ac 43, as indicated by the higher total polyamines in Ac 22. Moreover, the accessions showed different metabolic responses under photoautotrophy. These findings provide a better understanding of how the supporting material and CO₂ enrichment influence in vitro metabolism under photoautotrophic system in *P. glomerata*, wherein higher levels of gas exchange, enabled by use of Florialite® and CO₂ enrichment, increased total sugars as well as the levels of 20E in the plants. This information will be fundamental to optimize the in vitro culture systems of *P. glomerata* for the production of 20E.

Filippova V.N., Zorinyants S.E., Volodina S.O. and Smolenskaya I.N. (2003) Cell cultures of ecdysteroid-containing *Ajuga reptans* and *Serratula coronata* plants. *Russian Journal of Plant Physiology* 50(4), 501-508 [in English]/*Fiziologiya Rastenii* 50(4), 564-572 [in Russian].

Abstract: Cell suspension cultures of *Ajuga reptans* L. and *Serratula coronata* L. were derived from long-cultured calluses. Their growth patterns and morphophysiological characteristics indicated that these cells adapted well to grow in suspensions. During the growth cycle of cell suspension, the main ecdysteroid, among others present in plants, was 20-hydroxyecdysone (20E) for both cell lines. The content of 20E in cell suspension of *A. reptans* was 4–8 times higher than in the intact plant. After long culturing, the ecdysteroid profile in cell suspensions of *S. coronata* (20E, inokosterone, makisterone A, ecdyson, and a nonidentified metabolite) became similar to that of the intact plant. The ecdysteroids accumulated with a periodicity during subculturing cycle.

Flores R., Nicoloso F.T., Brondani D., Maldaner J., Cezarotto V., Giacomelli S.R. (2009) Extraction of ecdysterone from roots of Brazilian ginseng. *Ciencia Rural Santa Maria* 39(4), 1223-1226 [in Portuguese, with an English Abstract].

Abstract: This study aimed at optimizing the extraction method from ecdysterone of Brazilian ginseng. Root samples of two accessions (BRA and JB-UFSM) of *P. glomerata* were extracted in a Soxhlet with methanol or chloroform for 4h. In the second trial, the extraction was conducted in a Soxhlet or ultrasonic using methanol as a solvent. In *P. tuberosa*, the roots samples were extracted with methanol in a Soxhlet or in ultrasonic. The ecdysterone content was determined using high efficiency liquid chromatography methods. In both studied species, the highest ecdysterone content was detected from samples extracted in a Soxhlet and using methanol as a solvent. This extraction method has been successfully applied for determination of ecdysterone content from roots of Brazilian ginseng, and could be useful for the quality control of drugs and pharmaceutical formulations.

Flores R., Brondani D., Cezarotto V., Giacomelli S.R. and Nicoloso F.T. (2010) Micropropagation and β-ecdysone content of the Brazilian ginsengs *Pfaffia glomerata* and *Pfaffia tuberosa*. *In Vitro Cell. Dev. Biol.-Plant* 46, 210-217.

Abstract: The aim of this study was to investigate both a mass in vitro propagation system and the β-ecdysone content in roots and aerial parts of *Pfaffia glomerata* and *Pfaffia tuberosa*. Nodal segments of two genotypes (BRA and JB-UFSM) of *P. glomerata*, originated from aseptically grown plants, were cultivated on hormone-free Murashige and Skoog medium. For the proliferation of *P. tuberosa* shoots, nodal segments, originated from aseptically grown plants, were either cultivated on hormone-free Murashige and Skoog (MS) medium or were supplemented with 1.0µM thidiazuron (TDZ); the elongation and rooting of these plants were carried out on MS medium without TDZ. Plantlets of both species were acclimatized and transferred to field conditions. The β-ecdysone content in the plants was determined by high performance liquid chromatography. The BRA genotype showed a greater in vitro proliferation rate and β-ecdysone content than that of the JB-UFSM genotype. The culture of nodal segments of *P. tuberosa* on medium with 1.0µM TDZ with subsequent subcultivation of shoots on hormone-free medium was shown to be a suitable method for micropropagation due to the high multiplication rate and good plant development. Both species showed good adaptation to ex vitro conditions. The β-ecdysone content in micropropagated *P. tuberosa* was similar to that found in field-grown plants. For both species, the aerial parts accumulated higher β-ecdysone content than roots. These results reveal that micropropagation is a successful,

alternative method for rapid plant multiplication of both species of Brazilian ginseng. Furthermore, this study demonstrates that these two species have a potential for cultivation that is associated with high β -ecdysone production.

Flores R., Gimenes E.S. and Maldaner J. (2015) Production of β -ecdysone *in vitro*-cultured Brazilian ginseng *Pfaffia glomerata* (Spreng.) Pedersen. *Journal of Horticultural Science & Biotechnology* **90**(2), 109-114.

Abstract: In Brazil, several pharmaceutical industries produce phytomedicines from the roots of *Pfaffia glomerata* (Spreng.) Pedersen. In this work, callus cultures of *P. glomerata* were established in order to study whether *in vitro*-cultured tissue could produce β -Ecdysone. To induce calli, nodal segments were cultured on Murashige and Skoog (MS) medium with various levels of 6-benzylaminopurine (BAP; 1.0, 5.0, or 10.0 μ M), α -naphthaleneacetic acid (NAA; 0.5, 1.0, 5.0, or 10.0 μ M) or 2,4-dichlorophenoxyacetic (2,4-D; 0.5, 1.0, 5.0, or 10.0 μ M). β -Ecdysone concentrations [on a (w/w) dry weight (DW) basis] in *in vitro*-cultured and field-grown tissues were measured by high performance liquid chromatography. The level of β -ecdysone was related to the friability and shoot regeneration capacity of the different calli. Organogenic, friable calli grown on media supplemented with 10.0 μ M BAP, plus either 5.0 μ M NAA or 0.5 μ M 2,4-D, resulted in the highest β -ecdysone concentrations [0.282% (w/w) and 0.211% (w/w), respectively]. In contrast, the β -ecdysone concentrations in field-grown plants were 0.194% (w/w) in roots and 0.333% (w/w) in the aerial parts. Low levels of β -ecdysone [0.007% (w/w)] were detected in plantlets cultured on hormone-free MS medium, indicating the importance of culture conditions for the *in vitro* production of β -ecdysone. This is the first report on the presence of ecdysteroids in calli of *P. glomerata*.

Fuchino H., Nakamura H., Hakamatsuka T., Tanaka N., Cambie R.C. and Braggins J.E. (1997) Two new phytoecdysteroids from the fern *Schizaea dichotoma*. *Natural Medicines* **51**(5), 491-492.

Abstract: Two new phytoecdysteroids, named schizaeasterones A and B, were isolated from the fronds of *Schizaea dichotoma*. Their structures were determined to be (20R,22R,24R)-3 β ,11 α ,14 α ,20,22-pentahydroxy-24-methyl-5 β -cholest-7-en-6-one and (20R,22R,24 ξ ,25 ξ)-3 β ,11 α ,14 α ,20,22,26-hexahydroxy-24-ethyl-5 β -cholest-7-en-6-one, respectively, from spectral data.

Fujimoto Y., Kushiuro T. and Nakamura K. (1997) Biosynthesis of 20-hydroxyecdysone in *Ajuga hairy* roots: hydrogen migration from C-6 to C-5 during *cis*-A/B ring formation. *Tetrahedron Letters* **38**(15), 2697-2700.

Abstract: Feeding of deuterium labeled cholesterol including [6-(2)H]cholesterol and [3 α ,6-(2)H₂]-cholesterol to hairy roots of *Ajuga reptans* var. *atropurpurea* followed by (2)H-NMR analysis of the biosynthesized 20-hydroxyecdysone revealed that most of the deuterium atom located at C-6 of cholesterol migrated to the C-5 position of 20-hydroxyecdysone.

Fujimoto Y., Nakagawa T., Yamada J. and Morisaki M. (1996) Biosynthetic origin of C-26 and C-27 of the phytoecdysteroids cyasterone and 29-norcyasterone in *Ajuga hairy* roots. *Journal of the Chemical Society, Chemical Communications* (17), 2063-2064.

Abstract: Feeding of [¹³C₂] acetate to the hairy root culture of *Ajuga reptans* var. *atropurpurea* and ¹³C NMR analysis of the biosynthesized cyasterone and 29-norcyasterone reveal that the lactone carbonyl carbon of these phytoecdysteroids is derived from C-2 of mevalonate, whereas the methyl group on C-25 comes from C-6.

Fujimoto Y., Ohyama K., Nomura K., Hyodo R., Takahashi K., Yamada J., Morisaki M (2000) Biosynthesis of sterols and ecdysteroids in *Ajuga hairy* roots. *Lipids* **35**(3), 279-288.

Abstract: Hairy roots of *Ajuga reptans* var. *atropurpurea* produce clerosterol, 22-dehydroclerosterol, and cholesterol as sterol constituents, and 20-hydroxyecdysone, cyasterone, isocyasterone, and 29-norcyasterone as ecdysteroid constituents. To better understand the biosynthesis of these steroidal compounds, we carried out feeding studies of variously ²H- and ¹³C-labeled sterol substrates with *Ajuga hairy* roots. In this article, we review our studies in this field. Feeding of labeled desmosterols, 24-methylenecholesterol, and ¹³C₂-acetate established the mechanism of the biosynthesis of the two C₂₉-sterols and a newly accumulated codisterol, including the metabolic correlation of C-26 and C-27 methyl groups. In *Ajuga hairy* roots, 3 α -, 4 α -, and 4 β -hydrogens of cholesterol were all retained at their original positions after conversion into 20-hydroxyecdysone, in contrast to the observations in a fern and an insect. Furthermore, the origin of 5 β -H of 20-hydroxyecdysone was found to be C-6 hydrogen of cholesterol exclusively, which is inconsistent with the results in the fern and the insect. These data strongly support the intermediacy of 7-dehydrocholesterol 5 α ,6 α -epoxide. Moreover, 7-dehydrocholesterol, 3 β -hydroxy-5 β -cholest-7-en-6-one (5 β -ketol), and 3 β ,14 α -dihydroxy-5 β -cholest-7-en-6-one (5 β -ketodiol) were converted into 20-hydroxyecdysone. Thus, the pathway cholesterol \rightarrow 7-dehydrocholesterol \rightarrow 7-dehydrocholesterol 5 α ,6 α -epoxide \rightarrow 5 β -ketol \rightarrow 5 β -ketodiol is proposed for the early stages of 20-hydroxyecdysone biosynthesis. 3 β -Hydroxy-5 β -cholestan-6-one was also incorporated into 20-hydroxyecdysone, suggesting that the introduction of a 7-ene function is not necessarily next to cholesterol. C-25 Hydroxylation during 20-hydroxyecdysone biosynthesis was found to proceed

with *ca.* 70% retention and 30% inversion. Finally, clerosterol was shown to be a precursor of cyasterone and isocyasterone.

Gagalac-Nicolas M.G. and Sylianco C.Y.M. (1981) Anitmutagenic effects of *Cyanotis vaga* Lour. (Schultes) extracts. Bulletin of the Philippine Biochemical Society 4(1&2), 8-14.

Gaidi G., Miyamoto T., Laurens V. and Lacaille-Dubois M.-A. (2002) New acylated triterpene saponins from *Silene fortunei* that modulate lymphocyte proliferation. Journal of Natural Products 65(11), 1568-1572.

Abstract: Three new acylated triterpene saponins **1–3**, with a quillaic acid as aglycon, were isolated from the roots of *Silene fortunei* together with a known phytoecdysteroid (20-hydroxyecdysone). The compounds were characterized mainly by a combination of 2D NMR techniques, mass spectrometry, and chemical methods. Saponins **1–3**, jennisenosides C and D (**4, 5**), and **6** (deacylated form of **2/3** and **4/5**) were found to stimulate the proliferation of the Jurkat tumor cell lines at low concentration. At high concentration, **2/3** and **4/5** inhibited the proliferation of the cells and suggested the induction of apoptosis.

Galambosi B., Varga E., Hajdu Z. and Jokela K. (1997) Introduction of *Leuzea carthamoides* DC. as an adaptive medicinal plant in the Nordic climate. Drogenreport 10(16), 5-8.

Galbraith M.N. and Horn D.H.S. (1966) An insect-moulting hormone from a plant. Journal of the Chemical Society, Chemical Communications (24), 905-906.

No Abstract.

Galbraith M.N., Horn D.H.S., Porter Q.N. and Hackney R.J. (1968) Structure of podedcdysone A, a steroid with moulting hormone activity from the bark of *Podocarpus elatus* R.Br. Journal of the Chemical Society, Chemical Communications 971-972.

No Abstract.

Galbraith M.N. and Horn D.H.S. (1969) Insect moulting hormones: crustecdysone (20-hydroxyecdysone) from *Podocarpus elatus*. Australian Journal of Chemistry 22, 1045-1057.

Abstract: The insect moulting hormone crustecdysone has been isolated from the Australian brown pine, *Podocarpus elatus* R.Br., and detected in a number of other *Podocarpus* species from various parts of the world. A series of acetates and acetonides of crustecdysone has been prepared, and the mass and nuclear magnetic resonance spectra of these compounds analysed. A simple micro method of measuring the rate of acetylation of hydroxy groups in steroids has been developed to determine the steric environment of these groups.

Galbraith M.N., Horn D.H.S. and Middleton E.J. (1969) The structure of podedcdysone B, a new phytoecdysone. Journal of the Chemical Society, Chemical Communications 402-403.

No Abstract.

Galbraith, M. N., Horn, D. H. S., Middleton, E. J., Kaplanis, J. N., & Thompson, M. J. (1973) Structure of podedcdysone C, a steroid with a molting hormone activity from bark of *Podocarpus elatus*. *Experientia* 29, 782.

No Abstract.

Galiautdinov I.V., Baltaev U.A. and Odinokov V.N. (1999) 20-Hydroxyecdysone and its 22-acetate from *Serratula coronata*, one-step synthesis of shidasterone 22S-analog. *Chemicky Listy, Symposia* 93 S54.

Gallo M.B.C., Beltrame F.L., Vieira P.C., Cass Q.B., Fernandes J.B., da Silva M.F. dG.F. (2006) Quantitative determination of 20-hydroxyecdysone in methanol extract of twigs from *Vitex polygama* Cham. Journal of Chromatography B 832, 36-40.

Abstract: 20-Hydroxyecdysone (20E) is effective in stimulating protein synthesis, therefore, it has been largely used as anabolic agent in several commercial formulas. Phytochemical study of methanolic extract of twigs from *Vitex polygama*, used in traditional Brazilian medicine as emenagogue, yielded a large quantity of 20E. This finding led us to developing and validating a simple and reliable method to determine 20E in the surveyed extract. Chromatographic separation of 20E was achieved on a phenyl-hexyl-based column using reversed elution mode. Extract was cleaned-up by solid phase extraction employing C(18) cartridge, and an absolute recovery of 97% was acquired. External standard and standard addition calibration graphs were obtained and good linearity was accomplished ($r > 0.999$ for both curves). The limit of quantification and detection were determined. The results for accuracy fell within the -5 to +7% range.

Galyautdinov I.V., Sadretinova Z.R., Muslimov Z.S., Gareev V.F., Khalilov L.M. and Odinkov V.N. (2016) New minor ecdysteroids from the juice of *Serratula coronata* L. (Asteraceae). *Journal of Medicinal Plants Studies* 4(5), 30-34.

Abstract: Minor phytoecdysteroids - ajugasterone C 2-, 3-, and 11-acetates and calonysterone were isolated for the first time from the *Serratula coronata* plant. Out of these, ajugasterone 11-acetate is new ecdysteroid.

Gangan V.D., Pradhan P. and Sipahimalani A.T. (1997) Phytoecdysones from *Tinospora cordifolia*: structural elucidation of ecdysterone and makisterone A by 2D nmr spectroscopy. *Indian Journal of Chemistry B, Organic Chemistry including Medicinal Chemistry* 36(9), 787-792.

Abstract: The n-BuOH fraction of the methanolic extract of *T. cordifolia* stems. has been acetylated and subjected to exhaustive chromatography (column, radial and preparative TLC). Two phytoecdysones. viz. ecdysterone I and makisterone A 2 have been isolated as their polyacetates. Their structures have been elucidated by extensive 10 and 20 NMR studies.

Ganiev S. (1973) Ecdysterone from *Serratula sogdiana* and its influence on the metamorphosis of *Musca domestica*. *Uzbek. Biol. Journal* (2), 72-74 [in Russian].

Ganiev S.G. (1975) Ecdysone content in some plants of the tribe Cynareae Less., Family Asteraceae (Compositae). *Rastitelny Resursy* 11(1), 94-96 [in Russian].

Ganiev S.G. (1980) Amount of ecdysones in some plants of *Serratula* L. and *Rhaponticum* Ludw. *Rastitelny Resursy* 16(2), 193-198 [in Russian].

Gao C., Guo D., Qiao L., He W. and Lou Z (1991) NMR study on the structure of a new phytoecdysteroid. *Bopuxue Zazhi* (Chinese Journal of Magnetic Resonance) 8(4), 379-86 [in Chinese, with an English abstract] [CA 116:152145c].

Gao H., Ma X., Wen X., Chen D. and Si J. (2009) Chemical constituents from *Pfaffia paniculata*. *Zhongcaoyao* 40(4), 522-525 [in Chinese].

Abstract: For obtaining a more comprehensive understanding on the effective components, chem. constituents from the roots of *Pfaffia paniculata* Pedersen were studied. The chem. constituents were isolated and purified by silica gel and Sephadex LH-20 column chromatog. Their structures were elucidated by chem. and spectral analyses. Twelve compounds were obtained and identified as ecdysterone (I), 3-methoxy-4-hydroxy-benzoic acid (II), oleanolic acid 3-O- β -D-glucuronopyranoside (III), oleanolic acid 28-O- β -D-glucopyranoside ester (IV), oleanolic acid (V), stigmasterol- β -D-glucoside (VI), stigmasterol (VII), daucosterol (VIII), β -sitosterol (IX), 20-hydroxyecdysone-20,22-monoacetone (X), 1-O- β -D-glucopyranosyl-(2S,3S,4E)-2-[2'(R)-hydroxyl hexadecanoyl-amino]-4-octadecene-1,3-diol (XI) and oleanolic acid 3-O- β -D-glucuronopyranoside-Me ester (XII). Compounds II, III, IX, X and XII were isolated from this genus for the first time, and compounds I, IV and V were isolated from this plant for the first time.

Gao X.-y., Wang D.-w. and Li F.-m. (2000) Determination of ecdysterone in *Achyranthes bidentata* BL. and its activity promoting proliferation of osteoblast-like cells. *Acta Pharmaceutica Sinica* 35(11), 868-870 [in Chinese, with an English abstract].

Abstract:

Aim: To study the activity of ecdysterone from *Achyranthes bidentata* Bl. (AB) promoting proliferation of osteoblast-like (OB-like) UMR106 cells and to determine its content in AB by HPLC method.

Methods: Ecdysterone isolated from AB was cultured with OB-like cells UMR106 together in vitro and the proliferation of OB-like cells was determined by MTT assay. The chromatographic conditions for determining ecdysterone included an ODS column (250 mm x 4.6 mm, 5 microns), a mobile phase consisting of a mixture of water-acetonitrile-tetrahydrofuran (86:11:3), detection wavelength of 243 nm, and column temperature of 27 degrees C. Phenacetin was used as the internal standard.

Results: The ecdysterone from AB had significant activity promoting proliferation of OB-like cells, the proliferation was promoted by 41% (n = 3). The average recovery of ecdysterone was 96.2% (RSD = 2.1%), the calibration was linear in the range of 30-300 micrograms.mL⁻¹ (gamma = 0.9998).

Conclusion: Ecdysterone was screened quickly by cultivating with OB-like cells together in vitro. The HPLC method is accurate, fast and reproducible for the determination of ecdysterone in AB.

Gerard P.J., Ruf L.D., Lorimer S.D. and Heath A.C.G. (1997) Activity of extracts and compounds from New Zealand gymnosperms against larvae of *Lucilia cuprina* (Diptera: Calliphoridae). *New Zealand Journal of Agricultural Research* 40(2), 261-267.

Abstract: A modified serum-based bioassay was used to screen plant extracts and assess compound activity against newly hatched larvae of the Australian sheep blowfly *Lucilia cuprina*. Larval instar mortality and physical appearance were recorded after exposure for 24 h to the treatments. Extracts from New Zealand gymnosperms shown to have activity against *L. cuprina* larvae were: *Podocarpus totara*; *P. acuteifolius*; *Dacrycarpus dacrydioides*; *Halocarpus bidwillii*; *H. biformis*; *H. kirkii*; *Lagarostrobos colensoi*; *Lepidothamnus intermedius*; *L. laxifolius*; *Phyllocladus tricho-manoides*; *P. glaucus*, and *Agathis australis*. The phytoecdysones 20-hydroxyecdysone, 5,20-dihydroxyecdysone, and ponasterone A inhibited larval moulting. Dyshomoerythrine, dacrysterone, and nagilactone C were insecticidal, with the latter also retarding growth at sublethal rates.

Geszprych A. and Weglarz Z. (2002) Accumulation of biologically active compounds in the seeds of *Rhaponticum carthamoides* (Willd.) Illjin cultivated in Poland. *Folia Horticulturae* 14(1), 195-199.

Abstract: The aim of the present study was to determine ecdysterone (20-hydroxyecdysone) and sterol content in the seeds of *Rhaponticum carthamoides* cultivated in Poland. The effect of plant age, seed ripeness, and location of seeds in a fructification on the concentration of the above-mentioned compounds in seeds was investigated. The seeds of 4-year-old plants were characterised by a significantly higher content of ecdysterone and sterols in comparison with the seed of 2- and 3-year-old plants. The concentration of ecdysterone and sterols in the ripe seeds was distinctly higher than in those collected at milk- and wax-ripe stages. The concentration of investigated compounds (especially ecdysterone) in the seeds was related to their location in a fructification and was significantly higher in the achenes from the external part of a capitulum.

Ghedira K., Chemli R., Richard B., Zeches M. and Le Men-Olivier L. (1991) Contribution à l'étude pharmacopée traditionnelle de Tunisie: Étude des parties aériennes d'*Ajuga iva* (L.) Schreb. [Contribution to the study of the traditional pharmacopeia of Tunisia: study of the aerial parts of *Ajuga iva* (L.) Schreb.] *Plantes Médicinales et Phytothérapie* 25(2/3), 100-111 [in French with English abstract].

Ghosh D. and Laddha K.S. (2006) Extraction and monitoring of phytoecdysteroids through HPLC. *Journal of Chromatographic Science* 44(1), 22-26.

Abstract: The size of the phytoecdysteroids family is rapidly growing. Recent data shows over 250 ecdysteroid analogs have been identified so far in plants. It is theorized that there are over 1000 possible structures, which might occur in nature, but it is a fact that ecdysteroids usually occur in plants as a complex cocktail of structurally different compounds. Among these compounds, the major component is usually the common ecdysteroid-like 20-hydroxyecdysone. Ecdysteroids are polar steroids, almost sugar-like in their solubility properties. Extraction and purification of ecdysteroids (polyhydroxy steroids) is complicated by their polar nature and poor crystallizing properties. These properties make them difficult to separate from other polar plant constituents. Besides, this plant extract is very often processed by multistep procedures to isolate the major and minor ecdysteroids from the new or existing sources. A simplified scheme consisting of a few extraction steps for the purification of ecdysteroid from plants is in great demand. A quantitative approach through high-performance liquid chromatography has been initiated for developing an easy method for the extraction of ecdysteroids from *Ipomoea hederacea* (kaladana) seeds.

Girault J.P. (1998) Determination of ecdysteroid structure by 1D and 2D NMR. *Russian Journal of Plant Physiology* 45(3), 306-309.

Girault J.-P., Lafont R., Varga E., Hajdu Zs., Herke I and Szendrei K. (1988) Ecdysteroids from *Leuzea carthamoides*. *Phytochemistry* 27(3), 737-741.

Girault J.-P., Báthori M., Kalász H., Máthé I. and Lafont R (1996) Sidisterone, a C₂₄ ecdysteroid from *Silene dioica* and *Silene otites*. *Journal of Natural Products* 59(5), 522-524.

Abstract: Sidisterone (**1**), the first C₂₄ ecdysteroid, has been isolated from two species of the Caryophyllaceae in the genus *Silene* (*S. dioica* and *S. otites*) and identified through ¹H-, ¹³C-, and multidimensional NMR and molecular modeling techniques.

Girault J.-P., Báthori M., Varga E., Szendrei K. and Lafont R. (1990) Isolation and identification of new ecdysteroids from the Caryophyllaceae. *Journal of Natural Products* 53(2), 279-293.

Glashkin A.V., Sakipova Z.B., Sichkar A.A., Tuleuov B.I., Hasenova R.Z., Berkenov A.K. and Adekenov S.M. (2014) Ecdysteroids of *Silene gutensis* Feditsch plant and their physic-chemical research. *Vestnik Kaznmu* (5), 44-47 [in Russian, with an English abstract].

Glombitza K.-W., Kucera-Waldmann C. and Fricke U. (1989) Do roots of *Helleborus niger* contain cardioactive substances? *Planta Medica* 55, 107.

Poster Abstract.

Gorelick J., Iraqi R.H. and Bernstein N. (2020) Ecdysteroid content and therapeutic activity in elicited spinach accessions. *Plants* **9**, 727 (doi: 10.3390/plants9060727).

Abstract: While spinach is an established nutritionally important crop, its medicinal value is not as well known. Spinach is rich in ecdysteroids, insect hormone analogs with a number of medicinal properties including anti-oxidative, anti-inflammatory and even anabolic activity. However, the potential of spinach as a medicinal plant has not yet been developed. In this study, the ecdysteroid content of spinach was optimized to increase its therapeutic value. Spinach seeds from various sources were grown under controlled hydroponic conditions and analyzed for ecdysteroid content and related anabolic activity. Variations in ecdysteroid content and the related anabolic activity were observed among spinach accessions. A selected variety, *Spinacia oleracea* cv. Turkey, was exposed to various physical and chemical elicitors to increase and stabilize ecdysteroid content. A number of elicitors, including methyl salicylate and mechanical damage, significantly increased ecdysteroid content and anabolic activity 24 h after exposure. The effect was transient and disappeared 48 h thereafter. Further work is needed to identify the most suitable germplasm and elicitation conditions for optimal ecdysteroid content.

Gorovits M.B., Zatsny I.L. and Abubakirov N.K. (1974) Review: ecdysones in the plant world. *Rastitelny Resursy* **X**, 261-274 [in Russian].

Gosmann G., Gattuso S., Gattuso M., Fenner R., Pacheco E.F., Ferraz A., Savi L.A., Barardi C.R.M., Simoes C.M.O., Sortino M., Zacchino S., Gnerre C., Testa B. and Rates S.M.K. (2003) Botanical (morphological, micrographic), chemical and pharmacological characteristics of *Pfaffia* species (Amaranthaceae) native to South Brazil. *Revista Brasileira de Ciencias Farmaceuticas/Brazilian Journal of Pharmaceutical Sciences* **39**(2), 141-147.
Abstract: Some parameters for the quality control of *P. glomerata* and *P. paniculata* roots using their botanical and chemical characteristics are presented. It was also carried out an in vitro pharmacological screening to evaluate some biological properties of *P. glomerata* that could be related to its popular use as "tonic". Relating to biological assays, ethanolic extract from *P. glomerata* roots did not present antiviral, antiproliferative, antifungal or MAO inhibitory activities. The cytotoxicity evaluation of *P. glomerata* determined that IC₅₀ is >2,000 µg/mL. The main morphological and micrographic characteristics of *P. glomerata* and *P. paniculata* roots are described in this paper in order to aid in their unequivocal identification.

Graf B.L., Poulev A., Kuhn P., Grace M.H., Lila M.A. and Raskin I. (2014) Quinoa seeds leach phytoecdysteroids and other compounds with anti-diabetic properties. *Food Chemistry* **163**, 178-185.

Abstract: Quinoa (*Chenopodium quinoa* Willd.) contains high levels of biologically active phytoecdysteroids, which have been implicated in plant defense from insects, and have shown a range of beneficial pharmacological effects in mammals. We demonstrated that the most prevalent phytoecdysteroid, 20-hydroxyecdysone (20HE), was secreted (leached) from intact quinoa seeds into water during the initial stages of seed germination. Leaching efficiency was optimized by ethanol concentration (70% ethanol), temperature (80 °C), time (4 h), and solvent ratio (5 ml/g seed). When compared to extraction of macerated seeds, the leaching procedure released essentially all the 20HE available in the seeds (491 µg/g seed). The optimized quinoa leachate (QL), containing 0.86% 20HE, 1.00% total phytoecdysteroids, 2.59% flavonoid glycosides, 11.9% oil, and 20.4% protein, significantly lowered fasting blood glucose in obese, hyperglycemic mice. Leaching effectively releases and concentrates bioactive phytochemicals from quinoa seeds, providing an efficient means to produce a food-grade mixture that may be useful for anti-diabetic applications.

Grebenok R.J. and Adler J.H. (1991) Ecdysteroid distribution during development of spinach. *Phytochemistry* **30**(9), 2905-2910.

Abstract: During the growth and development of spinach (*Spinacia oleracea*), phytoecdysteroids are dynamically cycled between various leaves with an overall increase in total ecdysteroid content per plant. Within seeds, the embryo possesses 17 µg ecdysteroid per seed whereas the seed coat has only 1 µg ecdysteroid per seed. During the first 20 days of development the level of ecdysteroids on a µg per plant basis remains constant. After 20 days ecdysteroid content (µg per plant) increases with age. The physiological level of ecdysteroids within specific leaves cycles up and down with development on µg ecdysteroid per gram fresh weight basis. During this cycling ecdysteroids are synthesized in leaves subtending their site of accumulation, which is in the apical most leaves, as demonstrated by [2-¹⁴C]MVA radiolabel feed experiments. Movement of a post-sterol product, possibly ecdysteroids, is also supported by the radiolabel experiments. Transport of these products during growth and development of spinach would account for the physiological accumulation of ecdysteroids in the apical parts of the plant. The physiology of ecdysteroids observed in spinach provides the basis for a consistent interpretation of most previous reports of ecdysteroid levels in higher plants.

Grebenok R.J. and Adler J.H. (1993) Ecdysteroid biosynthesis during the ontogeny of spinach leaves. *Phytochemistry* **33**(2), 341-347.

Abstract: During the growth and development of spinach (*Spinacia oleracea*), phytoecdysteroid biosynthesis is observed later in the ontogeny of cotyledons, first and second leaves, whereas early in leaf ontogeny no biosynthesis of phytoecdysteroid is observed. An excised leaf assay for incorporation of various substrates [$2\text{-}^{14}\text{C}$]mevalonic acid (MVA), [$4\text{-}^{14}\text{C}$]cholesterol, [$2\text{-}^{14}\text{C}$]acetate and [$22,23\text{-}^3\text{H}$] α -ecdysone into phytoecdysteroids was developed. All four radiolabelled substrates can be incorporated into the ecdysteroids of spinach (20-hydroxyecdysone and polypodine B). The [$2\text{-}^{14}\text{C}$]MVA is apparently incorporated into the C_{27} -sterol lathosterol (5 α -cholest-7-en-3 β -ol) prior to radiolabel incorporation into spinach ecdysteroids. Leaves early in their ontogeny fail to produce radiolabelled lathosterol or ecdysteroids from [$2\text{-}^{14}\text{C}$]MVA in spite of an active C_{29} -sterol biosynthesis from [$2\text{-}^{14}\text{C}$]MVA. Conversion of [$22,23\text{-}^3\text{H}$] α -ecdysone into radiolabelled spinach ecdysteroids parallels the incorporation of [$2\text{-}^{14}\text{C}$]MVA into these ecdysteroids with leaf ontogeny. Lathosterol was structurally identified as a component of mature spinach leaves accounting for approximately 4% of the total sterol composition. The spinach excised leaf assay provides a model system for the study of ecdysteroid biosynthesis in plants.

Grebenok R.J., Ripa P.V. and Adler J.H. (1991) Occurrence and levels of ecdysteroids in spinach. *Lipids* **26**(8), 666-668.

Abstract: The dominant ecdysteroid in spinach, *Spinacia oleracea* L., is 20-hydroxyecdysone (2 β ,3 β ,14 α ,20R,22R,25-hexahydroxy-5 β -cholest-7-en-6-one) with the presence of a smaller amount of polypodine B (2 β ,3 β ,5 β ,14 α ,20R,22R,25-heptahydroxycholest-7-en-6-one). Ecdysteroids are present in the seed embryo at 14 μg ecdysteroid/seed, and ecdysteroid levels increase, in the plant during growth and development. During the onset of ecdysteroid production, the ratio of ecdysteroid to total sterol shifts from a value of one to about ten over the course of a week. Spinach may serve as a model to study ecdysteroid biosynthesis in plants.

Grebenok R.J., Venkatachari S. and Adler J.H. (1994) Biosynthesis of ecdysone and ecdysone phosphates in spinach. *Phytochemistry* **36**(6), 1399-1408.

Abstract: The polar ecdysteroid conjugate, ecdysone phosphate (2 β ,3 β ,14 α ,22R,25-pentahydroxy-7-en-6-one-3-phosphate) was identified in excised first leaves of spinach, where it is endogenously produced during 20-hydroxyecdysone biosynthesis. Radiolabelled [^{14}C]ecdysone phosphate was isolated from several excised leaf assays and was hydrolysed with wheat germ acid phosphatase to yield [^{14}C]ecdysone. Incorporation of ecdysone into excised first leaves followed by ^{32}P exposure produced a compound with ^{32}P activity, with chromatographic properties identical to those of the isolated [^{14}C]ecdysone phosphate and upon hydrolysis released ecdysone. In spinach first leaves with active ecdysteroid biosynthesis, ecdysone is present at 0.004% of the total free ecdysteroid and contained 6% of the total radioactivity from [$2\text{-}^{14}\text{C}$]mevalonic acid (MVA). These biosynthetically active tissues also produce radiolabelled lathosterol, ecdysone-3-phosphate and 20-hydroxyecdysone. In biosynthetically inactive tissue (immature apical organs) no radiolabelled lathosterol, ecdysone-3-phosphate, ecdysone or 20-hydroxyecdysone was produced from [$2\text{-}^{14}\text{C}$]MVA despite an active biosynthesis of C_{29} -sterols. Several intermediate and end product ecdysteroids, when incorporated into excised first leaves of spinach produced conjugates which were readily cleaved by wheat germ acid phosphatase. The ecdysteroid pathway appears to be regulated by the presence of ecdysteroid substrates.

Grebenok R.J., Galbraith D.W., Benveniste I., Feyereisen R. (1996) Ecdysone 20-monoxygenase, a cytochrome P_{450} enzyme from spinach, *Spinacia oleracea*. *Phytochemistry* **42**(4), 927-933.

Abstract: A microsomal preparation isolated from first leaves of 25-day-old spinach catalysed the hydroxylation of ecdysone to produce the insect moulting hormone, 20-hydroxyecdysone. Hydroxylation was dependent on NADPH and molecular oxygen, and was inhibited by carbon monoxide. Carbon monoxide inhibition was partially reversible by white light. Polyclonal antibodies to the Jerusalem artichoke NADPH-cytochrome P_{450} reductase inhibited the hydroxylation reaction as well as the spinach microsomal NADPH cytochrome c reductase. These results taken together establish ecdysone hydroxylation as a cytochrome P_{450} dependent reaction in spinach, which is known to synthesize large amounts of phytoecdysteroids.

Gu S., Zhang D., Xu L. and Yang S. (1997) Studies on the chemical constituents of *Podocarpus imbricatus* BI. (II). *Zhongguo Zhongyao Zazhi* **22**(3), 169-171 [in Chinese].

Abstract: From *Podocarpus imbricatus*, 5 compounds were isolated and identified as 2"-O-rhamnosylvitexin, hinokiflavone, ecdysone, β -sitosterol heptadecanoate, and daucosterol on the basis of spectral studies (UV, IR, ^1H NMR, ^{13}C NMR, MS) and chem. reactions.

Guibout L., Mamadaliyeva N., Balducci C., Girault J.-P. and Lafont R. (2015) The minor ecdysteroids from *Ajuga turkestanica*. *Phytochemical Analysis* **26** 293-300.

Abstract:

Introduction: *Ajuga turkestanica* is a plant used in traditional medicine for its high ecdysteroid content, including the presence of the particularly active turkesterone, which possess efficient anabolic activity.

Objectives: To isolate and identify minor ecdysteroids present in a semi-purified plant fraction containing ca. 70% turkesterone.

Material and methods: Multi-step preparative HPLC (combining RP- and NP-HPLC systems) was used to purify the different components present in the turkesterone fraction. Isolated compounds were identified by high-resolution mass spectrometry and 2D-NMR.

Results: Fourteen ecdysteroids (including turkesterone and 20-hydroxyecdysone) were isolated. Seven of these, all bearing an 11 α -hydroxy group, were previously unreported.

Conclusion: *Ajuga turkestanica* ecdysteroids are characterised by the abundance of 11 α -hydroxylated compounds and by the simultaneous presence of 24C, 27C, 28C and 29C ecdysteroids. It is expected that even more ecdysteroids are to be found in this plant since the starting material for this study lacked the less polar ecdysteroids. The simultaneous presence of 20-hydroxyecdysone and turkesterone (its 11 α -hydroxy analogue) as the two major ecdysteroids suggests that every ecdysteroid is probably present in both 11 α -hydroxy and 11-deoxy forms.

Guo D.-a. and Lou Z.-c. (1992) Separation and quantitative determination of three phytoecdysteroids in *Rhaponticum uniflorum* by high-performance liquid chromatography. *Journal of Chinese Pharmaceutical Sciences* **1**, 60-66.

Abstract: An analytical method for the separation and quantitative determination of three phytoecdysteroids in *Rhaponticum uniflorum* by high performance liquid chromatography has been developed. Three phytoecdysteroids, ecdysterone (I), rhapontisterone (II) and turkesterone (III) were separated and determined quantitatively by using the solvent system MeOH-H₂O (40:60) at 242 nm with flow rate of 1.5 ml/min, the operation can be completed in 10 minutes. Using the external standard method, the content of three phytoecdysteroids in the roots and aerial parts of *Rhaponticum uniflorum* obtained from different localities was determined. It is proved that this method is sensitive and accurate, and with good reproducibility.

Guo D.-A., Lou Z.C., Gao C.Y., Qiao L. and Peng J.R. (1991) Phytoecdysteroids of *Rhaponticum uniflorum* root. *Yaoxue Xuebao (Acta Pharmaceutica Sinica)* **26**(6), 442-446 [in Chinese, with an English abstract].

Abstract: Three phytoecdysteroids I, II and III were isolated from the root of *Rhaponticum uniflorum* (L.) DC. and their structures were elucidated by chemical and spectroscopic methods (UV, IR, EI-MS, FAB-MS, ¹HNMR, ¹³CNMR, 1H-1H COSY, 1H-1H NOESY, 1H-13CCOSY and CD). Compound II is new and named rhapontisterone, its structure was confirmed as (20R, 22R, 24S)-2 beta, 3 beta, 11 alpha, 14 alpha, 20, 22, 24-heptahydroxy-5 beta cholest-7-en-6-one. The other two, I and III, were identified as ecdysterone and turkesterone, respectively, both are known compounds, but turkesterone was isolated for the first time from the title plant.

Guo J.-y., Wang W.-z., Wu L.-j., Wang J.-w. Li Y.-n. and Gao H.-y. (2011) Isolation and identification of chemical constituents from *Taxus cuspidata* Sieb. et Zucc. (II). *Journal of Shenyang Pharmaceutical University* (5), 355-359 [in Chinese].

Guo K., Ren X., Mu R-F., Zhou T-T., Li D., Hu H., Liu Y. and Li S-H. (2021) Ecdysteroids and spirosterone steroids from the traditional Chinese medicine *Paris polyphylla* var. *yunnanensis*. *Phytochemical Letters* **45**, 117-120.

Abstract: Phytochemical investigation of the rhizomes of *Paris polyphylla* var. *yunnanensis* led to the isolation of four ecdysteroids (1– 4) including two new ecdysteroids (1– 2), and two known spirostane steroids (5– 6). Their structures were elucidated by extensive spectroscopic analyses as well as comparisons with those reported in the literature. All the isolates were assayed for their immunosuppressive and adipogenesis inhibitory activities but were inactive.

Guo Y.Y., Kojima K., Lin L.B., Fu X.W., Zhao C.Q., Hatano K., Chen Y.J. and Ogihara Y. (1999) A new N-methyltetrahydroprotoberberine alkaloid from *Tinospora hainanensis*. *Chemical and Pharmaceutical Bulletin* **47**(2), 287-289.

Abstract: Several species of the general *Tinospora* (Menispermaceae) are widely distributed over Asia and Africa and are well-known for their medicinal properties. From the stems of *Tinospora hainanensis*, a new N-methyltetrahydroprotoberberine alkaloid, N-methyltetrahydrocolumbamine, was isolated and characterized on the basis of NMR, MS and X-ray.

Gvazava L.N. and Kukoladze V.S. (2010) Phytoecdysteroids from *Digitalis ciliata* and *D. purpurea* leaves. *Chemistry of Natural Compounds* **46**(1) 146-147

Abstract: Ecdysteroids are chemically polyhydroxysteroids that contain specific structural elements such as 2,3-diol and 14 -hydroxy,7,6-ketone groups and cis-fused rings A and B. These features enable them to be classified as a

separate group of natural compounds [1]. Ecdysones exhibit a specific effect on insect metamorphosis processes. Thus, juvenile and molting hormones were discovered among this group of compounds [2, 3]. A third generation of insecticides that had excellent selectivity and lacked harmful environmental effects could be created by using these properties against insects [4, 5]. Herein we report results from a study of ecdysteroids isolated from mother liquors after separation of steroidal saponins and cardenolides, in particular acetyldigitoxin and digalen-neo from *Digitalis ciliata* [6] and *D. purpurea* [7], respectively. Cardenolides were exhaustively extracted from raw material (1 kg air-dried *D. ciliata* leaves and 0.8 kg, *D. purpurea*). The aqueous mother liquors were extracted with n-BuOH and purified from ballast substances by aqueous NaOH (5%). The purified n-BuOH extracts were condensed. The resulting precipitates were separated by filtration and dried to afford total ecdysteroids (5.84 g, 0.584% yield, *D. ciliata* and 3.92 g, 0.49% yield, *D. purpurea*). These were separated over columns of silica gel (L 100/160 m, Czech Rep.) and Al₂O₃. We used solvent systems CHCl₃:MeOH (15:1, 1; 9:1, 2; 4:1, 3) and CHCl₃:MeOH:H₂O (4:1:0.1, 4) [8]. Elution of total ecdysteroids from *D. ciliata* first by system 1 and then system 2 isolated polygodine B acetate and benzoate and polygodine B. Elution of the column by systems 3 and 4 produced 20-hydroxyecdysone acetate and benzoate and 20-hydroxyecdysone. Elution of total ecdysteroids from *D. purpurea* first by system 3 and then system 4 produced 20-hydroxyecdysone acetate and 20-hydroxyecdysone. Rechromatography of the total over a column of Al₂O₃ using systems 2 and 3 isolated viticosterone E. The compounds were identified by the following data. Polygodine B, C₂₇H₄₄O₈, mp 251–252°C (acetone), [J]_D 20 +92.2° (c 0.30, MeOH). UV spectrum: 245 nm (log 4.01). IR spectrum (KBr, , cm⁻¹): 3350–3450, 1687 (C=O), 1640. MS m/z 478 [M – H₂O]⁺. PMR spectrum (C₅D₅N, , ppm): 6.23 (1H, s, H-7), 4.0–4.3 (2H, H-2,3), 3.85 (H-22), 3.59 (H-9), 1.58 (CH₃-21), 1.37 (6H, CH₃-26,27), 1.20 (CH₃-18), 1.15 (CH₃-19) [9, 10]. Polygodine B-22-O-acetate, C₂₉H₄₆O₉, mp 149–152°C (MeOH:H₂O), [J]_D 20 +122° (c 0.28, MeOH). IR spectrum (KBr, , cm⁻¹): 3400–3500 (OH), 1685 (C=O), 1735, 1260 (ester). MS m/z 520 [M – H₂O]⁺. PMR spectrum (C₅D₅N, , ppm, J/Hz): 6.25 (1H, br.s, H-7), 5.49 (1H, d, J = 7.6, H-22), 4.10 (2H, m, H-2, H-3), 3.62 (1H, m, H-9), 2.02 (3H, s, OAc), CH₃: 1.65, 1.35, 1.18 (2) [8]. Polygodine B-22-O-benzoate, C₃₄H₄₈O₉, mp 196–198°C (MeOH:H₂O). IR spectrum (KBr, , cm⁻¹): 3430 (OH), 1655 (C=O), 1710, 1280 (ester), 1612, 1585, 715 (arom.). PMR spectrum (C₅D₅N, , ppm, J/Hz): 8.24 (2H, dd, J = 1.2, 7.6) and 7.35 (3H, br.m), aromatic protons; 6.17 (1H, br.s, H-7), 5.70 (1H, d, J = 7.5, H-22), 4.16 (2H, m, H-2, H-3), 3.52 (1H, m, H-9), CH₃: 1.65, 1.17 (2), 1.06, 1.02 [11]. 20-Hydroxyecdysone, C₂₇H₄₄O₇, mp 242–243°C (acetone), [J]_D 20 +62.0° (c 1.4, MeOH). UV spectrum: 243 nm (log 4.05). IR spectrum (KBr, , cm⁻¹): 3500–3350 (OH), 1660(7-6-ketone). MS m/z 462 [M – H₂O]⁺. PMR spectrum

Hang D.T.T., Hang N.T.M., Anh H.L.T., Nhiem N.X., Hue C.T., Binh P.T., Dat N.T., Nam N.H., Yen P.H., Minh C.V., Hung N.V. and Kiem P.V. (2015) ¹H and ¹³C NMR assignments of new ecdysteroids from *Callisia fragrans*. *Magnetic Resonance in Chemistry* **53**(5) 379-382 (doi: 10.1002/mrc.4214)

Hardman R. and Benjamin TV. (1976) The co-occurrence of ecdysones with bufadienolides and steroidal saponins in the genus *Helleborus*. *Phytochemistry* **15**, 1515-1516.

Abstract: A method is described for the detection of ecdysones in small quantities of plant material. When applied to 14 taxa of *Helleborus* L., ecdysterone and 5 β-hydroxyecdysterone were detected and quantified in 11; the other 3, which are morphologically distinct, gave a negative result. The co-occurrence of ecdysones with bufadienolides and steroidal saponins is discussed. The isolation of the ecdysones from the aerial parts of *H. orientalis* hybrids is described.

Hardman R. and Benjamin T.V. (1980) The detection of *Helleborus* ecdysteroids by mass spectrometry and by GLC analysis of their TSIM-ethers. *Planta Medica* **39**, 148-152.

Abstract: 20-Hydroxyecdysone (ecdysterone) (2β, 3β, 14α, 20R, 22R, 25-hexahydroxy-5β-cholest-7-en-6-one) and 5β,20-dihydroxyecdysone (Polygodine B) present in extracts of species of *Helleborus* were detected as their trimethylsilylimidazole (TSIM) derivatives by GLC analysis. The MS analysis of 5β,20-dihydroxyecdysone in the purified extract and the GLC characteristics of the silylated ecdysteroids are described and discussed as a means of their identification.

Hardman R. and Mahar S.M.A. (1978) Isolation, identification and quantification of ecdysones from *Aerva tomentosa* and *Pandiaka involucreta*. *Planta Medica* **33**, 278-279.

He G-x., Liang X-l., Ouyang W., Yi G-q., Li Y-y., Zhao J-p., Khan I. (2013) Chemical constituents from *Ajuga nipponensis*. *Journal of Chinese Medicinal Materials* (12), 1950-1953 [in Chinese, with and English abstract].

Abstract: Chem. constituents of *Ajuga nipponensis* were studied. The chem. constituents were isolated by repeated silica gel column chromatog. and their structures were elucidated by physiochem. properties and spectral anal. Ten compounds were isolated and identified as: hexadecanoic acid (1), ajuforrestin A (2), β-sitosterol (3), acacetin (4), apigenin (5), ajugamacrin B (6), ursolic acid (7), β-ecdysone (8), 8-acetylharpagide (9) and daucosterol (10). Compounds 1-7 and 10 are isolated from this plant for the first time.

Heftmann E., Sauer H.H. and Bennett R.D. (1968) Biosynthesis of ecdysterone from cholesterol by a plant. Die Naturwissenschaften 55, 37-38.

Heinrich G. and Hoffmeister H. (1967) Ecdyson als Begleitsubstanz des Ecdysterons in *Polypodium vulgare* L. [Ecdysone accompanies ecdysterone in *Polypodium vulgare* L.] Experientia 23, 995 [in German].

Abstract: Ecdysone, one of the moulting hormones of Arthropodes, was found for the first time in the plant kingdom too. From rhizomes of *Polypodium vulgare* L. we isolated 0.002% ecdysone and 0.07% ecdysterone.

Heinrich G. and Hoffmeister H. (1968) 5 β -Hydroxyecdysterone, ein Pflanzensteroid mit Häutungshormonaktivität aus *Polypodium vulgare* L. [5 β -hydroxyecdysterone, a plant steroid with moulting hormone activity from *Polypodium vulgare* L.] Tetrahedron Letters (58), 6063-6064 [in German].

Die Rhizomen des Farnes *Polypodium vulgare* enthalten die beiden von den Prothorakaldriisen der Insekten gebildeten Steroidhäutungshormone Ecdysterone und Ecdyson (1,2). Wir konnten jetzt zwei weitere Steroide aus der handelsüblichen Droge (Rhizoma Polypodii depur.conc.) isolieren. Wie die Strukturaufklärung ergab, handelt es sich bei der einen Substanz um 5 β -Hydroxyecdysterone (I). Mit I, das in einer Menge von 400 mg/kg Rhizomen vorliegt, steht ein neues Häutungshormon zur Verfügung, dessen Aktivität gleich groß ist wie die der wirksamsten Hormone dieser Stoffgruppe, des Ecdysterons (II): 0,005 μ g lösen im Calliphora-Test (3) die Verpuppung aus.

Hikino H. (1976) Ecdysterone and ecdysone from *Polypodium virginianum*. Journal of Natural Products 39, 246-247.

Hikino H., Hikino Y., Nomoto K. and Takemoto T. (1968) Cyasterone, an insect metamorphosing substance from *Cyathula capitata*: structure. Tetrahedron 24, 4895-4906.

Abstract: A novel C₂₉ insect-metamorphosing substance, cyasterone, has been isolated from *Cyathula capitata* (Amaranthaceae). Chemical and physico-chemical studies of cyasterone and its derivatives (II-V), and in particular its transformation into 14 α -hydroxy-2,3-seco-5 β -pregn-7-ene-6,20-dione-2,3-dial (VI) and 2,4-dimethyl-3-(2-oxoethyl)-4-butanolide (VII) have established the structure of cyasterone as shown in formula I.

Hikino H., Nomoto K. and Takemoto T. (1969a) Structure of sengosterone, a novel C₂₉ insect-moulting substance from *Cyathula capitata*. Tetrahedron Letters (18), 1417-1420.

No Abstract.

Hikino H., Kohama T. and Takemoto T. (1969b) Biosynthesis of ponasterone A, an insect-moulting substance from *Podocarpus macrophyllus*. Chemical and Pharmaceutical Bulletin 17, 415.

No Abstract.

Hikino H., Arihara S. and Takemoto T. (1969c) Ponasteroside A, a glycoside of insect metamorphosing substance from *Pteridium aquilinum* var. *latiusculum*: structure and absolute configuration. Tetrahedron 25, 3909-3917.

No Abstract.

Hikino H. and Hikino Y. (1970) Arthropod molting hormones. Fortschritte der Chemie organischer Naturstoffe 28, 256-312.

Abstract: While much is known about the hormones of vertebrates, knowledge of the hormones of invertebrates is far less complete. However, the chemistry of the molting hormones and the juvenile hormones of insects has made surprisingly rapid advances during the past few years and has now become a subject of research which is attracting the interest of both chemists and biologists. This review article is an attempt to summarize recent developments in our knowledge regarding the chemistry, synthesis and metabolism of the arthropod molting hormones, but will in the main exclude consideration of biological properties which have been frequently the object of excellent reviews (71).

Hikino H., Kohama T. and Takemoto T. (1970) Biosynthesis of ponasterone A and ecdysterone from cholesterol in *Podocarpus macrophyllus*. Phytochemistry 9, 367-369.

Abstract: Administration of cholesterol-4-¹⁴C to *Podocarpus macrophyllus* seedlings resulted in the formation of the insect-metamorphosing substances ponasterone A and ecdysterone in radioactive form.

Hikino H., Nomoto K. and Takemoto T. (1970b) Poststerone, a metabolite of insect metamorphosing substances from *Cyathula capitata*. Steroids 16, 393-400.

No Abstract.

Hikino H., Nomoto K., Ino R. and Takemoto T. (1970c) Structure of precyasterone, a novel C₂₉ insect moulting substance from *Cyathula capitata*. Chemical and Pharmaceutical Bulletin 18(5), 1078-1080.

No Abstract.

Hikino H., Nomoto K. and Takemoto T. (1970d) Sengosterone, an insect metamorphosing substance from *Cyathula capitata*: structure. Tetrahedron 26, 887-898.

Abstract: A novel C₂₉ insect metamorphosing substance, sengosterone, has been isolated from *Cyathula capitata* (Amaranthaceae) and shown to have structure I by chemical and physico-chemical studies.

Hikino H., Nomoto K. and Takemoto T. (1971) Cyasterone, an insect metamorphosing substance from *Cyathula capitata*: absolute configuration. Tetrahedron 27, 315-321.

Abstract: Chemical and physico-chemical studies on cyasterone and its derivatives (IV–VII) have established the stereostructure of cyasterone as shown in formula II.

Hikino H., Nomoto K. and Takemoto T. (1971b) Structure of isocyasterone and epicyasterone, novel C₂₉ insect-moulting substances from *Cyathula capitata*. Chemical and Pharmaceutical Bulletin 19(2), 433-435.

Hikino H., Nomoto K. and Takemoto T. (1971c) Isocyasterone, an insect metamorphosing substance from *Cyathula capitata*. Phytochemistry 10, 3173-3178.

Abstract: A novel C₂₉ insect-metamorphosing substance, isocyasterone, has been isolated from *Cyathula capitata* (Amaranthaceae). Chemical and spectroscopic investigations of isocyasterone and its triacetate (II) have shown that isocyasterone has stereostructure I.

Hikino, H., Jin, H., & Takemoto, T. (1971d) Occurrence of insect-moulting substance ecdysterone and inokosterone in callus tissues of *Achyranthes*. Chemical and Pharmaceutical Bulletin 19(2), 438-439.

No Abstract.

Hikino H., Okuyama T., Jin H. and Takemoto T. (1973) Screening of Japanese ferns for phytoecdysones. I. Chemical and Pharmaceutical Bulletin 21, 2292-2302.

Abstract: Japanese ferns from 20 families, representing 76 genera, 283 species, 39 varieties, and 1 form, have been subjected to screening tests by means of bioassay for the presence of phytoecdysones. A total of 170 species, 22 varieties, and 1 form have been found to show the insect moulting hormone activity. The taxonomical relationship is discussed.

Hikino H., Jin H. and Takemoto T. (1975a) Tissue culture of *Achyranthes* and formation of phytoecdysones in cultured tissues. Yakugaku Zasshi 95, 581-589 [in Japanese, with an English abstract].

Hikino H., Okuyama T., Arihara S., Hikino Y., Takemoto T., Mori H. and Shibata K. (1975b) Shidasterone, an insect metamorphosing substance from *Blechnum niponicum*: structure. Chemical and Pharmaceutical Bulletin 23(7), 1458-1479.

Abstract: The structure of shidasterone, the phytoecdysone isolated from *Blechnum niponicum* (Blechnaceae) has been studied. In connection with this, stereoisomers of the cholestane-20, 22-diol derivatives in regard to C-20 and C-22 have been synthesized and their chemical and spectral properties have been examined. Chemical and physico-chemical data and, in particular, the ¹³C NMR spectrum of shidasterone have revealed that shidasterone is identified as 22, 25-oxido-5 β -cholest-7-en-6-one-2 β , 3 β , 14 α , 20-tetraol (1).

Hikino H., Jin H. and Takemoto T. (1975c) Biosynthesis of inokosterone and ecdysterone from cholesterol and mevalonic acid in *Achyranthes*. Yakugaku Zasshi 95(5), 590-595 [in Japanese, with an English abstract].

Abstract: Incorporation of [4-¹⁴C]cholesterol and [2-¹⁴C]mevalonic acid lactone into the phytoecdysones, inokosterone and ecdysterone, in *Achyranthes fauriei* seedlings and their homogenate, respectively, was demonstrated.

Hikino H., Mohri K., Okuyama T., Takemoto T and Yen K-Y. (1976a) Phytoecdysones from *Diplazium donianum*. Steroids 28(5), 649-654.

Abstract: From *Diplazium donianum*, makisterone A, makisterone D, and an unidentified stereoisomer of makisterone B have been isolated. The presence of two other unidentified phytoecdysones has been noted.

Hikino H., Mohri K., Hikino Y., Arihara S. and Takemoto T. (1976b) Inokosterone, an insect metamorphosing substance from *Acyranthes fauriei*: absolute configuration and synthesis. *Tetrahedron* **32**, 3015-3021.

Ho R., Girault J.-P., Cousteau P.-Y., Bianchini J.-P., Raharivelomanana P. and Lafont R. (2008) Isolation of a new class of ecdysteroid conjugates (glucosyl-ferulates) using a combination of liquid chromatographic methods. *Journal of Chromatographic Science* **46**, 102-110.

Abstract: The Polynesian medicinal fern *Microsorium membranifolium* contains very large amounts of ecdysteroids, including ecdysone, 20-hydroxyecdysone, 2-deoxy-20-hydroxyecdysone, and 2-deoxyecdysone. It also contains large amounts of unusual ecdysteroids which have been unambiguously identified by mass spectrometry and nuclear magnetic resonance. A new class of ecdysteroid conjugates (3-glucosyl-ferulates of 2-deoxyecdysone and 2-deoxy-20-hydroxyecdysone) is isolated, together with a new glycoside (2-deoxyecdysone 25-rhamnoside). The simultaneous presence of a sugar and an aromatic moiety results in a very particular chromatographic behavior of these conjugates. They behave like flavonoids and polyphenols when using the classical purification on polyamide, aimed at removing the latter from crude plant extracts, and would therefore be lost. They elute as non-polar ecdysteroids on reversed-phase high-performance liquid chromatography (RP-HPLC), whereas their behavior on normal-phase (NP) HPLC is strongly dependent on the mobile phase composition. Our data highlight the importance of selectivity in the choice of HPLC methods used for ecdysteroid separations.

Ho R., Teai T., Loquet D., Bianchini J.-P., Girault J.-P., Lafont R. and Raharivelomanana P. (2007) Phytoecdysteroids in the genus *Microsorium* (Polypodiaceae) of French Polynesia. *Natural Product Communications* **2**, 803-806.

Abstract: A chemical survey of the six species of *Microsorium* in French Polynesia has been performed to determine and quantify the phytoecdysteroids. The content and composition of these compounds in the fronds of each of the six species were established. The highest concentrations of ecdysteroids were found in *M. membranifolium* (1.6% w/w) and *M. scolopendria* (0.47%), used in Polynesian traditional medicine. Seven phytoecdysteroids were quantified in these species and the major components were ecdysone, 20-hydroxyecdysone, and 2-deoxy-20-hydroxyecdysone, besides the minor ones (inokosterone, makisterone A, makisterone C, and 2-deoxyecdysone). Both the fronds of *M. membranifolium* and the rhizomes of *M. scolopendria* grown in French Polynesia could be considered as uncommonly rich sources of ecdysteroids.

Ho R., Girault J.-P., Raharivelomanana P. and Lafont R. (2012) E- and Z-Isomers of new phytoecdysteroid conjugates from French Polynesian *Microsorium membranifolium* (Polypodiaceae) fronds. *Molecules* **17** 11598-11606.

Abstract: Phytochemical investigation of the fronds of *Microsorium membranifolium* resulted in the isolation of a new phytoecdysteroid, E-2-deoxy-20-hydroxyecdysone 3-[4-(1-β-D-glucopyranosyl)]-caffeate (1), together with two known phytoecdysteroids, E-2-deoxy-20-hydroxyecdysone 3-[4-(1-β-D-glucopyranosyl)]-ferulate (2), E-2-deoxyecdysone 3-[4-(1-β-D-glucopyranosyl)]-ferulate (3). Their respective Z-isomers 4-6 were also observed and identified for the first time. The new structures were elucidated on the basis of extensive spectroscopic data analysis (1D, 2D-NMR and HR-MS techniques).

Hoffmeister H., Heinrich G., Staal G.B. and van der Burg W.J. (1967) Über das Vorkommen von Ecdysteron in Eiben [About the presence of ecdysterone in yew]. *Experientia* **54**, 471 [in German].
No Abstract.

Hou S.-s., Wang G.-l. and Xia K.-m. (1980) Preliminary studies on phytoecdysones of *Murdannia triquetra* (Wall) Brückn. *Acta Botanica Sinica* **22**(2), 207-208.
No Abstract.

Hou S.-s., Wang G.-l. and Xia K.-m. (1981) Further studies on phytoecdysone of *Murdannia triquetra* (Wall) Brückn. *Acta Botanica Sinica* **23**(2), 166-168 [in Chinese].
No Abstract.

Hou S.-s., Wang G.-l. and Xia K.-m. (1982) The isolation and identification of phytoecdysones from *Dacrydium pierrei* Hickel. *Acta Botanica Sinica* **24**(4), 347-354 [in Chinese, with an English abstract].

Abstract: Three crystals were isolated from the bark of *Dacrydium pierrei* Hickel and were identified by melting point spectral data (UV, IR, MS, NMR) and GC. Hplc. Crystal I is β-ecdysone, Crystal II is a jugasterone C. Crystal III consists of ponasterone A (IIIA) and a new phytoecdysone named dacryhainansterone (IIIB). Total yield of three crystals is 0.4%.

Hsieh C-W., Ko W-C., Chang C-K., Chen G-J. and Tsai J-C. (2016) Antioxidant and hepatoprotective effects of *Ajuga nipponensis* extract by ultrasonic-assisted extraction. *Asian Pacific Journal of Tropical Medicine* 9(5), 420-425.

Abstract:

Objective: To investigate suitable condition for extraction of the active components from *Ajuga nipponensis* (*A. nipponensis*).

Methods: Orthogonal experimental design was used to determine the optimal extraction parameters for ecdysterones and flavonoids. Finally, the hepatoprotective abilities of *A. nipponensis* extracts were evaluated by CCl₄-induced animal models.

Results: Maximum yields of flavonoids (7.87 ± 0.10) mg/g and ecdysterones (0.73 ± 0.02) mg/g could be obtained when the extraction time was 50 min, the extraction temperature was 60 °C, and the ratio of sample to 70% (v/v) ethanol was 1:20 (w/w). The antioxidant property of *A. nipponensis* was correlated to the concentration of its extracts. At 5 mg/mL, *A. nipponensis* extract scavenged 84.8% of DPPH radical and had absorbance values of 2.43 ± 0.04 reducing power. Upon CCl₄-induced liver injury, glutamic oxaloacetic transaminase and glutamic pyruvic transaminase decreased significantly after the mice were treated with *A. nipponensis*. Histological researches also explained that *A. nipponensis* reduced the extent of liver lesions induced by CCl₄.

Conclusions: *A. nipponensis* exhibited potent antioxidant activity in chemical experimental models and hepatoprotective effect against CCl₄-induced liver damage.

Hu J., Shi X., Mao X., Li H., Chen J. and Shi J. (2014) Ecdysteroids from the ethanol extract of *Diplopterygium rufopilosum*. *Phytochemistry Letters* 8 73–76.

Abstract: Phytochemical investigation of the ethanol extract of *Diplopterygium rufopilosum* resulted in the isolation of three new ecdysteroids, (22R,24R,25S,26S)-2 beta,3 beta,14 alpha,20R-tetrahydroxy-26 alpha-methoxy-6-oxo-stigmast-7-ene-22,26-lactone (1), (22R, 24R, 25S)-2 beta,3b,14a,20R,26S-pentahydroxy-6-oxo-stigmast-7-ene-22,26-lactone (2), and (22R, 25S)-2 beta,3 beta, 14 alpha,20R,24S-pentahydroxy-6,26-dioxo-stigmast-7-ene-22,26-lactone (3), together with two known compounds (4) and (5). Their structures were determined on the basis of spectroscopic analyses, including 1D-NMR, 2D-NMR, and HR-ESI-MS. The isolated ecdysteroids were evaluated in vitro for antimicrobial properties, and exhibited moderate antibacterial activities against the tested oral pathogens.

Hu K., Wang Y., Zhang Z. and Fu D. (2018) Determination of *Achyranthes bidentata* Bl's steroids by high-performance liquid chromatography. *Guangdong Chemical Industry* (5), 47-49 [in Chinese].

Abstract: A high-performance liquid chromatographic method was developed for the separation from *Cyathula officinalis* Kuan and *Achyranthes bidentata* Blume of cyasterone and β -ecdysterone. The analysis column was CN chromatographic column and the device was Agilent LC1200. As a result, chromatographic conditions were that mobile phase was chromatographically pure acetonitrile, the flow-rate was 1.5 mL/min, the column temperature was 30°C and detection wavelength was 242 nm. Under such conditions, the relationship between cyasterone's concentration and peak area was preferably linear, and regression equation was $y=15.8 x-21.14$, $R^2=0.9992$; the relationship between β -ecdysterone's concentration and peak area was preferably linear, and regression equation was $y=10.843 x-462.17$, $R^2=0.9894$. This method was accurate, sensitive and rapid, and it has been successfully applied to measure *Cyathula officinalis* Kuan and *Achyranthes bidentata* Blume cyasterone and β -ecdysterone.

Hu L., Fang L., Li R. and Wang X. (2020) Determination of β -ecdysterone in characteristic ethnic medicine *Achyranthes aspera*. *China Pharmacist* (8), 1625-1627 [in Chinese].

Abstract: Objective: To establish a method for determining the content of β -moulting steroids in the characteristic national medicine ox-knee (rough-haired ox-knee, wild ox-knee and willow ox-knee). Methods: The sample was extracted with 70% methanol reflux, with Symmetry C18 column (250 mm \times 4.6 mm, 5 μ m) as the column of colour spectrum, and acetylene-0.1% formic acid solution (15:85) as the column. Flow phase, flow rate: 1.0 ml.min⁻¹, sample: 10 μ l, detection wavelength: 250 nm, column temperature: 35 degrees C. Results: β -moulting ketones at 0.048 to 1.1.5 There is a good linear relationship in the range of 940 sg ($r=1.000 0$), with an average recovery rate of 94.17 per cent and an RSD of 0.93 per cent (n=6). The 24 batches of soil cow knee samples collected were measured, with coarse-haired ox-knees and wild ox-knee and willow ox-knee content range is 0.053 1% to 0.122 9%, 0.069 3% to 0.092 0% and 0.085 2% to 0.207 0%; In the mixed counterfeit Guangdong tu niu knee and Sichuan cow knee were not detected β -moulting steroids. Conclusion: The method has good repetition and recovery rate and can be used as a quantitative analysis method for β -moulting steroids in ox-knee.

Huang H., Wang X-q., Du Y. and Li C-f. (2017) Simultaneous determination of six components in the flowers of *Rhaponticum uniflorum* by HPLC. *Chinese Journal of Pharmaceutical Analysis* (6), 956-961 [in Chinese].

Abstract: Objective: To establish an HPLC method for simultaneous determination of quercetin-3-O- α -L-rhamnoside, luteolin, apigenin, 5,7,4'-trihydroxy-3'-methoxyflavone, ecdysterone and hemislin B glucoside in the

flowers of *Rhaponticum uniflorum*. Methods: The analysis was performed on an YMC-Pack C₁₈ column (4.6 mm×250 mm, 5.0 μm) at 35°C, eluted with a gradient program using acetonitrile (A) 0.2% phosphoric acid aqueous solution (B) as the mobile phase. The flow-rate was 0.8 mL·min⁻¹, and the detection wavelength was 254 nm. Results: The linear ranges of six components in the flowers of *R. uniflorum* were 0.090-2.24 μg (r=1.000 0) for quercetin-3-O-α-L-rhamnoside, 0.048-1.19 μg (r=0.999 8) for luteolin, 0.040-1.00 μg (r=0.999 9) for apigenin, 0.008-0.20 μg (r=0.999 8) for 5,7,4'-trihydroxy-3'-methoxyflavone, 0.080-2.00 μg (r=1.000 0) for ecdysterone, and 0.092-2.31 μg (r=1.000 0) for hemislin B glucoside. The average recoveries of six components were between 99.2%-103.9% with RSD of 0.82%-2.9%. The contents of quercetin-3-O-α-L-rhamnoside, luteolin, apigenin, 5,7,4'-trihydroxy-3'-methoxyflavone, ecdysterone and hemislin B glucoside in eight batches of the flowers of *R. uniflorum* were 0.66%-1.26%, 0.19%-0.60%, 0.08%-0.24%, 0.03%-0.05%, 0.31%-0.99%, and 0.42%-2.26%. Conclusion: The method established in this research for determination of the six components was feasible and suitable to evaluate the quality of the flowers of *R. uniflorum*.

Huang M-f., Li N. and Jia X-g. (2008) Progress in studies of *Rhaponticum carthamoides*. Journal of Shenyang Pharmaceutical University 7 17pp

Abstract: Objective To review the studies on biological characteristics, chemical constituents, pharmacological effects of *Rhaponticum carthamoides*. Methods On the basis of more than 40 published literatures within 30 years, the biological characteristics of *Rhaponticum carthamoides* were briefly reviewed. The chemical constituents were classified according to their structural types; their pharmacological effects were also summarized. Results The *Rhaponticum carthamoides* was a light-favored plant with good adaptability to light conditions, and it contained many kinds of chemical constituents, primarily flavonoids, phytoecdysteroids, triterpenoids, phenolic acids, essential oil and polysaccharides etc. It had been proved that *Rhaponticum carthamoides* possessed the cell-growth promoting, hypolipidemic, antiatherosclerotic, antimicrobial, antitumor, immunotropic, tonic, and anti-stress regulation effects. Conclusions Many studies have been performed on the chemical constituents and pharmacological actions of *Rhaponticum carthamoides* so far, and further researches on biological activities and its mechanism are to be continued.

Huang X-c., Guo Y-w., Zhou W-l., Zuo J-p. and Wang Z-t. (2003). Ecdysteroids from the stems of *Diploclisia glaucescens*. Tianran Chanwu Yanjiu Yu Kaifa (Natural Product Research and Development) 15(2), 93-97.

Abstract: Five ecdysteroids were isolated from the EtOH extract of the stems of *Diploclisia glaucescens*. Their structures were identified as paristerone 20,22 monoacetone(1), paristerone(2), β ecdysterone(3), makisterone C(4) and capitasterone(5), respectively, on the basis of chemical and physical evidence. This is the first report of paristerone 20,22 monoacetone isolated from natural source. Compounds 2,4, and 5 were also isolated for the first time from *D. glaucescens*.

Huang X., Gao W., Gu K. and Ma C. (2009) Chemical constituents of *Paris pubescens*. Zhongcaoyao 40(9), 1366-1369 [in Chinese].

Abstract: Chem. constituents of *Paris pubescens* stems and roots were researched. Solvent method was applied to extract, normal phase silica gel chromatog., Sephadex LH-20 and RP-HPLC were used to isolate and purify, and structure was identified by ¹H-NMR and ¹³C-NMR. Eleven components were isolated from Et acetate layer and n-butanol layer. They were β-sitosterol (I), stigmaterol (II), di-Bu phthalate (III), β-ecdysone (IV), pennogenin-3-O-α-L-binofuranosyl (1→4)-[α-L-rhamnopyranosyl-(1→2)]-β-D-glucopyranoside (V), diosgenin-3-O-α-L-binofuranosyl (1→4)-[α-L-rhamnopyranosyl-(1→2)]-β-D-glucopyranoside (VI), pennogenin-3-O-α-L-binofuranosyl (1→4)-β-D-glucopyranoside (VII), diosgenin-3-O-α-L-binofuranosyl (1→4)-β-D-glucopyranoside (VIII), pennogenin-3-O-α-L-rhamnopyranosyl(1→4)-α-L-rhamnopyranosyl(1→4)-[α-L-rhamnopyranosyl-(1→2)]-β-D-glucopyranoside (IX), quercetin (X) and kaempferol (XI). Compound I-XI were isolated from the plant for the first time, and compound III, X were isolated from *Paris* for the first time.

Huang X., Gao W., Man S., Zuo J. and Wang Y. (2009) Chemical constituents from herbs of *Paris verticillata*. China Journal of Chinese Materia Medica (14), 1812-1815 [in Chinese].

Abstract: Objective: To study the chemical constituents in herbs of *Paris verticillata*. Method: The compounds were isolated by column chromatography with silica gel and purified by Sephadex LH-20 and RP-HPLC. The structures were identified by means of NMR analysis. Results: Nine compounds were isolated from the EtOAc extract and the n-BuOH extract of *P. verticillata*. Their structures were identified as β-sitosterol (1), stigmaterol (2), daucosterol (3), β-ecdysterone (4), 4-hydroxymethyl-γ-butyrolactone (5), diosgenin-3-O-α-L-arabinofuranosyl(1→4)-[α-L-rhamnopyranosyl(1→2)]-β-D-glycopyranoside (6), pennogenin-3-O-α-L-arabinofuranosyl(1→4)-β-D-glycopyranoside (7), pennogenin-3-O-α-L-arabinofuranosyl(1→4)-[α-L-rhamnopyranosyl(1→2)]-β-D-glycopyranoside (8) and pennogenin-3-O-α-L-rhamnopyranosyl(1→4)-α-L-rhamnopyranosyl(1→4)-[α-L-rhamnopyranosyl(1→2)]-β-D-glycopyranoside (9). Conclusion: Compounds 1-9 are isolated from *P. verticillata* for

the first time, and compounds 3 and 5 are isolated from the genus *Paris* for the first time. The compounds 6-9 showed certain inhibition activeness of LA-795 cells, especially, the effects of compounds 6,8 and 9 were more significant.

Huang X., Gao W., Zhao W., Zhang T. and Xu J. (2010) Flavone and steroid chemical constituents from rhizome of *Paris axialis*. *Zhongguo Zhongyao Zazhi* 35(22), 2994-2998 [in Chinese].

Abstract: This paper is to study the chem. constituents from the rhizome of *Paris axialis*. The compounds were isolated by column chromatog. with silica gel and purified by Sephadex LH-20 column chromatog. and preparative RP-HPLC. The structures were identified by means of spectroscopic methods. 14 Compounds were isolated from the EtOAc extract and the n-BuOH extract of *P. axialis*. Their structures were identified as daucosterol (1), stigmasterol-3-O- β -D-glycopyranoside (2), β -ecdysterone (3), pennogenin-3-O- α -L-arabinofuranosyl(1->4)-[α -L-rhamnopyranosyl(1->2)]- β -D-glycopyranoside (4), diosgenin-3-O- α -L-rhamnopyranosyl(1->4)- α -L-rhamnopyranosyl(1->4)[α -L-rhamnopyranosyl(1->2)]- β -D-glycopyranoside (5), kaempferol (6), rutin (7), myricitrin (8), 4,2',4'-trihydroxychalcone (9), isorhamnetin-3-O- β -D-glycopyranoside (10), isorhamnetin-3-O- α -L-rhamnopyranosyl(1->2)- β -D-glycopyranoside (11), isorhamnetin-3-O- β -D-glucopyranosyl(1->6)- β -D-glycopyranoside (12), kavaflavone (13), and amentoflavone (14). Compounds 1-3 and 6-14 are isolated from *P. axialis* for the first time; and compounds 7-10, 13, 14 are isolated from the genus *Paris* for the first time.

Hunyadi A., Tóth G., Simon A., Mák M., Kele Z., Máthé I. and Báthori M. (2004) Two new ecdysteroids from *Serratula wolffii*. *Journal of Natural Products* 67 1070-1072.

Abstract: 11 α -Hydroxypoststerone (1) and herkesterone (2), two new natural ecdysteroids, were isolated from the herb *Serratula wolffii*. The former compound is the first 11-hydroxylated C21 ecdysteroid, while the latter is a new ecdysteroid with a 7,9(11)-dien-6-one chromophore. Their structures were determined using a combination of spectroscopic techniques.

Hunyadi A., Gergely A., Simon A., Toth G., Veress G. and Bathori M. (2007a) Preparative-scale chromatography of ecdysteroids of *Serratula wolffii* Andrae. *Journal of Chromatographic Science* 45, 76-86.

Abstract: Numerous ecdysteroids are isolated from the herb of *Serratula wolffii* Andrae, a cultivated plant. The isolation procedure includes a variety of low-pressure liquid chromatography, thin-layer chromatography (TLC), gel chromatography, and high-performance liquid chromatography (HPLC) methods. The progress of separation is monitored by TLC, and the final proof of purity is carried out by HPLC. The isolation process involves the removal of proteins, flavonoids, chlorophylls, other sterines, etc. The purification also includes the separation of the target ecdysteroids from each other. Isolation of the pure compounds requires 2-8 chromatographic steps. The consecutive steps are based on the different physicochemical properties of the ecdysteroids. In some cases, a special peak-cut method employing a flush of dichloromethane into the dichloromethane-isopropanol-water mobile phase is used. This flush of dichloromethane leads to an almost perfect separation of otherwise unresolved peaks. Two ecdysteroids, 25-hydroxydacryhainansterone and 14-epi-20-hydroxyecdysone, are identified as natural products for the first time. The structure-chiroptical relationships for some ecdysteroids are also discussed.

Hunyadi A., Báthori M. and Kálmán S. (2007b) Ecdysteroid sources in the Carpathian basin. 11-Hydroxy substituted ecdysteroids from *Serratula wolffii* Andrae. *Acta Pharmaceutica Hungarica* 77(1), 11-18 [in Hungarian, with an English abstract].

Abstract: Seven ecdysteroids have been isolated from the methanolic extract of the herb *Serratula wolffii* Andrae. The isolation process involved the removal of polar and apolar contaminants, and also included the separation of the target ecdysteroids from each other by using combined chromatographic methods. Isolation of the pure compounds required a minimum of 2 and a maximum of 7 consecutive chromatographic steps with different selectivity. Six of the isolated ecdysteroids showed the presence of an 11 α -hydroxyl group on the steroidal skeleton, which structure may enhance the increasing effect of ecdysteroids on the protein synthesis of mammals, according to literature data. Ajugasterone C, one of these compounds was found to be present in remarkable amount in the plant. One ecdysteroid, 11 α -hydroxypoststerone was identified as a new natural compound.

Hunyadi A., Herke I., Lengyel K., Báthori M., Kele Z., Simon A., Tóth G. And Szendrei K. (2016) Ecdysteroid-containing food supplements on the European market: evidence for spinach product counterfeiting. *Scientific Reports* (DOI: 10.1038/srep37322).

Abstract: Phytoecdysteroids like 20-hydroxyecdysone ("ecdysterone") can exert a mild, non-hormonal anabolic/adaptogenic activity in mammals, and as such, are frequently used in food supplements. Spinach is well-known for its relatively low ecdysteroid content. *Cyanotis arachnoidea*, a plant native in China, is among the richest sources of phytoecdysteroids, and extracts of this plant are marketed in tons per year amounts via the internet at highly competitive prices. Here we report the investigation of a series of food supplements produced in Germany and claimed to contain spinach extracts. Twelve ecdysteroids including two new compounds were isolated and utilized as

marker compounds. A comparative analysis of the products with Cyanotis and spinach extracts provides evidence that they were manufactured from Cyanotis extracts instead of spinach as stated. Based on the chromatographic fingerprints, 20-hydroxyecdysone 2- and 3-acetate are suggested as diagnostic markers for related quality control. This case appears to represent an unusual type of dietary supplement counterfeiting: undeclared extracts from alternative plants would supposedly 'guarantee' product efficacy.

Hussein I.A., Srivedavyasari R., El-Hela A.A., Mohammad A.I. and Ross S.A. (2020) Chemical constituents from *Silene schimperiana* Boiss. belonging to Caryophyllaceae and their chemotaxonomic significance. *Biochemical Systematics and Ecology* 92, article 104113, pp4.

Abstract: Phytochemical investigation of *Silene schimperiana* Boiss. ethanolic extract led to the isolation of fifteen compounds (1–15). The isolated compounds were identified by their NMR, MS spectral data analyses and comparing with published data as: vanillic acid (1), ferulic acid (2), caffeic acid (3), ethyl ferulate (4), apigenin (5), hesperetin (6), diosmetin (7), luteolin (8), kaempferol (9), quercetin (10), ecdysterone (11), hesperedin (12), diosmin (13), kaempferol-3-O-rutinoside (14) and rutin (15). The lack of chemical and biological investigations on this plant encouraged us to carry out the above-mentioned work

Hyodo R. and Fujimoto Y. (2000) Biosynthesis of 20-hydroxyecdysone in *Ajuga* hairy roots: the possibility of 7-ene introduction at a late stage. *Phytochemistry* 53, 733-737.

Abstract: Administration of [3 α -2H]-3 β -hydroxy-5 β -cholestan-6-one to hairy roots of *Ajuga reptans* var. *atropurpurea* followed by 2H-NMR spectroscopic analysis of the resulting 20-hydroxyecdysone so formed revealed that the substrate was efficiently incorporated into the latter. Additionally, [5 β ,7 α ,7 β -2H3]-2 β ,3 β -dihydroxy-5 β -cholestan-6-one was converted into 20-hydroxyecdysone. These findings clearly indicate that *Ajuga* hairy roots are capable of introducing a double bond at the 7-position at a late stage of 20-hydroxyecdysone biosynthesis, suggesting the possibility of an alternative biosynthetic pathway which does not involve 7-dehydrocholesterol as an obligatory intermediate.

Hyodo R., Ohyama K. and Fujimoto Y. (2000) Biosynthesis of 20-hydroxyecdysone in *Ajuga* hairy roots: the possibility of 7-ene introduction at a later stage. 2000 International Chemical Congress of Pacific Basin Societies, Honolulu, Hawaii, abstract 376.
Conference Abstract.

Ikan R. and Ravid U. (1970) The isolation and identification of ecdysterone from *Ajuga iva*. *Planta Medica* 20(1), 33-35.

Abstract: Ecdysterone, an insect-molting hormone, has been isolated from *Ajuga iva* and identified by means of chromatographic (column, TLC) and spectroscopic (UV, IR, NMR, MS) methods.

Ikan R. and Ravid U. (1971) The isolation and identification of cyasterone from *Ajuga chia* (Labiatae). *Phytochemistry* 10, 1659-1661.

Ikan R., Ravid U., Trosset D. and Shulman E. (1971) Ecdysterone: an insect moulting hormone from *Acyranthes aspersa* (Amaranthaceae). *Experientia* 27(5), 504-505.

Imai S., Fujioka S., Nakanishi K., Koreeda M. and Kurokawa T. (1967) Extraction of ponasterone A and ecdysterone from Podocarpaceae and related plants. *Steroids* 10(5), 557-565.

Abstract: A general procedure for extracting substances possessing insect moulting hormone activity, i.e., certain polyhydroxy steroids, from plant leaves is described. The screening of plants closely related to *Podocarpus Nakaii* for the ecdysones and ponasterones has led to the isolation of ponasterone A from *P. macrophyllus*, *P. chinensis*, and *Taxus cuspidata*, and the isolation of ecdysterone from *P. macrophyllus* and *Taxus cuspidata*.

Imai S., Fujioka S., Murata E., Sasakawa Y. and Nakanishi K. (1968) The structures of three additional phytoecdysones from *Podocarpus macrophyllus*, makisterone B, C and D. *Tetrahedron Letters* (36), 3887-3890.

Imai S., Hori M., Fujioka S., Murata E., Goto M. and Nakanishi K. (1968b) Isolation of four new phytoecdysones, makisterone A, B, C, D and the structure of makisterone A, a C₂₈ steroid. *Tetrahedron Letters* (36), 3883-3886.
No Abstract.

Imai S., Fujioka S., Murata E. and Otsuka K. (1969a) Structure of the phytoecdysone, ajugasterone B. *Journal of the Chemical Society, Chemical Communications* 82-83.
No Abstract.

Imai S., Murata E., Fujioka S., Koreeda M. and Nakanishi K. (1969b) Structure of ajugasterone C, a phytoecdysone with an 11-hydroxy-group. *Journal of the Chemical Society, Chemical Communications* 546-547.

Abstract: The new phytoecdysone isolated from *Ajuga japonica* Miq. is shown to be 2 β ,3 β ,11 α ,14 α ,20,22-hexahydroxy-5 β -cholest-7-en-6-one.

Imai S., Toyosato T., Sakai M., Sato Y., Fujioka S., Murata E. and Goto M. (1969c) Isolation of cyasterone and ecdysterone from plant materials. *Chemical and Pharmaceutical Bulletin* 17(2), 340-342.

Abstract: The isolations of the following substances are described : cyasterone and ecdysterone from *Ajuga decumbens* THUNB., *A. incisa* MAXIM. and *A. nipponensis* MAKINO, and ecdysterone from *Trillium Smallii* MAXIM., *T. Tschonokii* MAXIM., *Stachyurus praecox* SIEB. et Zucc. and *Polypodium japonicum* MAKINO.

Imai S., Toyosato T., Sakai M., Sato Y., Fujioka S., Murata E. and Goto M. (1969d) Screening results of plants for phytoecdysones. *Chemical and Pharmaceutical Bulletin* 17(2), 335-339.

Abstract: 1056 species of plants and 351 crude drugs were screened by the Chilo dipping test looking for the phytoecdysones. 13 species of Pteridophyta and 23 species of Gymnospermae and Angiospermae were newly found to show the insect-moulting activity. The chemotaxonomical relations were discussed.

Imai S., Murata E., Fujioka S., Matsuoka T., Koreeda M. and Nakanishi K. (1970a) Structures of stachysterone A, the first natural 27-carbon steroid with a rearranged methyl group, and stachysterone B. *Journal of the American Chemical Society* 92(25), 7510-7512.

No Abstract.

Imai S., Murata E., Fujioka S. and Matsuoka T. (1970) Structures of stachysterones C and D. *Journal of the Chemical Society, Chemical Communications* 352-353.

Abstract: Structures (I) and (II) have been derived for stachysterones C and D, respectively.

Ishola I.O., Ochieng C.O., Olayemi S.O., Jimoh M.O. and Lawal S.M. (2014) Potential of novel phytoecdysteroids isolated from *Vitex doniana* in the treatment of depression: involvement of monoaminergic systems. *Pharmacology, Biochemistry and Behavior* 127, 90-100.

Abstract: *Vitex doniana* Sweet (Verbanaceae) is used in traditional African medicine for the treatment of neurological disorders including depression. In our previous studies, three new phytoecdysteroids were isolated from methanol stem bark extract of *V. doniana* (VD) (11 β -hydroxy-20-deoxyshidasterone, 21-hydroxyshidasterone, and 2,3-acetonide-24-hydroxyecdysone) along with known ecdysteroids. This study was designed to investigate antidepressant-like effect of VD and the isolated phytoecdysteroids in behavioral models of despair, forced-swim test (FST) and tail-suspension test (TST) in mice. VD (100 and 200mg/kg, p.o.) treatment reduced ($P < 0.05$) the duration of immobility in both tests without affecting the locomotor activity and exploratory behavior as observed in the open field test. Similarly, 21-hydroxyshidasterone, 11 β -hydroxy-20-deoxyshidasterone, ajugasterone and 24-hydroxyecdysone acute oral treatments significantly reduced immobility time with peak effect at 10mg/kg, which was similar to the effect of conventional antidepressants (imipramine and fluoxetine) in the FST. Conversely, pretreatment of mice with yohimbine (1mg/kg, i.p., α_2 -adrenoceptor antagonist), ketanserin (5mg/kg, i.p., 5-HT_{2A/2C} receptor antagonist) or sulpiride (dopamine D₂ receptor antagonist) prevented the antidepressant-like effect of 21-hydroxyshidasterone while the effects of 11 β -hydroxy-20-deoxyshidasterone and 24-hydroxyecdysone were blocked by yohimbine or ketanserin in the FST. Moreover, the anti-immobility effect elicited by ajugasterone was prevented by prazosin (62.5 μ g/kg, i.p., α_1 -adrenoceptor antagonist) pretreatment. Our findings demonstrated that *V. doniana* and its phytoecdysteroids constituents elicited antidepressant-like effect in behavioral paradigm of despair. Furthermore, 21-hydroxyshidasterone produces its antidepressant-like effect through interaction with α_2 -adrenoceptor, 5-HT_{2A/2C} receptor and dopamine D₂-receptors but 11 β -hydroxy-20-deoxyshidasterone and 24-hydroxyecdysone effects depend on interaction with α_2 -adrenoceptor and 5-HT_{2A/2C} receptors while ajugasterone produces its action through interaction with post-synaptic α_1 -adrenoceptors. Thus, phytoecdysteroids could play a pivotal role in the treatment of major depression.

Issaadi H.M., Tsai Y-C., Chang F-R. And Hunyadi A. (2017) Centrifugal partition chromatography in the isolation of minor ecdysteroids from *Cyanotis arachnoidea*. *Journal of Chromatography B* 1054, 44-49.

Abstract: Phytoecdysteroids are known for their various beneficial bioactivities in mammals including a non-hormonal anabolic and adaptogenic effect. *Cyanotis arachnoidea* extracts are extensively utilized worldwide as ecdysteroid-rich materials for various purposes, e.g. food supplementation, use in agriculture and aquaculture, etc. Preparative chromatography of ecdysteroids requires extensive use of methods of different selectivity, and only a very limited number of papers are available on related application of modern liquid-liquid chromatographic techniques.

In this work, a centrifugal partition chromatography (CPC) method was developed for the isolation of two minor ecdysteroids, dactryhainansterone and calonysterone, from a pre-purified commercial extract of *Cyanotis arachnoidea*. The biphasic solvent system was optimized by HPLC, and was composed of *n*-hexane – ethyl acetate – methanol – water (1:5:1:5, v/v/v/v). The isolated dactryhainansterone and calonysterone represented 99.1% and 99.7% purity, respectively.

Calonysterone exerts a stronger effect on the protein kinase B (Akt) phosphorylation in mammalian skeletal muscle cells than the abundant 20-hydroxyecdysone, while no related data are available on dactryhainansterone. Despite their presence in food supplements, neither compound has appropriately been assessed for safety and efficacy. The reported method allows the gram scale isolation of these compounds, opening ways to their in-depth pharmacological investigation.

Iyer R.T., Ayengar K.N.N. and Rangaswami S. (1973) Occurrence of cheilanthone-B in *Cheilanthes mysurensis*. Indian Journal of Chemistry 11, 1336-1338.

Jadhav A.N., Pawar R.S., Arula B. and Khan I.A. (2007) Ecdysteroid glycosides from *Sida rhombifolia* L. Chemistry & Biodiversity 4, 225-230.

Abstract: Seven ecdysteroids, including the three new compounds 1-3, were isolated from *Sida rhombifolia* L. Their structures and configurations were determined by extensive spectroscopic techniques in combination with chemical derivatization. The four known compounds--ecdysone (4), 20-hydroxyecdysone (5), 2-deoxy-20-hydroxyecdysone-3-O-beta-D-glucopyranoside (6), and 20-hydroxyecdysone-3-O-beta-D-glucopyranoside (7)--are reported for the first time from this plant.

Jaiswal Y., Liang Z., Ho A., Chen H., Williams L. and Zhao Z. (2018) Tissue based metabolite profiling and qualitative comparison of two species of *Achyranthes* roots by use of UHPLC-QTOF MS and laser micro dissection. Journal of Pharmaceutical Analysis 8, 10-19 (doi: 10.1016/j.jpha.2017.06.006).

Abstract: *Achyranthes bidentata* and *Achyranthes aspera* are saponin and steroid rich medicinal plants, used extensively for therapeutic treatments in Traditional Chinese Medicine (TCM) and Ayurveda. *A. bidentata* is reported to be one of the rare and extensively exploited medicinal plant species that face the issue of being endangered. Finding qualitative substitute with identical phyto-constituents contributing to similar composition and pharmacological benefits will help in reducing the burden of exploitation of the natural habitats of such plants. In the present study, a comparative metabolite analysis of the whole drug and specific tissues isolated by laser micro-dissection (LMD) was carried out for both the selected species, by use of ultra-high performance liquid chromatography-quadrupole time-of-flight mass spectrometry (UHPLC-QTOF MS). The results of the study indicate that the cortex and the medullary ray tissues are rich in their content of steroidal and saponin constituents such as (25S)-inokosterone-20,22-acetonide, ginsenoside Ro, bidentatoside II and achyranthoside B. Metabolite profiling of the whole tissues of both the species indicates presence of identical constituents. Thus, it is inferred that *A. bidentata* and *A. aspera* can be used as qualitative substitutes for each other

Janeczko A., Oklestkova J., Tarkowska D. and Drygas B. (2021) Naturally occurring ecdysteroids in *Triticum aestivum* L. and evaluation of fenarimol as a potential inhibitor of their biosynthesis in plants. International Journal of Molecular Sciences 22, article 2855 (doi: 10.3390/ijms22062855).

Abstract: Ecdysteroids (ECs) are steroid hormones originally found in the animal kingdom where they function as insect molting hormones. Interestingly, a relatively high number of these substances can also be formed in plant cells. Moreover, ECs have certain regulatory effects on plant physiology, but their role in plants still requires further study. One of the main aims of the present study was to verify a hypothesis that fenarimol, an inhibitor of the biosynthesis of ECs in the animal kingdom, also affects the content of endogenous ECs in plants using winter wheat *Triticum aestivum* L. as a model plant. The levels of endogenous ECs in winter wheat, including the estimation of their changes during a course of different temperature treatments, have been determined using a sensitive analytical method based on UHPLC-MS/MS. Under our experimental conditions, four substances of EC character were detected in the tissue of interest in amounts ranging from less than 1 to over 200 $\mu\text{g} \cdot \text{g}^{-1}$ FW: 20-hydroxyecdysone, polypodine B, turkesterone, and isovitexirone. Among them, turkesterone was observed to be the most abundant EC and accumulated mainly in the crowns and leaves of wheat. Importantly, the level of ECs was observed to be dependent on the age of the plants, as well as on growth conditions (especially temperature). Fenarimol, an inhibitor of a cytochrome P450 monooxygenase, was shown to significantly decrease the level of naturally occurring ECs in experimental plants, which may indicate its potential use in studies related to the bio-synthesis and physiological function of these substances in plants.

Jayasinghe U.L.B. (1995) Pilot plant scale extraction of ecdysterone from *Diploclisia glaucescens*. ACGC Chemical Research communications 3, 38-39.

Jayasinghe L., Jayasooriya C.P., Oyama K. and Fujimoto Y. (2002) 3-Deoxy-1 β ,20-dihydroxyecdysone from the leaves of *Diploclisia glaucescens*. *Steroids* **67**, 555-558.

Abstract: Chemical investigation of methanol extract of the leaves of *Diploclisia glaucescens* of the family Menispermaceae furnished a new ecdysteroid, 3-deoxy-1 β ,20-dihydroxyecdysone. The structure of the new ecdysteroid was established on detailed analysis of spectral data. The 3-deoxy ecdysteroid showed 40% potency of 20-hydroxyecdysone in the spiracle index assay using the fourth instar larvae of the silkworm *Bombyx mori*.

Jayasinghe L., Kumarihamy B.M.M., Arundathie B.G.S., Dissanayake L., Hara N. and Fujimoto Y. (2003a) A new ecdysteroid, 2-deoxy-5 β ,20-dihydroxyecdysone from the fruits of *Diploclisia glaucescens*. *Steroids* **68**(5), 447-450.

Abstract: Chemical investigation of ethyl acetate extract of the fruits of *Diploclisia glaucescens* of the family Menispermaceae furnished a new ecdysteroid 2-deoxy-5 β ,20-dihydroxyecdysone, together with 20-hydroxyecdysone, 3-deoxy-1 β ,20-dihydroxyecdysone, 2-deoxy-20-hydroxyecdysone, 24-ethyl-20-hydroxyecdysone (makisterone C). Latter two ecdysteroids are reported first time from the family Menispermaceae.

Jayasinghe L., Jayasooriya C.P., Hara N. and Fujimoto Y. (2003b) A pyridine ring-containing ecdysteroid from *Diploclisia glaucescens*. *Tetrahedron Letters* **44**, 8769-8771.

Abstract: A novel pyridine ring-containing ecdysteroid, named diploclidine, was isolated from the methanol extract of the leaves of *Diploclisia glaucescens*, and its structure was determined by spectral means.

Jayasinghe U.L.B., Balasooriya B.A.I.S., Hara N. and Fujimoto Y. (2005) Steroidal and triterpenoidal saponins from the fruits of *Diploclisia glaucescens*. *Natural Product Research* **19**(3), 245-251.

Abstract: Chemical investigation of methanol extract of the fruits of *Diploclisia glaucescens* furnished 3-O- β -D-glucopyranosyl-20-hydroxyecdysone, 3-O-[β -D-glucopyranosyl-(1 \rightarrow 2)- β -D-glucopyranosyl]serjanic acid 28-O- β -D-glucopyranosyl ester and 3-O-[β -D-xylopyranosyl(1 \rightarrow 2)- β -D-glucopyranosyl-(1 \rightarrow 2)- β -D-glucopyranosyl]serjanic acid 28-O- β -D-glucopyranosyl ester. The latter saponin was found to be a new natural product while the other two are reported for the first time from the family Menispermaceae.

Jenett-Siems K., Krause N., Siems K., Jakupovic S., Wallukat G. and Melzig M.F. (2012) Chemical composition and biological activity of *Paris quadrifolia* L. *Zeitschrift für Naturforschung* **67c** 565-570.

Abstract: A study of the components of *Paris quadrifolia* was undertaken to identify compounds with potential influence on cardiac cells, since previous reports suggested a cardiotoxic risk of this plant. Compounds isolated and identified included one new steroidal saponin, (23S,24S)-spirosta-5,25(27)-diene-1 β ,3 β ,21,23,24-pentol-1-O-beta-D-apiofuranosyl-(1 \rightarrow 3)-alpha-L-rhamnopyranosyl-(1 \rightarrow 2)-[beta-D-xylopyranosyl-(1 \rightarrow 3)]-beta-D-glucopyranoside 21-O-beta-D-apiofuranoside 24-O-beta-D-fucopyranoside (1), demonstrating quite unusual structural features, as well as the known compounds 26-O-beta-D-glucopyranosyl-(25R)-5-en-furost-3 β ,17 α ,22 α ,26-tetraol-3-O-alpha-L-rhamnopyranosyl-(1 \rightarrow 4)-alpha-L-rhamnopyranosyl-(1 \rightarrow 4)-[alpha-L-rhamnopyranosyl-(1 \rightarrow 2)]-beta-D-glucopyranoside (2), pennogenin 3-O-alpha-L-rhamnopyranosyl-(1 \rightarrow 4)-alpha-L-rhamnopyranosyl-(1 \rightarrow 4)-[alpha-L-rhamnopyranosyl-(1 \rightarrow 2)]-beta-D-glucopyranoside (3), 7-O-beta-D-glucopyranosyl-kaempferol-3-O-beta-D-glucopyranosyl-(1 \rightarrow 2)-beta-D-galactopyranoside (4), kaempferol-3-O-beta-D-glucopyranosyl-(1 \rightarrow 2)-beta-D-galactopyranoside (5), 5-hydroxyecdysterone (6), and 20-hydroxyecdysone (7). The pennogenin derivative 3 showed strong cardiotoxic effects in an in vitro cellular model system, whereas the respective furostanol derivative 2 was inactive.

Jia A., Li Y., Zhou J. and Gao J. (2010) Three phytoecdysteroids from *Sagina japonica* and potential biotransforming pathways of japonicone. *Chemistry of Natural Compounds* **46**(5) 738-741.

Abstract: In continuation of the series of studies on the chemical components of the whole plant of *Sagina japonica* Ohwi, another two phytoecdysteroids named 22,25-epoxy-24-methylene-2,3,14,20-tetrahydrocholest-7-en-6-one, or japonicone (1) and shidasterone (2), along with the previously reported compound 20-hydroxyecdysone (3), have been isolated on the basis of polyspectroscopic methods (1H NMR, 13C NMR, HMQC, HMBC, MS, and IR). The heteronuclear multiple bond coherence (HMBC) data of shidasterone (2) was supplemented for first time. Potential biotransforming pathways of japonicone (1) were discussed

Jiang P., Liu Y., Sun Y-P., Pan J., Guan W., Xu Z-P. Li X-M., Wang S-Y., Mei Y., Yang B-Y., and Kuang H-X. (2021) Ecdysteroids from the aerial parts of *Paris verticillata*. *Chemistry and Biodiversity* (10.1002/cbdv.202100239).

Abstract: Two new ecdysteroids 14-epi-polypodine B (1) and 22-oxo-hydroxyecdysterone (2), along with nine known compounds, polypodine B (3), viticosterone E (4), 20-hydroxyecdysone-2-acetate (5), 22-oxo-20-hydroxyecdysone (6), 5-hydroxyecdysone (7), pinnatasterone (8), 3-epi-20-hydroxyecdysone (9), ecdysterone (10) and stachysterone B (11), were isolated from the aerial parts of *Paris verticillata*. The structures of all compounds were elucidated by extensive spectroscopic analysis, quantum chemical calculations and ANN-

PRA/DP4⁺ probability analysis. Among them, the absolute configuration of compound **1** and **2** was unambiguous determined by ECD. Also, the isolated compounds were assessed for their cytotoxic activities. Compounds **2**, **3** and **7** exhibited significant cytotoxic activities against PC12, LN299 and SMCC7721 cells.

Jiang X. and Li X. (1997) Phytoecdysones of the aerial parts of the uniflower Swiss centaury (*Rhaponticum uniflorum*). *Zhongcaoyao* (Chinese Traditional and Herbal Drugs) **28**(5), 262-264 [in Chinese, with an English abstract].

Jiang Y-T., Yan W-J., Qi C-L., Hou J-Q., Zhong Y-Y., Li H-J., Wang H. and Li P. (2017) Triterpenoid saponins from the roots of *Cyathula officinalis* and their inhibitory effects on nitric oxide production. *Chinese Journal of Natural Medicines* **15**(6), 463-466.

Abstract: The present study was designed to investigate the chemical constituents of the roots of *Cyathula officinalis*. Compounds were isolated by silica gel, Sephadex LH-20, ODS column chromatography, and preparative HPLC. Their structures were determined on the basis of 1D and 2D NMR techniques, mass spectrometry, and chemical methods. One new oleanane-type triterpenoid saponin, 28-O-[α -L-rhamnopyranosyl-(1 \rightarrow 3)- β -D-glucuronopyranosyl-(1 \rightarrow 3)- β -D-glucopyranosyl] hederagenin (**1**), was isolated from the roots of *Cyathula officinalis*. The anti-inflammatory activities of the isolates were evaluated for their inhibitory effects against LPS-induced nitric oxide (NO) production in RAW 264.7 macrophages cells. Compounds **2**, **4**, and **6** exhibited moderate anti-inflammatory activities.

Jiang Z., Qian J., Dong H., Yang J., Yu X., Chen J., Chen H., Shi Q. and Jia L. (2017) The traditional Chinese medicine *Achyranthes bidentata* and our *de novo* conception of its metastatic chemoprevention: from phytochemistry to pharmacology. *Scientific Reports* **7**: 3888 (doi:10.1038/s41598-017-02054-y) 13 pages.

Abstract: Our recent biosystems analysis revealed similarities between embryonic implantation and cancer cell adhesion, which suggests that abortifacients may be good for safe and effective metastatic chemoprevention targeting circulating tumor cells (CTC). Here we test the hypothesis by using the well-known abortion herb *Achyranthes bidentata* Blume (*A. bidentata*). Five compounds were separated from the herb root. Among them, ginsenoside Ro was the most potent in inhibiting embryonic implantation within non-cytotoxic concentrations. It specifically inhibited the metastatic dissemination capability of colon cancer cells HT29, including the migration and invasion ability, and their adhesion to human endothelium through inhibiting integrin α v β 6, MMP-2, MMP-9, and ERK phosphorylation by HT29. Pretreatment of nude mice with oral ginsenoside Ro followed by HT29 intravenous inoculation and 40-day oral ginsenoside Ro significantly prevented lung metastasis with downregulation of integrin α v β 6 and no toxicity. The present study firstly introduces the new conception of utilizing safe and effective abortion botanic medicines for CTC-based metastatic chemoprevention.

Jimenez-Aspee F., Theoduloz C., Pormetter L., Mettke J., Avila F. and Schmeda-Hirschmann G. (2019) Andean *Prumnopitys andina* (Podocarpaceae) fruit extracts: characterization of secondary metabolites and potential cryoprotective effect. *Molecules* **24**, 4028 (doi: 10.3390/molecules24224028).

Abstract: The fruits from the Chilean Podocarpaceae *Prumnopitys andina* have been consumed since pre-Hispanic times. Little is known about the composition and biological properties of this fruit. The aim of this work was to identify the secondary metabolites of the edible part of *P. andina* fruits and to assess their antioxidant activity by means of chemical and cell-based assays. Methanol extracts from *P. andina* fruits were fractionated on a XAD7 resin and the main compounds were isolated by chromatographic means. Antioxidant activity was determined by means of 2,2-diphenyl-1-picrylhydrazyl radical (DPPH), ferric reducing power (FRAP), trolox equivalent antioxidant capacity (TEAC) and oxygen radical absorbance capacity (ORAC) assays. The cytoprotective activity of the extract against oxidative and dicarbonyl stress was evaluated in human gastric epithelial cells (AGS). The total intracellular antioxidant activity (TAA) of the extract was determined in AGS cells. The inhibition of meat lipoperoxidation was evaluated under simulated gastric digestion conditions. Rutin, caffeic acid β -glucoside and 20-hydroxyecdysone were identified as major components of the fruit extract. Additional compounds were identified by high-performance liquid chromatography diode-array detector mass spectrometry (HPLC-DAD-MSⁿ) and/or co-injection with standards. Extracts showed dose-dependent cytoprotective effects against oxidative and dicarbonyl-induced damage in AGS cells. The TAA increased with the pre-incubation of AGS cells with the extract. This is the first report on the composition and biological activity of this Andean fruit.

Jin H., Hikino H. and Takemoto T. (1975) Seasonal dynamics of the accumulation of phytoecdysones in *Achyranthes fauriei*. *Yakugaku Zasshi* **95**(5), 596-598 [in Japanese, with an English abstract].

Abstract: Dynamics of the accumulation of the phytoecdysones, inokosterone and ecdysterone, in *Achyranthes fauriei* were examined by gas-liquid chromatography and it was found that the content of phytoecdysones dose not vary throughout the year and even during the germinating period.

Jin Y-s., Xu W. and Li Y-s. (2011) Isolation and identification of ecdysterones from root of *Silene viscidula* Franch. Journal of Shenyang Pharmaceutical University (4), 269-271 + 282 [in Chinese].

Abstract: The constituents of the extract from the root of *Silene viscidula* Franch were studied. The chem. constituents were isolated by silica gel column, open ODS column, Sephadex LH-20 column chromatog., and semi-preparative HPLC. Chem. characters and spectroscopic anal. were employed for the structural identification. Five ecdysterone compounds were obtained and their structures were identified as 20-hydroxyecdysone (1), 1-epi-integristerone A (2), abutasterone (3), stachysterone (4) and 15-hydroxystachysterone A (5), along with two sterol compounds daucosterol (6) and β -sitosterol (7). Compounds 2-5 are obtained from the genus *Silene* for the first time.

Jitchum V., Promdang S., Tongtavee N. and Petchsri S. (2016) Screening of some species of Thai microsoroid ferns for phytoecdysteroids. SDU Research Journal 9(3), 81-97 [in Thai, with an English Abstract].

Abstract: Phytochemical screening investigation in the fronds of the seven species of Thai microsoroid ferns (Polypodiaceae) resulted in the isolation of 10 significant phytoecdysteroids (PEs). These fern in the conservation area of northern Thailand were collected and compared with previously recognized taxa. In addition, *M. insigne*, *M. punctatum*, and *P. scolopendria* represent an excellent source of PEs. This is the pioneer work that reports the study on the screening of the PEs from the dried aerial parts of *M. insigne*. Unfortunately, the ecdysteroids in *M. membranaceum* could not be found by using HPLC because of its insolubility in this solvent system. Moreover, the variety of PEs found in Thai microsoroid ferns could not potentially be distinguished from their ferns at the genus level. The voucher specimens of the study species were deposited at the Botanical Laboratory, Science Department, Faculty of Liberal Arts and Sciences, Kasetsart University, Kamphang Saen Campus.

Jizba J., Herout V. and Šorm F. (1967a) Polypodine B - A novel ecdysone-like substance from plant material. Tetrahedron Letters (51), 5139-5143.
No Abstract.

Jizba J., Herout V. and Šorm F. (1967b) Isolation of ecdysterone (crustecdysone) from *Polypodium vulgare* L. rhizomes. Tetrahedron Letters (18), 1689-1691.
No Abstract.

Jizba J., Dolejš L., Herout V., Šorm F., Fehlhäber H.-W., Snatzke G., Tschesche R. and Wulff G. (1971) Polypodosaponin, ein neuer Saponintyp aus *Polypodium vulgare*. Chemische Berichte 104, 837-846.

Abstract: From *Polypodium vulgare* L. a saponin, polypodosaponin (1), was isolated. It is easily converted into 26-0-methyl-polypodosaponin (2) during the isolation with methanol. The structure elucidation was mainly made with 2. The corresponding aglykon (26-0-methylpolypodogenin) has been shown to have the structure of a 3P-hydroxy-26E-methoxy-225.26Eepoxy-6-oxo-5cc-cholest-7-ene (4). In the saponin 2, the 3P-hydroxy group is attached to an α -L-rhamnopyranosyl-(1- \rightarrow 2)- β -D-glucopyranosyl moiety. 2 shows the typical properties of a saponin, and it is representing a new type of steroid saponins..

Jizba J., Vašíčková S. and Herout V. (1974a) Components of the fern *Polypodium aureum* L. Collection of Czechoslovak Chemical Communications 39, 501-505.

Jizba J., Dolejš L. and Herout V. (1974b) Polypodoaurein, a new phytoecdysone from *Polypodium aureum* L. Phytochemistry 13, 1915-1916.

Abstract: A new phytoecdysone was isolated from *Polypodium aureum* L. and its structure determined as 25-O-methylecdysone.

John R., Shajita P.P., Devassy A. et al. (2017) Hairy-root cultures of *Achyranthes aspera* Linn. As a novel route for the production of 20-hydroxyecdysone. Acta Physiologiae Plantarum 39(12), Article 255

Abstract: An efficient transformation system for hairy-root induction in *Achyranthes aspera* L. was established and investigated as a novel method for the production of 20-hydroxyecdysone. Four *Agrobacterium rhizogenes* strains (MTCC 2364, NCIM 5140, A4, and ATCC 15834) were examined for their frequency of transformation in the presence of various concentrations of acetosyringone. Strain ATCC 15834 showed significantly higher transformation efficiency and was capable of inducing hairy roots from different explants (young leaf, hypocotyls, cotyledon, and stem segments) of *A. aspera* in the presence of 100 μ M acetosyringone. The hairy root transformation of the lines (AS001 and AS002) was confirmed by PCR amplification of rol B locus using primers specific for rol B gene. Hairy roots grown in the Murashige and Skoog (MS) liquid medium augmented with 30 g/L sucrose exhibited the highest biomass accumulation and this medium was found to be superior to Gamborg's B5 medium and Whites root culture medium. HPLC and LC-Q-TOF analyses of the hairy-root extract of *A. aspera* revealed the ability of hairy-root lines to synthesise the phytoecdysteroid 20-hydroxyecdysone.

John R., Shajitha P.P., Devassy A. and Mathew L. (2018) Effect of elicitation and precursor feeding on accumulation of 20-hydroxyecdysone in *Achyranthes aspera* Linn. cell suspension cultures. *Physiology and Molecular Biology of Plants* 24(2), 275-284.

Abstract: 20-Hydroxyecdysone is one of the most common ecdysteroids in plants with potential therapeutic applications. In this study, cell suspension cultures of *Achyranthes aspera* were raised in shake flasks to investigate the production of 20-hydroxyecdysone. The quantification and characterization of 20-hydroxyecdysone in the cultures were done by High performance liquid chromatography (HPLC) and Liquid Chromatography-quadrupole time-of-flight mass spectrometry (LC-Q-TOF) analyses. For raising the suspension, calli initiated from in vitro grown leaf explants were cultured in liquid Murashige and Skoog (MS) medium augmented with combinations of 2, 4-dichlorophenoxyacetic acid (1 mg L^{-1}) and α -naphthaleneacetic acid (1 mg L^{-1}). Maximum growth index of the cell suspension was 9.9, which was achieved during 20th day of culture (final phase of exponential growth). At this stage, the biomass accumulated was $1.09 \pm 0.09 \text{ g dry weight (DW)}$ and the 20-hydroxyecdysone concentration was $0.24 \text{ mg g}^{-1} \text{ DW}$. Eliciting the cultures with 0.6 mM Methyl jasmonate for 6 days; enhanced the production of 20-hydroxyecdysone production to $0.35 \text{ mg g}^{-1} \text{ DW}$. By augmenting the cultures with the precursors namely cholesterol (10 mg L^{-1}) and 7-dehydrocholesterol (10 mg L^{-1}), production of 20-hydroxyecdysone was boosted to $0.31 \text{ mg g}^{-1} \text{ DW}$ and $0.28 \text{ mg g}^{-1} \text{ DW}$ respectively.

Joly R.A., Svahn C.M., Bennett R.D. and Heftmann E. (1969) Investigation of intermediate steps in the biosynthesis of ecdysterone from cholesterol in *Podocarpus elata*. *Phytochemistry* 8, 1917-1920.

Abstract: Both 4-14C- and 26-14C-cholesterol were incorporated into ecdysterone by *Podocarpus elata*. This demonstrates that no degradation of the side chain is involved in the biosynthetic pathway. However, three possible intermediates in ecdysterone biosynthesis, 25-hydroxycholesterol-26-14C, cholesterol 5a,6a-epoxide-4-14C, and cholesterol 5b,6b-epoxide-14C were not converted to ecdysterone. The significance of these results in the biosynthesis of ecdysterone is discussed.

Jones C.G. and Firm R.D. (1978) The role of phytoecdysteroids in bracken fern, *Pteridium aquilinum* (L.), as a defence against phytophagous insect attack. *Journal of Chemical Ecology* 4(2), 117-138.

Abstract: Analysis of green bracken fronds collected during July, August, and October, 1975, for phytoecdysteroids showed that these compounds occur in only trace amounts ($0.25\text{--}0.53 \text{ }\mu\text{g/kg}$ fresh weight [FW]). The effect of ecdysteroids on the feeding behavior of seven species of insect showed that four species were deterred at ecdysteroid concentrations at or above 60 mg/kg FW diet; one species of insect at 6 mg/kg or above, and two species which were not affected at the higher concentrations. It was concluded that the levels of phytoecdysteroids in bracken would not deter insects from feeding on the plant. The previously published data relevant to the possible role of phytoecdysteroids as defense compounds are also discussed.

Josephraj Kumar A., Subrahmanyam B. and Devakumar C. (2000) Growth-regulatory activity of silver fern extract on the cotton bollworm, *Helicoverpa armigera* (Hubner). *Insect Science Applications* 20(4), 295-302.

Abstract: Methanolic extracts of stems and roots of silver fern, *Cheilanthes farinosa* Kaulf., (Polypodiaceae: Pteridophyta) incorporated into a semi-synthetic diet significantly extended the larval period, reduced pupal weight and adversely affected pupation of the cotton bollworm, *Helicoverpa armigera* (Hübner) (Noctuidae: Lepidoptera). Early third instars were more susceptible to the treatment than late third instars. A steroidal fraction ($R_f 0.7$) from the methanolic extract significantly reduced mean larval weight but did not affect pupation. A major ecdysteroidal component (R_t 9.233 min.) was purified by reverse phase HPLC of this fraction, along with two minor components. The total yield of ecdysteroids was 564 mg/kg of shade-dried root and stem matter. Pooled HPLC fractions, when injected into final instars, resulted in dose-dependent effects on larval-pupal transformation. Adults emerging from treated larvae failed to lay eggs and were shorter-lived than their untreated counterparts.

Ju Y., Liang H., Du K., Guo Z. and Meng D. (2020) Isolation of triterpenoids and phytosterones from *Achyranthes bidentata* Bl. To treat breast cancer based on network pharmacology. *Natural Product Research* DOI: 10.1080/14786419.2020.1805603

Abstract: Breast cancer (BC) is a cancer with a high incidence and mortality of female, threatening women's physical and mental health. *Achyranthes bidentata* Blume, a traditional Chinese medicine, has been popular in folk due to its effective pharmacology activities including anti-inflammatory and anti-tumour effects. In order to identify the active ingredients from the roots of *A. bidentata* for treating breast cancer, the separation guided by network pharmacology analysis was employed which led to the isolation of 7 triterpenoids and 10 phytosterones. According to the *in vitro* experiments, the isolated compounds showed significant inhibitory activities on MCF-7 cells as well as anti-inflammatory activities by attenuating the production of NO and TNF- α in LPS-induced macrophage cells. In conclusion, this study successfully predicted and confirmed the positive impact of triterpenoids and phytosterones on breast cancer, which provided a foundation for further *in vivo* studies.

Kalász H., Liktör-Busa E., Janicsák G. and Báthori M. (2006) Role of preparative rotation planar chromatography in the isolation of ecdysteroids. *Journal of Liquid Chromatography & Related Techniques* 29, 2095-2109.

Abstract: A reliable isolation procedure is elaborated for the purification of ecdysteroids from *Serratula wolffii*. The procedure is also applicable to other plant sources. The general ecdysteroid isolation procedure was improved by the introduction of preparative rotation planar chromatography (RPC) to the purification process. Effective and simple cleanup and vacuum reversed-phase column chromatographic separation was completed with RPC, or repeated RPC, to obtain pure ecdysteroids (ajugasterone C, dacyrhainansterone, 22-deoxy-integristerone A, 20-hydroxyecdysone, and 2 new ecdysteroids) from the crude extract. This paper discusses the advantages of this method as the final step of ecdysteroid isolation.

Kamble A. (2018) Phytochemical studies on *Achyranthes aspera*. *World Scientific News* 100, 16-34.

Abstract: Phytochemical analysis was carried out on the plant *Achyranthes aspera* which revealed the presence of medicinally important bioactive compounds. The presence of phytochemical compounds in the plant *Achyranthes aspera* was evaluated in root using different solvents such as Petroleum ether, Chloroform, Methanol. In the present study, the preliminary phytochemical screening of leaf and flower extracts of *Achyranthes aspera* showed the presence of phytochemicals such as alkaloids, carbohydrates, flavonoids, proteins, aminoacids, tannins, phenols, steroids, glycosides and saponins

Kapare V., Satdive R., Fulzele D. and Malpathak N. (2016) Tailoring the in vitro production of 20-hydroxyecdysone using PGDs in *Sesuvium portulacastrum*. *Journal of Chemical, Biological and Physical Sciences, Section B: Biological Sciences* 6(4), 1207-1215.

Kapare V., Satdive R., Fulzele D.P. and Malpathak N. (2017) Impact of gamma irradiation induced variation in cell growth and phytoecdysteroid production in *Sesuvium portulacastrum*. *Journal of Plant Growth and Regulation* 36, 919-930.

Abstract: Abiotic stress in the form of gamma irradiation has been shown a potent inducer of oxidative stress in plant cell cultures which produce higher amounts of commercially important secondary metabolites. In the present study, the impact of low doses of gamma irradiation on growth and accumulation of 20-hydroxyecdysone of *Sesuvium portulacastrum* was investigated. Shoot cultures were established on Murashige and Skoog medium supplemented with indole-3-acetic acid (0.5 mg L^{-1}) and N^6 -benzylaminopurine (2.0 mg L^{-1}). Mutations were induced in tissue culture by treating multiple shoots at low doses of gamma irradiation in the range from 5 to 40 Gy. In vitro cell growth and 20-hydroxyecdysone were assessed during M_1T_1 , M_1T_2 , M_1T_3 , and M_1T_4 generations. A gamma radiation dose of 20 Gy was calculated as 50% of the lethal value (LD_{50}). The survival rates of multiple shoot cultures exposed to high doses were gradually reduced in the course of increased order of generations. High-gamma irradiation doses were harmful to growth and 20-hydroxyecdysone production. The accumulation of 20-hydroxyecdysone of $0.139 \text{ mg/plant dry weight}$ was significantly two-fold higher than non-irradiated shoot cultures. The stressed shoots increased 13-fold 20-hydroxyecdysone at 20 Gy during the M_1T_4 generation compared to the yield of the M_1T_1 generation. The ex vitro plants produced $0.321 \text{ mg/plant dry weight}$ of 20-hydroxyecdysone which was remarkably greater than the untreated control. The present study postulated that gamma radiation induced metabolic changes and easy-to-achieve putative mutant comprised with the high amount of 20-hydroxyecdysone.

Kaplanis J.N., Thompson M.J., Robbins W.E. and Bryce B.M. (1967) Insect hormones: alpha ecdysone and 20-hydroxyecdysone in bracken fern. *Science* 157, 1436-1438.

Abstract: The two major molting hormones of insects, alpha ecdysone and 20-hydroxyecdysone, were isolated in crystalline form from dry pinnae of the bracken fern, *Pteridium aquilinum* (L.). Bracken is the first plant found to contain both of the major insect ecdysones, and it is the first known plant source of alpha ecdysone.

Kashchenko N.I. and Olennikov D.N. (2019) New ecdysteroids and flavonoids from *Silene* plants (Caryophyllaceae). XIIIth International Symposium on the Chemistry of Natural Compounds, October 16th-19th, 2019, Shanghai, China, p113 [abstract].

Kayani W.K., Rani R., Ihsan-ul-Haq and Mizra B. (2014) Seasonal and geographical impact on the morphology and 20-hydroxyecdysone content in different tissue types of wild *Ajuga bracteosa* Wall. ex Benth. *Steroids* 87, 12-20.

Abstract: *Ajuga bracteosa* is an endangered medicinal herb which contains several natural products of therapeutic importance like 20-hydroxyecdysone (20-HE). As geography and habitat play a crucial role in the metabolism and morphology of a plant, the present study was aimed at evaluating the impact of phytogeography, season and tissue type on morphology and 20-HE content of *A. bracteosa*. The results revealed large morphological variations in various ecotypes of *A. bracteosa*. However, plants from the same altitude, regardless of their phytogeography, represented similar morphology. Effect of habitat on 20-HE content remained non-significant except for Karot ($1608 \text{ } \mu\text{g/g}$) and Kahuta ($728 \text{ } \mu\text{g/g}$). Effect of tissue types was significant (p value <0.016) for 20-HE content and

followed ascending order: root < stem < leaf < flower, representing the tender aerial tissues' hormonal supremacy. Seasons showed a significant impact (p value <0.001) on 20-HE content with the pattern: winter (1902 $\mu\text{g/g}$) > spring (1071 $\mu\text{g/g}$) > summer (617 $\mu\text{g/g}$). The aerial tissue types contained more 20-HE content in all seasons; especially during winter its amount radically rose in flowers (μ = 2814 $\mu\text{g/g}$). The aerial portion of Karot ecotype harvested in winter offers a valuable source of 20-HE. To confirm the effect of low temperature on 20-HE content, profiling of *A. bracteosa* raised *in vitro* at different temperature regime was carried out. On the basis of these results we hypothesize that chilling cold hampers vegetative growth and triggers stress induced 20-HE accumulation as a defense response.

Kayani W.K., Dilshad E., Ahmed T., Ismail H. And Mirza B. (2016a) Evaluation of *Ajuga bracteosa* for antioxidant, anti-inflammatory, analgesic, antidepressant and anticoagulant activities. *Complementary and Alternative Medicine* **16**, 375 (doi: 10.1186/s12906-016-1363-y).

Abstract:

Background: *Ajuga bracteosa* has been extensively used traditionally for the treatment of a variety of diseases. The aim of the study was to scientifically validate the wide-scale exploitation of *A. bracteosa* in folk medicine various *in vitro* and *in vivo* assays. Moreover, these activities were related to the intrinsic biologically active phytoecdysteroids of *A. bracteosa*.

Methods: Aerial and root parts of *A. bracteosa* were first extracted separately with chloroform (AbCA and AbCR) and the residue was again extracted with methanol (AbMA and AbMR). Total flavonoid and phenolic contents were assayed as quercetin (QE) and gallic acid equivalents (GAE), respectively. These extracts were analyzed for *in vitro* antioxidant assessment including DPPH and H_2O_2 (% inhibition of free radicals), and reducing power and phosphomolybdenum methods (ascorbic acid equivalents AAE mg/g DW). Further, these extracts were assayed *in vivo* in separate groups of Sprague-Dawley rats for carrageenan induced rat paw edema inhibition, hotplate antinociception, forced swim antidepressant and anticoagulation. Dose of each crude extract and standard drug given to rats was 200 mg/Kg- and 10 mg/10 mL/Kg body weight respectively. Plant extracts and standard drugs were administered orally, 60 min prior to the conduction of assays. Moreover, biologically active phytoecdysteroids were screened in *A. bracteosa* with the help of RP-HPLC.

Results: AbMA represented highest values of flavonoids (QE 1.98 % DW) and phenolic contents (GAE 5.94 % DW), significantly scavenged DPPH radicals (IC_{50} 36.9) and reduced ferric ions with 718.4 mg ascorbic acid equivalent/g (AAE). Highest total antioxidant capacity was expressed by AbMR (927 mg AAE) with an IC_{50} value 19.1 $\mu\text{g/mL}$. The extracts which were found potent anti-oxidants, were also good at *in vivo* activities. AbMA significantly reduced edema in all the three hours of treatment (67.9, 70.3 and 74.3 %). AbMA also showed maximum nociceptor suppression in analgesic assay by delaying the time to start licking of paws in rats (57.7 ± 4.9 s). In addition, maximum anti-coagulation was also exhibited by AbMA (89.3 s), while all extracts were found strong antidepressants (≤ 15.66 s immobility time). Screening of biologically active phytoecdysteroids revealed the presence of 20-hydroxyecdysone (20-HE), makisterone (MKA), cyasterone (CYP) and ajujalactone (AJL). Total phytoecdysteroid content found in *A. bracteosa* was 1232.5 $\mu\text{g/g}$ DW and 20-HE was most abundant (1232.5 $\mu\text{g/g}$ DW) as compared to other phytoecdysteroids.

Conclusion: Based on the tested *in vitro* and *in vivo* activities, AbMA was found to be a promising bioactive extract. These activities can be attributed to the intrinsic polyphenols and phytoecdysteroids contents of *A. bracteosa*.

Kayani W.K., Palazon J., Cusido R.M. and Mirza B. (2016) The effect of *rol* genes on phytoecdysteroid biosynthesis in *Ajuga bracteosa* differs between transgenic plants and hairy roots. *Royal Society of Chemistry Advances* **6**, 22700-22708.

Abstract: Phytoecdysteroids are secondary metabolites biosynthesized by plants as a defense strategy against phytophagous insects. These are also used in certain medical preparations to reduce depression and prevent infections. We studied the effect of *rol* genes on phytoecdysteroid biosynthesis in *Ajuga bracteosa* by transformation through *Agrobacterium tumefaciens* strain GV3101 harboring pPCV002-ABC. Transformation was confirmed by PCR. Among seven independently generated transgenic lines, a significant increase of phytoecdysteroids was observed in lines 3 and 4 (6728 and 6759 $\mu\text{g g}^{-1}$ total phytoecdysteroids, respectively) that was 14.5-fold higher than the control plants. Both these lines showed relatively high expression of the *rolC* gene verified by semi-quantitative RT-PCR and densitometric analysis. We also obtained transgenic hairy root lines of *A. bracteosa* by transformation with *A. rhizogenes* strains LBA-9402, A4 and ARqual. Semi-quantitative RT-PCR and densitometric molecular imaging of 9 transgenic root lines obtained from LBA 9402 revealed a high *rolC* expression. Transgenic hairy root lines also exhibited phytoecdysteroid production, particularly lines A4-2 and 9402-01 that showed a yield of 4449 and 4123 $\mu\text{g g}^{-1}$ dry weight, respectively. Complete transgenic plants developed from hairy roots had more phytoecdysteroid content than the parent hairy root lines, suggesting the presence of a possible sink (leaves). Considering the ecdysteroid negative feedback inhibition, we hypothesize that the unavailability of a suitable sink prevents further biosynthesis of phytoecdysteroids in hairy roots. Moreover, sengosterone was not detected in

untransformed plants, pPCV002-ABC-generated transgenic plants or untransformed roots, but its presence in some high ecdysteroid-producing hairy root lines suggests its *de novo* biosynthesis in roots.

Kayani W.K., Palazon J., Cusido R.M. and Mirza B. (2017) Effect of *pRi* T-DNA genes and elicitation on morphology and phytoecdysteroid biosynthesis in *Ajuga bracteosa* hairy roots. RSC Advances 7, 47945-47953.
Abstract: The medicinal plant *Ajuga bracteosa* is a rich source of biologically active and metabolism-enhancing phytoecdysteroids. Transgenic hairy roots from *A. bracteosa* were obtained by infection with *Agrobacterium rhizogenes* strains A4, LBA-9402 and ARqua1. The 59 selectively established and *rolC*-positive hairy root lines increased in size up to 6.6-fold (L42) after one month of *in vitro* culturing and produced a phytoecdysteroid content ranging from 69.3 to 4449 $\mu\text{g g}^{-1}$ (L3 and A2, respectively). The clones of these transgenic roots were maintained for successive subcultures on hormone-free medium to obtain a stabilized morphology. Hairy roots displayed four different morphologies: typical hairy root (THR) (59%), callus-like (CM) (17%), thick (TK) (14%) and thin (TN) (10%). The growth rate of the transgenic hairy root lines varied according to their morphology, with CM showing the highest rate (3.93-fold per month). However, THR exhibited the highest phytoecdysteroid content (1538.5 $\mu\text{g g}^{-1}$). Considering the hairy root morphology in relation to the *pRi* TR-DNA genes, 100% of the hairy root lines harbored the *mas1* and *ags* genes and 90% of CM lines the *aux1* gene. Strikingly, the clones with TK morphology were all positive for the TL- and TR-DNA genes. In contrast, 0% of TN clones harbored the *aux1* and *ags* genes, and only 16.7% the *mas1* gene. All CM and TK hairy roots were positive for the *ags* gene, while *mas1* was present in all hairy root types except TN. As expected, all the root lines considered in this work were positive for the *rol C* gene. Eleven hairy root lines displaying a high growth rate and phytoecdysteroid content were elicited with methyl jasmonate (MeJ) and coronatine (Cor). MeJ doubled the phytoecdysteroid content after 14 days of elicitation (8356 $\mu\text{g g}^{-1}$ in L2) compared with unelicited control hairy roots, and in *in vitro*-grown untransformed roots the increase was 5.6-fold.

Keckeis K., Sarker S.D. and Dinan L. (2000) Phytoecdysteroids from *Atriplex nummularia*. Fitoterapia 71(4), 456-458.

Abstract: A bioassay for ecdysteroid agonists/antagonists and ecdysteroid-specific radioimmunoassays, together with a photo-diode-array detector-monitored high-performance liquid chromatography, have been used to analyse a methanol extract of the seeds of *Atriplex nummularia*. This yielded two phytoecdysteroids, 20-hydroxyecdysone (1) and polypodine B (2).

Khaliq-uz-Zaman S.M., Simin K. and Ahamad V.U. (2000) Chemical constituents from *Asparagus dumosus*. Fitoterapia 71, 331-333.

Abstract: The isolation and characterization of calonysterone (1), blechnoside B (2), 20-hydroxyecdysone and isovanillin from the whole plant of *Asparagus dumosus* are being reported for the first time from this source.

Khanavi M., Najafi B., Sadati S.N., Abai M.R. and Vatandoost H. (2017) Chemical constitute and larvicidal activity of fractions of *Ajuga chamaecistus tomentella* plant against malaria vector *Anopheles stephensi*. Journal of Arthropod-borne Diseases 11(1), 116-123.

Abstract:

Background: The genus *Ajuga*, belongs to Lamiaceae family, is one of the exclusive subspecies in the flora of Iran. The plants of this genus are used traditionally for treatment of joints pain, gout, jaundice, and as insecticide.

Methods: larvicidal activity of methanol 80% extract and partition fractions of hexane, chloroform, and ethyl acetate obtained from aerial parts of *Ajuga chamaecistus* subspecies *tomentella* against malaria vector *An. stephensi* was evaluated. Phytochemical study of active fraction was analyzed using column chromatography and spectroscopy.

Results: According to the results, among different fractions, hexane fraction has the most larvicidal activity with mortality rate of 100% in concentration of 102 ppm and LC₅₀ of 95.66ppm. The structure of compound 1, main phytoecdysteroid compound separated from hexane fraction, was determined to be ajugalide-E.

Conclusion: The results suggested that the hexane fraction of *Ajuga chamaecistus* subsp *tomentella* could be used as a natural and biodegradable insecticide.

Kholodova Y.D. (1981) Chugaev's reaction for the analysis of steroids. Proceedings of the Symposium on the Analysis of Steroids, Eger, Hungary, pp. 519-522.

Kholodova Y.D., Baltaev U., Volovenko V.O., Gorovits M.B. and Abubakirov N.K. (1979) Phytoecdysones of *Serratula xeranthemoides*. Khimiya Prirodnykh Soedinenii (2), 171-174 [in Russian]/Chemistry of Natural Compounds (2), 144-146 [in English].

Kilinc H., Masullo M., Bottone A., Karayildirim T., Alankus O. and Piacente S. (2018) Chemical constituents of *Silene montbretiana*. Natural Product Research (doi.org/10.1080/14786419.2018.1451998).

Abstract: A new steroidal glycoside, 3- O- β -d-glucopyranosyl-3 β ,25-dihydroxy-5 β -cholest-7-en-6-one 25- O- β -d-glucopyranoside (1), together with six known steroidal derivatives (2-7), one cerebroside (8) and one flavonoid (9) were isolated from *Silene montbretiana* Boiss (Caryophyllaceae), a perennial herb growing mainly in the Middle and East Anatolia, Azerbaijan, Iran, and Turkey. Their structures were established by the extensive use of 1D and 2D NMR experiments along with ESI-MS analyses. The cytotoxicity against the cancer A549 (human alveolar basal carcinoma) and Hela (human epitheloid cervix carcinoma) cell lines has been evaluated. None of the tested compounds, in a range of concentrations between 12.5 and 100 μ M, caused a significant reduction of the cell number.

Kim K.I., Ku C-S., Kim M-J., Park Y.J., Ryu H.W., Song H-H., Kim J.H. and Oh S-R. (2015) Phytoecdysones from the roots of *Achyranthes japonica* Nakai and their anti-atopy activity. *Journal of Applied Biological Chemistry* 58(1), 13-19 [Article in Korean, with an Abstract in English].

Abstract: The roots of *Achyranthes japonica* Nakai were extracted with 100% aqueous and concentrated subfraction was separated with ultra-performance liquid chromatography-based activity profiling. Three compounds were isolated from the subfraction 5 through the repeated prep- high performance liquid chromatography column chromatography. According to the results of physico-chemical and spectroscopic data including NMR and MS, the chemical structures of the compounds were determined as ecdysterone (1), 25S-inokosterone (2), and 25R-inokosterone (3). Three phytoecdysones were showed weak inhibitory activity for thymus and activation-regulated chemokine expression levels in tumor necrosis factor (TNF)- α plus IFN- γ induced HaCaT cells, respectively. However, those compounds 1-3 were exhibited the most potent inhibition (80–95% at 200 μ g/mL) against TNF- α expression levels in A23187 plus phorbol-myristate acetate-induced RBL-2H3 cells. As result, 100% aqueous extract of *A. japonica* has an excellent anti-atopy activity. It could be used to a large range of functional anti-atopy cosmetics.

Kim O.T., Manickavasagam M., Kim Y.J., Jin M.R., Kim K.S., Seong N.S. and Hwang B. (2005) Genetic transformation of *Ajuga multiflora* Bunge with *Agrobacterium rhizogenes* and 20-hydroxyecdysone production in hairy roots. *Journal of Plant Biology* 48(2), 258-262.

Abstract: An efficient transformation system for *Ajuga multiflora* Bunge was established by using *Agrobacterium rhizogenes* strain A4. After inoculation with the bacteria, we obtained a number of hairy-root clones from micro-calli of the explant petioles. One fast-growing line showed the highest production of 20-hydroxyecdysone (20-HE). PCR amplification of rooting locus (rol) genes revealed that the left hand-transferred DNA of the root-inducing plasmid was inserted into the genome of our transformed *Ajuga* hairy roots. This integration was further confirmed by DNA-DNA hybridization. The 20-HE content in hairy roots was 10 times higher than that measured in the wild type.

Kiran P., Mammen D. and Mammen D. (2012) Ecdysterone, phospholipids and phenolics in the seeds of *Amaranthus hypochondriacus*. *Journal of Pharmaceutical Research* 1(3), 68-69.

Abstract: The grain amaranth available in India, *Amaranthus hypochondriacus* Linn, is subjected to chemical analysis, wherein the seeds were found to contain 20- β -hydroxyecdysterone, upto 110 μ g per gm, in addition to β -sitosterol, campesterol and stigmasterol. The oil content of the seeds was 5.49% consisting of glycerides of linoleic acid (40%), oleic acid (27%), palmitic acid (27%) and stearic acid (3%). Phospholipids and galactolipids amounted to 1.266%. The total phenolic was 1.4 mg/gm in terms of gallotannins. The flavonoid located was quercetin (in traces) and phenolic acids present were vanillic and syringic acids. The total antioxidant potential of the seeds was estimated to be IC 50 = 43.75.

Kissmer B. and Wichtl M. (1987) Ecdysone aus Wurzeln und Samen von *Helleborus*-Arten [Ecdysone from roots and seeds of *Helleborus* species]. *Archiv der Pharmazie* 320, 541-546 [in German].

Zusammenfassung: Aus den Wurzeln und Samen von *Helleborus odoratus* WALDST. et KIT. und von *Helleborus purpurascens* WALDST. et KIT. konnten vier Ecdysteroide isoliert werden: β -Ecdyson (1), 5- α -Hydroxyecdyson (2), 5- α -Hydroxyecdyson-3- α -D-glucosid (3) und 5- β -Hydroxyecdyson-3- β -D-glucosid (4).

Klein R. (2007) Preliminary research on *Aster tataricus*. High Falls Gardens, <http://highfallsgardens.net/botanicalstudies/papers/index.html>

Knight J.C. and Pettit G.R. (1969) Arizona flora: the sterols of *Peniocereus greggii*. *Phytochemistry* 8, 477-482.

Abstract: Roots of the cactus *Peniocereus greggii* have been found to contain sucrose, peniocerol, desoxyviperidone, viperidone, viperidinone, and β -sitosterol. Two other sterols, which may be artifacts arising from desoxyviperidone, were also isolated.

Kolegova N.A. and Volodin V.V. (1999) Determination of ecdysteroids by reversed-phase high-performance liquid chromatography using eluent systems with electron-donor additives. *Journal of Analytical Chemistry* 54(12), 1139-1141.

Kozhanova A.M., Tuleuov B.I., Kudabayeva P.K., Temirgazyev B.S., Seilkhanov T.M., Seidakhmetova R.B., Salkayeva L.K. and Adekenov S.M. (2020) Synthesis, NMR spectroscopic study of α -, β - and γ -cyclodextrin inclusion complexes of 2-deoxyecdysone and their anti-inflammatory activity. *Macroheterocycles (Russia)* 13(3), 292-297.

Abstract: 2-Deoxyecdysone-3 β ,14 α ,22R,25-tetrahydroxy-5 β (H)-cholest-7-ene-6-one has been isolated from *Silene wolgensis* (Hornem.) Bess. ex. Spreng. of Caryophyllaceae Juss. family for the first time. The complex formation of phytoecdysteroid with α -, β - and γ -cyclodextrins has been studied by NMR spectroscopy. Based on the change in the chemical shifts of protons of substrate and receptors, it has been established that 2-deoxyecdysone interacts with α -, β - and γ -cyclodextrins with formation of supramolecular inclusion complexes of 1:1 stoichiometric ratio with the entry of ring A of the steroid nucleus of the substrate molecule into the internal cavity of the receptor. Anti-inflammatory activity of the complexes has been studied.

Krasnov E.A., Saratikov A.S. and Yakunina G.D. (1976) Inokosterone and ecdysterone in *Rhaponticum carthamoides*. *Khimiya Prirodnykh Soedinenii* (5), 550 [in Russian]/*Chemistry of Natural Products* 494-495 [in English].

No Abstract.

Krasnov E. A., Saratikov A. S. and Yakunina G. D. (1977) Active substances of *Leuzea carthamoides*. *Izvestiya Sibirskogo Otdeleniya Akademii Nauk SSR Seriya Biologicheskikh Nauk* 1, 93-96. [in Russian with English abstract]

Kubo I. (1992) Recent examples of natural products isolation by countercurrent chromatographic methods. *Journal of Liquid Chromatography* 15(15/16), 2843-2855.

Abstract: The efficient isolation of two steroidal glycoalkaloids from the Andean potato "papa negra", three bitter quassinoids from the bark of *Castela tortuosa*, and several phytoecdysteroids from the root barks of *Vitex strickeri* was accomplished by countercurrent chromatographic methods.

Kubo I., Klocke J.A., Ganjian I., Ichikawa N and Matsumoto T. (1983) Efficient isolation of phytoecdysones from *Ajuga* plants by high-performance liquid chromatography and droplet counter-current chromatography. *Journal of Chromatography* 257, 157-161.

No Abstract.

Kubo I., Matsumoto A., Hanke F.J. and Ayafor J.F. (1984) Efficient isolation of a large amount of 20-hydroxyecdysone from *Vitex madiensis* (Verbenaceae) by droplet counter-current chromatography. *Agricultural and Biological Chemistry* 48(6), 1683-1684.

No Abstract.

Kubo I., Matsumoto A., Hanke F.J. and Ayafor J.F. (1985) Analytical droplet counter-current chromatography isolation of 20-hydroxyecdysone from *Vitex thyrsoflora* (Verbenaceae). *Journal of Chromatography* 321, 246-248.

No Abstract.

Kubo I., Asaka Y., Stout M.J. and Nakatsu T. (1990) Structure of a novel phytoecdysteroid, vitexirone, and efficient isolation of phytoecdysteroids from *Vitex fisherii*. *Journal of Chemical Ecology* 16(8), 2581-2588.

Abstract: A novel phytoecdysteroid, vitexirone, has been isolated from a MeOH extract of the root bark of the East African medicinal plant *Vitex fisherii* by recycling high-performance liquid chromatography on a semipreparative scale. In addition, three known phytoecdysteroids, 20-hydroxyecdysone, ajugasterone C, and turkesterone, also were isolated. The structure of vitexirone has been established spectroscopically. The position and stereochemistry of the 11- α -hydroxy group of ajugasterone C and vitexirone were confirmed by the ^1H - ^1H homonuclear COSY NMR data. These phytoecdysteroids disrupt the molting process of the pink bollworm *Pectinophora gossypiella*.

Kubo I. and Nakatsu T. (1991) Recent examples of preparative-scale recycling high performance liquid chromatography in natural products chemistry. *LC-GC International* 4(7), 37-42.

Kumar A. (2014) Structure elucidation of makisterone A from methanolic extract of *Tinospora cordifolia* and used as antidepressant agent. *Journal of Chemical and Pharmaceutical Research* 6(11), 744-749.

Abstract: *Tinospora cordifolia* is an annual or perennial Ayurvedic plant which is used in several traditional medicines to cure various diseases. Common names are Amrita, Guduchi, Shindilakodi (Tamil) and Gulancha. Makisterone-A is extracted from the stem of *Tinospora cordifolia* by Soxhlet by repeated washing with predistilled methanol (organic solvent) under reflux. TLC and Column Chromatography techniques are used to isolate and purify Makisterone-A and its structure elucidated by Infra-red and NMR spectroscopy techniques. Makisterone-A was found to be very effective in reducing the depressant potential.

Kumpun S., Maria A., Crouzet S., Evrard-Todeschi N., Girault J.-P. and Lafont R. (2011) Ecdysteroids from *Chenopodium quinoa* Willd., an ancient Andean crop of high nutritional value. *Food Chemistry* **125** 1226-1234.

Abstract: *Chenopodium quinoa* Willd. (=quinoa) is a crop cultivated since ancient times by the Incas, which has a high nutritional value. Quinoa belongs to Chenopodiaceae, a family containing many ecdysteroid-containing species, including spinach.

Quinoa seeds have been investigated for their ecdysteroid content. Besides large amounts of 20-hydroxyecdysone, they contain lower amounts of makisterone A, 24-*epi*-makisterone A, 24(28)-dehydromakisterone A and polypodine B, together with a wide array of minor ecdysteroids, among which three were isolated as new natural compounds, i.e. 24,25-dehydroinokosterone, 25,27-dehydroinokosterone and 5 β -hydroxy-24(28)-dehydromakisterone A. Ecdysteroids are concentrated in the bran, and their amount varies with the quinoa variety used. Most of the 20-hydroxyecdysone is recovered undegraded within the seeds even after 20 min boiling, and is thus susceptible to evoke significant (beneficial) pharmacological effects on humans who eat quinoa regularly. The same applies to most quinoa-based foods, which retain most of their original ecdysteroid content.

Kunert O., Haslinger E., Schmid M.G., Reiner J., Bucar F., Mulatu E., Abebe D. and Debella A. (2000) Three saponins, a steroid, and a flavanol glycoside from *Achyranthes aspera* [sic]. *Monatshefte für Chemie* **131**(2), 195-204.

Abstract: Three bisdesmosidic saponins, 20-hydroxyecdysone, and quercetin-3-O- β -D-galactoside were isolated from the methanol extract of the aerial parts of *Achyranthes aspera* L. (*Amaranthaceae*). Their structures were established on the basis of NMR spectroscopic analysis; the complete ^1H and ^{13}C assignments of the compounds were achieved by means of 2D NMR studies.

Kusamba C., Nicoletti M., Federici E., Multari G., Galeffi C. and Palazzino G. (1995) Isolation of ecdysteroids from three species of *Palisota*. *Fitoterapia* **LXVI**(2), 175-178.

Kusano G., Takemoto T., Beisler J.A. and Sato Y. (1975) Steroidal constituents of *Solanum xanthocarpum*. *Phytochemistry* **14**, 529-532 [this paper concerns ecdysteroid-related compounds].

Abstract: From the extract of the fruits of *Solanum xanthocarpum*, cycloartanol (I), cycloartenol (II), sitosterol (III), stigmasterol (IV), campesterol (V), cholesterol (VI), sitosteryl glucoside (VII), stigmasteryl glucoside (VIII), solamargine (IX), and β -solamargine (X) were identified and an isolated steroid (XI) was identical with 4 α -methyl-(24R)-ethylcholest-7-en-3 β -ol synthesized from campesterol.

Laddha K.S. and Ghosh D. (2005) Isolation of 20-hydroxyecdysone from Indian medicinal plant *Achyranthes aspera* and development of simple HPLC analysis. *Natural Products* **1**(1/2) 1-4.

Abstract: Develop a simple procedure for extraction and isolation of 20-hydroxyecdysone from *Achyranthes aspera* and characterized by DSC, UV, IR, CD, ^1H and ^{13}C NMR, MS and quantified by HPLC

Lafont R., Porter C.J., Williams E., Read H., Morgan E.D. and Wilson I.D. (1993) The application of off-line HPTLC-MS-MS to the identification of ecdysteroids in plant and arthropod samples. *Journal of Planar Chromatography* **6**, 421-424.

Lakshmi T., Rajendran R., Ezhilarasan D. and Silvester A. (2017) High-performance liquid chromatography and tandem mass spectrometric analysis of beta ecdysone from *Achyranthes aspera* extract: an antimalarial drug. *Journal of Advanced Pharmacy Education & Research* **7**(2), 61-65.

Abstract: The objective of the paper is to isolate beta ecdysone, an insect molting hormone in *Achyranthes aspera* seed extract by high-performance liquid chromatography (HPLC) and tandem mass spectrometry analysis. Refined extract of *A. aspera* is obtained from Green chem herbal extracts and formulations, Bengaluru. Beta ecdysone is purchased from Sigma. HPLC analysis is performed by Shimadzu LC-20AD prominence gradient system and mass spectra is detected by Shimadzu LC-MS 8030 triple quadrupole mass analyzer. The result reveals that content of beta ecdysone in *A. aspera* extract was 0.05% w/w. Spectra generated for beta ecdysone standard in positive selective ion monitoring gave protonated molecule $[\text{M}+\text{H}]^+$ 481 and in negative selective ion monitoring gave deprotonated molecule $[\text{M}-\text{H}]^-$ 479. Mass value (m/z) of beta ecdysone is 480.65. Same mass of $[\text{M}+\text{H}]^+$ 481 and $[\text{M}-\text{H}]^-$ 479 in spectra generated for *A. aspera* extract eluted at retention time 12.5 min, confirms the presence of beta ecdysone.

Beta ecdysone is found to be richly present in *A. aspera* seed extract further in vivo studies should be done to prove its anti-malarial activity to combat the vector borne diseases.

Laosooksathit S., Preecha P. and Suksamrarn A. (2003) Ecdysteroids as insect control agents: a new ecdysteroid from stem bark of *Vitex canescens*. The Journal of KMITNB 13(1), 1-13.

Abstract: A new ecdysteroid, P5, has been isolated from the stem bark of aged trees (with flower) of vitex canescens. 20-Hydroxyecdysone, makisterone A, 24-epi-makisterone A, and canescensterone have also been isolated from both young trees (without flower) and aged trees (with flower) of vitex canescens. The methods of extraction of the ecdysteroids from vitex canescens are given and the melting points, infrared absorption spectra, nuclear magnetic resonance spectra and mass spectra of the ecdysteroids are reported.

Large T., Lafont R., Morgan E.D. and Wilson I.D. (1992) Micellar capillary electrophoresis of ecdysteroids. Analytical Proceedings 29, 386-388.

Larguet H. (2011) Les molécules térpeniques (ecdystéroïdes) des fleurs de *Serratula cichoracea*: recherche d'activités antimicrobienne et antioxydante [Terpenoid molecules (ecdysteroids) of the flowers of *Serratula cichoracea*: research on antimicrobial and antioxidant activities]. Ph.D. Thesis, Université Mentouri Constantine, Algeria, pp. 75

Lei X.Y., Xia J., Wang J.W. and Zhang L.P. (2018) Comparative transcriptome analysis identified genes putatively involved in 20-hydroxyecdysone biosynthesis in *Cyanothis arachnoidea*. International Journal of Molecular Sciences 19, 1885 pp15 (doi: 10.3390/ijms19071885).

Abstract: *Cyanothis arachnoidea* contains a rich array of phytoecdysteroids, including 20-hydroxyecdysone (20E), which displays important agrochemical, medicinal, and pharmacological effects. To date, the biosynthetic pathway of 20E, especially the downstream pathway, remains largely unknown. To identify candidate genes involved in 20E biosynthesis, the comparative transcriptome of *C. arachnoidea* leaf and root was constructed. In total, 86.5 million clean reads were obtained and assembled into 79,835 unigenes, of which 39,425 unigenes were successfully annotated. The expression levels of 2427 unigenes were up-regulated in roots with a higher accumulation of 20E. Further assignments with Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways identified 49 unigenes referring to the phytoecdysteroid backbone biosynthesis (including 15 mevalonate pathway genes, 15 non-mevalonate pathway genes, and 19 genes for the biosynthesis from farnesyl pyrophosphate to cholesterol). Moreover, higher expression levels of mevalonate pathway genes in roots of *C. arachnoidea* were confirmed by real-time quantitative PCR. Twenty unigenes encoding CYP450s were identified to be new candidate genes for the bioreaction from cholesterol to 20E. In addition, 90 transcription factors highly expressed in the roots and 15,315 unigenes containing 19,158 simple sequence repeats (SSRs) were identified. The transcriptome data of our study provides a valuable resource for the understanding of 20E biosynthesis in *C. arachnoidea*.

Lev S.V., Zakerova R.P., Saatov Z., Gorovits M.B. and Abubakirov N.K. (1990) Ecdysteroids of tissue and cell cultures of *Ajuga turkestanica*. Khimiya Prirodnikh Soedinenii (1), 51-52 [in Russian].

Abstract: The possibility has been studied of obtaining ecdysteroids — ecdysterone and turkesterone — with the aid of a culture of the tissues and cells of the plant *Ajuga turkestanica*. Conditions have been selected under which the yield of ecdysteroids in the culture of tissues and cells is comparable with amounts of the same substances in the organs of the whole plant.

Li F., Wu H., Sun L-l., Wu H., Wang R., Li S-p. Wang W-y., Dai L., Zhang Z-r., Fu J. and Deng R. (2018) Quantitative analysis of multi-components by single marker and fingerprint analysis of *Achyranthes bidentata* Blume. Journal of Chromatographic Science 56(7), 595-603 (doi: 10.1093/chromsci/BMY031).

Abstract: A simple and effective method of high performance liquid chromatography (HPLC) with diode array detection was established to identify the origin of *Achyranthes bidentata* Blume and evaluate its quality, based on chromatographic fingerprint combined with the similarity analysis, hierarchical cluster analysis and the quantitative analysis of multi-components by single marker (QAMS). In the chromatographic fingerprint, 16 peaks were selected as the common model to evaluate the similarities among 18 batches (S₁–S₁₈) of *A. bidentata* Blume samples collected from different origins in China. The similarities values for 18 batches of samples were more than 0.75, which compared with control fingerprint. Furthermore, 18 batches of *A. bidentata* Blume samples were categorized into two groups for quantitative analysis, the quantification of three bioactive constituents (β -ecdysterone, cyasterone and 5-hydroxymethyl furfural) between QAMS and external standard method proved the consistency of the two methods, the three constituents showed good regression ($R > 0.9995$) within linear ranges, and their recoveries were within the range of 97.6–101.5%. This study demonstrated that the quality of *A. bidentata* Blume can be successfully evaluated by means of a combination of HPLC chromatographic fingerprint and QAMS approach.

Li H-b., Hu J., Chen J-c. and Qiu M-h. (2005) Chemical constituents of *Tinospora craveniana*. Natural Product Research and Development (2), 125-127 [in Chinese].

Abstract: 7 compounds were isolated from the roots of the *Tinospora craveniana* S.Y.Hu. The chemical structure of these compounds was determined by the spectral analysis to be: columbin (1), isocolumbin (isocolumbin, 2), Bamatin (palmatine, 3), β -glutasterol (β -sitosterol, 4), 20 beta-Hydroxyl moulting hormone (20-hydroxyecdysone, 5), Bama Tubin (palmatrubine, 6), 20 beta-hydroxymoulting hormone-2-O- β -glucoside (20-hydroxyecdysone 2-O- β -D-glucopyranoside, 7).

Li J., Li H-J. and Qi H. (2007) Simultaneous qualitation and quantification of four phytoecdysones in Radix *Achyranthis Bidentatae* by high-performance liquid chromatography with diode array detection. Biomedical Chromatography 21, 823-828.

Abstract: A high-performance liquid chromatographic method with diode array detection was developed and validated to simultaneously identify and quantify four phytoecdysones, i.e., polypodine B (1), ecdysterone (2), 25-R inokosterone (3) and 25-S inokosterone (4), in Radix *Achyranthis Bidentatae*. The analysis was performed using a C(18) column with 6% tetrahydrofuran aqueous and acetonitrile isocratic elution, and the detection was carried out by DAD at 242 nm. The identities of the analytes were determined by comparing the retention time and UV spectrum with those of reference compounds. The validation of the method included linearity, sensitivity, recovery and stability. All calibration curves of the four phytoecdysones showed good linear regression ($r^2 \geq 0.9993$). The limit of detection (S/N = 3) and limit of quantification (S/N = 10) were less than 7.5 and 12.3 ng, respectively. The method provided desirable repeatability with overall intra- and inter-day variations of less than 4.67%. The obtained recoveries varied between 95.1 and 104.4% while the relative standard deviations were below 4.85% (n = 3). The analysis results indicated that the contents of the investigated phytoecdysones in Radix *Achyranthis Bidentatae* from different locations were highly variant, and the genuine crude drug indigenous to Henan province did not possess the highest concentration of phytoecdysones.

Li J., Qi H., Qi L-W., Yi L. and Li P. (2007) Simultaneous determination of main phytoecdysones and triterpenoids in Radix *Achyranthes Bidentatae* by high-performance liquid chromatography with diode array-evaporative light scattering detectors and mass spectrometry. Analytica Chimica Acta 596, 264-272.

Abstract: A liquid chromatographic method was developed for simultaneous determination of two main types of bioactive compounds: four phytoecdysones and eight triterpenoids in radix *achyranthis bidentatae* (RAB), i.e., polypodine B (1), ecdysterone (2), 25-R inokosterone (3), 25-S inokosterone (4), ginsenoside Ro (5), chikusetsusaponin IVa (6), zingibroside R1 (7), chikusetsusaponin IVa ethyl ester (8), 28-deglucosyl-chikusetsusaponin IVa (9), PJS-1 (10), 28-deglucosyl-chikusetsusaponin IVa butyl ester (11), and oleanolic acid (12). Optimum separations were obtained with a Zorbax C18 column, using a gradient elution with 0.08% aqueous formic acid (containing 5% isopropyl alcohol) and acetonitrile as mobile phase. Phytoecdysones were detected by diode array detector (DAD) at 242 nm, whereas triterpenoids were monitored by evaporative light scattering detector (ELSD) connected in series with DAD, temperature for the drift tube was 110 degrees C and the nitrogen flow rate was 3.2 L min⁻¹. The identity of the analytes was confirmed using retention times, ultraviolet absorbance and mass spectral data in comparison with reference compounds. The method was validated for acceptable precision (intra- and inter-day variation $\leq 4.87\%$), accuracy (recovery $\geq 88.9\%$) and sensitivity (LOD ≤ 0.43 microg mL⁻¹ (DAD) and 26.0 microg mL⁻¹ (ELSD), LOQ ≤ 0.97 microg mL⁻¹ (DAD) and 46.5 microg mL⁻¹ (ELSD), respectively). This rapid and reliable method was applied for the analysis of four cultivated and ten commercial samples. The results demonstrated that the method is suitable for routine analysis and quality control of RAB.

Li J-t., Teng H-m. and Hu Z-h. (2007) Accumulation dynamic of ecdysterone in vegetative organs of *Achyranthese bidentata*. Chinese Traditional and Herbal Drugs 38, 1570-1573 [in Chinese].

Abstract: Objective: To reveal the accumulation of moulting steroids in different nutrient organs in ox-knee plants. Methods: Efficient liquid chromatography. Results: The amount of moulting steroids in the root, stem and leaves of ox-knee was significantly different. The amount of moulting steroids in young organs is high, and with the development and maturity of nutritional organs, the amount of moulting steroids varies significantly with the change of physical waiting period. By the harvest period (November), the amount of moulting steroids in the nutritional organs of the ox-knee was: > root > stem. Conclusion: The peak of moulting steroids in the root of the ox-knee is consistent with its peak yield, and when the ground partially withers in early November, it should be the appropriate harvest period for the ox-knee, which coincides with the traditional ox-knee harvest period. However, at this time the leaves and stems also contain a higher amount of moulting steroids, so it is recommended to carry out comprehensive development and utilization of the above-ground part of the ox-knee.

Li J-ti., Zhang Y-h., Guo X-s., Wang C., Li K. and Hu Z-h. (2014) Effect of IBA on growth of *Achyranthes bidentata* and the accumulation of major medicinal components. Northern Horticulture (20), 153-155 [in Chinese].

Abstract: Taking seedlings of *Achyranthes bidentata* as material, the effect of indole butyric acid (IBA) with different concentrations on the growth of *Achyranthes bidentata* and content of oleanolic acid and ecdysterone were studied. The results showed that 1.0mg/L IBA treatment significantly increased plant height, root length, root dry weight, and improved the accumulation of oleanolic acid and ecdysterone in the root of *Achyranthes bidentata*. The influence of IBA on content of oleanolic acid was not significant, but IBA significantly inhibited the ecdysterone in leaves.

Li J-T., Qi W-Z., Guo X-S., Wang C., Zhang Y-H., Wang D. and Su H-H. (2015) Effects of methyl jasmonate on growth and major medicinal components accumulation of *Achyranthes bidentata*. *Guihaia* (6), 875-879 and 929 [in Chinese].

Abstract: To study the effects of methyl jasmonate on the growth and the major medicinal components (oleanolic acid and ecdysterone) accumulation of *Achyranthes bidentata*. Seeds of *A. bidentata* were soaked for 3 h in different concentrations (0,0.1,0.15,0.2,0.25 mmol.L⁻¹) of methyl jasmonate, and the control groups were soaked in the same volume of distilled water in this study. The determination of plant height, root length, fresh weight of aboveground and root of *A. bidentata* were done after 60 d. The contents of ecdysterone and oleanolic acid in roots and leaves of *A. bidentata* were determined by HPLC. The results showed that the 0.15 mmol.L⁻¹ MeJA treatment sample was beneficial for plant height, root length, fresh weight of aboveground and root. Compared with control, the biomass respectively be increased (43.9%,38.7%,26.4%,64.0%, P <0.05) observably. The 0.15 mmol.L⁻¹MeJA treatment sample was also beneficial for oleanolic acid accumulation in roots and leaves of *A. bidentata*. It had the highest amount respectively (114.3% and 60%,P <0.05). The 0.25 mmol.L⁻¹ MeJA treatment sample was significantly promoted ecdysterone accumulation in roots, but inhibited oleanolic acid accumulation in roots and leaves and ecdysterone in leaves of *A. bidentata*. These results suggested that 0.15 mmol.L⁻¹ MeJA soaking was beneficial for the growth, oleanolic acid accumulation in roots and leaves, and would significantly promote ecdysterone accumulation in roots of *A. bidentata*, and be helpful to improve the quality and yield of medicinal materials.

Li J., Jadhav A.N. and Khan I.A. (2009) Triterpenoids from Brazilian ginseng, *Pfaffia paniculata*. *Planta Medica* (doi: 10.1055/s-0029-1240631).

Abstract: Two new nortriterpenoids, pfaffine A and B (1- 2), were isolated from the roots of *Pfaffia paniculata* Kuntze, along with ten known compounds including four ecdysteroids, ecdysone (3), 20-hydroxyecdysone (4), pterosterone (5), rapisterone (6), five triterpenoids, pfaffic acid (7), pfameric acid (8), mesembryanthemoidigenic acid (9), calenduloside E 6'-methyl ester (10), oleanolic acid 28-O-beta-D-glucopyranoside (11), and one monoterpene glycoside (+)-angelicoidenol-2-O-beta-D-glucopyranoside (12). The structures of the new compounds were elucidated as 3 beta,16 beta-dihydroxy-30-norolean-12,20(29)-dien-28-oic acid (1), and 3 beta-hydroxy-30-norolean-12,20(29)-dien-28-oic acid-28-O-beta-D-glucoside (2) through the extensive analysis of 1D- (1H, 13C, DEPT) and 2D-NMR (COSY, HSQC, HMBC, NOESY) spectra, as well as by a chemical method.

Li J., Huang J., Li P. and Zhang Z. (2010) Influence of crop rotation on contents of sterols and triterpenes of *Achyranthes bidentata*. *Shizhen Guoyi Guoyao* 21(10), 2433-2434 | [in Chinese].

Abstract: The influence of crop rotation on contents of sterols and triterpenes of *Achyranthes bidentata* was studied. HPLC/DAD/ELSD was used to determine contents of polypodine, β-ecdysone, 25-R inokosterone, 25-S inokosterone, ginsenoside Ro, chikusetsusaponin IVa, zingibroside R1 and 28-deglucose chikusetsusaponin IVa. The linear ranges of polypodine, β-ecdysone, 25-R inokosterone, 25-S inokosterone, ginsenoside Ro, chikusetsusaponin IVa, zingibroside R1 and 28-deglucose chikusetsusaponin IVa were 1.57-157.00 μg/mL, 4.45-445.00 μg/mL, 1.12-112.00 μg/mL, 1.46-146.00 μg/mL, 51.40-1028.00 μg/mL, 52.00-1040.00 μg/mL, 15.90-318.00 μg/mL and 14.90-298.00 μg/mL, resp. The average recoveries were 97.9%, 96.2%, 97.9%, 96.9%, 88.9%, 98.3%, 97.0% and 97.2% with RSD of 4.46%, 1.33%, 4.10%, 3.03%, 3.51%, 3.66%, 3.84% and 4.47%, resp. The results showed that crop rotation could promote the contents of sterols and triterpenes of *Achyranthes bidentata*.

Li J., Qi W., Han X., Wang C., Liu X., Liu Y. and Ma Y. (2016) Effects of plant growth substance on growth and major medicinal components accumulation of *Achyranthes bidentata* BI. *Journal of Henan Normal University (Natural Science Edition)* (5), 101-105 [in Chinese].

Abstract: The effect of plant growth substance on the growth and major medicinal components of *Achyranthes bidentata* BI. was investigated by spraying cultured seedlings with different combinations of exogenous hormones and phytochemical method. The results showed that 1.0mg·L⁻¹ IBA+1.0mg·L⁻¹ MJ(T4) coordination treatment significantly promoted underground biomass increasing in *A. bidentata*. 1.0mg·L⁻¹ IBA+1.0mg·L⁻¹ 6-BA(T3)coordination treatment was benefit for oleanolic acid accumulation in root, whereas 1.0mg·L⁻¹ IBA+2.0mg·L⁻¹ NAA(T2)and 1.0mg·L⁻¹ IBA+1.0mg·L⁻¹ 6-BA(T3)treatment also significantly promoted ecdysterone accumulation in roots. Taking into account of each index in this experiment,1.0mg·L⁻¹ IBA+1.0mg·L⁻¹

6-BA(T3) treatment was advantageous to growth, the oleanolic acid and ecdysterone total contents increasing in root of *A. bidentata* and are beneficial to the improvement the output and quality of *A bidentata* crude drug.

Li S.-H., Zhang H.-J., Yao P., Niu X.-M., Xiang W. and Sun H.-D. (2002) Non-taxane compounds from the bark of *Taxus yunnanensis*. *Journal of Asian Natural Products Research* 4(2), 147-154.

Abstract: From the bark of *Taxus yunnanensis*, 15 non-taxane compounds were isolated. Through spectroscopic methods such as ID and 2D NMR and MS experiments, one of them was determined as a new abietane-type diterpenoid named taxayunnin (1). The other 14 known compounds were identified as taxamairin C (2), taxamairin A (3), 3beta-hydroxy-sandaracopimaric acid (4), (+)-3-hydroxy-isodrimenin (5), rubrosterone (6), ponasterone A (7), ecdysterone (8), 20-hydroxy-echysone-20,22-monoacetone (9), 7-oxositosterol (10), stigmast-4-en-6beta-ol-3-one (11), 5alpha,6beta-dihydroxy-daucosterol (12), beta-sitosterol (13), daucosterol (14), 1-O-beta-D-glucopyranosyl-(2S, 3R, 4E, 8Z)-2-N-(2'-hydroxypalmitoyl)-octadeca-sphinga-4,8-dienine (15), respectively. Compounds 4-6, 9-12 and 15 were isolated from *Taxus* plants for the first time.

Li W.F., Song Q.S., Xiang W. and Yang S.M. (2006) Study on the chemical constituents of *Sida szechuensis* Matsuda. *Zhongcaoyao (Chinese Traditional and Herbal Drugs)* 37(9), 1304-1306 [in Chinese].

Abstract: Badusan is the herb of malvaceae *sida rhombifolia* genera plant *sida szechuensis* matsuda. As a local used variety of common Chinese herbal cowheeb in Yunnan, it is also called xiaobadu, mimazhuang and xiaonianyao. According to the record in materia medica of Diannan, cowheeb, also called badusan, cold in property, bitter in flavour, can be used to cure milk obstruction, mammary carbuncle and red swelling for women, to eliminate swollen tumefaction poison, to cure hematuria and blood strangury for children and to eliminate skin pruritus, wind and heat. In the folk of China, badusan is used to cure acute mammitis, acute adenopharyngitis, enteritis, bacterial dysentery, women menoschesis, furuncle, tumefaction and traumatic injuries. The chem. components of Badusan were researched. In order to explore its effective components, the upper ground part of badusan was studied on chem. component. The seven compounds separated are resp. identified to be α -ecdysone (I), β -ecdysone (II), polypodine B (III), pterosterone (IV) β -sitosterol (V), astragalol 6''-O- (4-hydroxycinnamate) (VI) and 1,2,3,4-tetrahydro-1-methyl- β -carboline-3-carboxylic acid (VII). The compound (IV), (VI) and (VII) were obtained from this plant for the first time.

Li X., Qian J., Li X., Yan S. and Chi D. (2013) A study on ecdysterone production of *Ajuga lobata* D. Don cell lines in suspension culture. *Chinese Agricultural Science Bulletin* 29, 127-133.

Abstract: In order to acquire the suspension cultivation production that cells growth quickly, biomass liveweight larger and edysterone content higher, the inducing condition for obtaining the granular embryonic callus was studied by chosen culture medium and screened the phytohormone, and the *Ajuga lobata* D. Don cell suspension culture system was established by optimizing from three aspects, which were the culture medium type, plant hormone species and their proportion, amount of inoculation in this paper. The results show that: MS solid medium added 0.4 mg/L 2,4-D could induce the *Ajuga lobata* D. Don's root segments to form unconsolidated granular embryogenic callus. Comparing the growth status of cell lines in 3 kinds of liquid culture medium such as WPM, MS, 1/2 MS found that cells were grew best in MS. 2,4-D affected cell growth obviously. The cell biomass was 1.3 times higher than which cultured in culture medium added NAA. Experiments on the effect of phytohormone ratio on the *Ajuga lobata* D. Don cell suspension culture shown that, with 6-BA concentration increasing, ecdysterone content increased. Compared with that in control group, edysterone content in the cells cultured in medium added 0.5 mg/L 6-BA was significantly higher ($P < 0.05$). The cells cultured in liquid medium with added 1 mg/L 6-BA accumulated very significantly higher edysterone than that accumulated in the cells in control group ($P < 0.01$). Though, the most prosper phytohormone proportion was 0.4 mg/L 2,4-D + 0.5 mg/L 6-BA, under which the net cell biomass could reach (4.3652 ± 1.0739) g after cultured 7 days, the ecdysterone content could reach (4.5692 ± 0.2044) mg/g of cell dry weight, the cell lines were more uniform and grew faster. When the inoculation amount was 15%, cell lines grew best at the 7th day after inoculation, cell growth rate was 1.109 g/d, cell regeneration index could reach 3.45. It was the most proper inoculation quantity. The optimal cell suspension culture system was established, which would benefit the ecdysterone production.

Li X.Q., Wang J.H., Wang S.X. and Li X. (1998) New phytoecdysterones from *Rhaponticum uniflorum*. *Journal of Chinese Medicinal Chemistry* 8(3), 199-201 [in Chinese, with English abstract].

Li X.Q., Wang J.H., Wang S.X. and Li X. (2000a) A new phytoecdysone from the roots of *Rhaponticum uniflorum*. *Journal of Asian Natural Products Research* 2(3), 225-229.

Abstract: A new ecdysteroid named rhapontisterone R₁ (1) together with two known phytoecdysones, rhapontisterone (2) and ecdysterone (3) were isolated from the roots of *Rhaponticum uniflorum* (L.) DC. The new compound was shown to be 2 β ,3 β ,11 α ,14 α ,20 ξ ,22 ξ -hexahydroxy-stigma-7,24(28)-dien-6-oxo-28,25-carbolactone. The structure has been determined primarily on the basis of physico-chemical properties and spectral analysis.

Li X.Q., Wang J.H., Wang S.X. and Li X. (2000b) The phytosterones in *Rhaponticum uniflorum* (L.) DC. Journal of Shenyang Pharmaceutical University 17(4), 260-263 [in Chinese, with English abstract].

Abstract: *Rhaponticum uniflorum* (L.) DC. Four plant steroids were isolated from the roots and identified as moulting steroids; ecdysterone (1), ajugasterone C (2), ecdysterone-3-O- β -D-glucoside (ecdysterone-3-O- β -D-glucopyranoside) (3) and ecdysterone-25-O- β -D-glucoside (ecdysterone-25-O- β -D-glucopyranoside) (4), based on the physicochemical constants and spectral data, where compounds 3,4 are isolated for the first time from the plant.

Li Y., Cai L., Dong J-W., Xing Y., Duan W-H., Zhou H. and Ding Z-T. (2015) Innovative approach to the accumulation of rubrosterone by fermentation of *Asparagus filicinus* with *Fusarium oxysporum*. Journal of Agricultural and Food Chemistry 63, 6596-6602.

Abstract: Rubrosterone, possessing various remarkable bioactivities, is an insect-molting C19-steroid. However, only very small amounts are available for biological tests due to its remarkably limited content from plant sources. Fungi of genus *Fusarium* have been reported to have the ability to convert C27-steroids into C19-steroids. In this study, *Asparagus filicinus*, containing a high content of 20-hydroxyecdysone, was utilized to accumulate rubrosterone through solid fermentation by *Fusarium oxysporum*. The results showed that *F. oxysporum* had the ability to facilitate the complete biotransformation of 20-hydroxyecdysone to rubrosterone by solid-state fermentation. The present method could be an innovative and efficient approach to accumulate rubrosterone with an outstanding conversion ratio.

Li Y-c., Duan J., Yang S-m., Gao B. and Zhou J. (2016) Content determination of β -ecdysone in *Cyanotis* extract. Yunnan Chemical Technology (2), 28-30 [in Chinese, with an English abstract].

Liang J., Tang C. and Qin A. (2014) Determination of β -ecdysterone in *Achyranthes bidentata* Radix from different areas by HPLC. Asia-Pacific Traditional Medicine (1), 30-31 [in Chinese].

Abstract: Objective: To determine the content of β -ecdysterone in *Achyranthes bidentata* Radix from different areas and evaluate their qualities. Methods: the content of β -ecdysterone in *Achyranthes bidentata* Radix from different areas was measured by HPLC. Kromasil C18 column (250mm \times 4.6mm, 5 μ m) was used, with the mobile phase consisting of acetonitrile-water (15: 85). Results: The linear range of β -ecdysterone was 0.2064 ~ 2.58 μ g ($r = 0.9992$), the average recovery rate was 99.34%. Conclusion: There was a difference between the content of β -ecdysterone in *Achyranthes bidentata* Radix from different areas, but they meet the Pharmacopoeia requirements.

Liang X-k., Lei J-w., Gong H-y., Tang W-w., Ji L., Xie C-x., Liu D-f. (2019) Simultaneous determination of the contents of five components in *Achyranthes bidentatae* radix and rhizome by HPLC. Journal of Chinese Medicinal Materials (7), 1578-1583 [in Chinese].

Abstract: Objective: To determine the contents of five components (25R-inokosterone, 25S-inokosterone, ecdysterone, ginsenoside R0, chikusetsu saponin IVa) in *Achyranthes bidentata* Radix and rhizome. Methods: HPLC method was used to determine the contents of five components in *Achyranthes bidentata* radix, and the quality was comprehensively evaluated by cluster analysis and principal component analysis. Results: There was little difference in the contents of ecdysterone, 25S-inokosterone and 25R-inokosterone in *Achyranthes bidentata* Radix from different areas and different specifications, while there was great difference in the contents of ginsenoside R0 and chikusetsu saponin IVa; Cluster analysis and principal component analysis all divided *Achyranthes bidentata* radix into four categories. The contents of ecdysterone, 25R-inokosterone, ginsenoside R0, chikusetsu saponin IVa in rhizome were higher than that in *Achyranthes bidentata* radix, there was no significant difference in the content of 25S-inokosterone; Cluster analysis could distinguish the *Achyranthes bidentata* radix from rhizome. Conclusion: The quality of *Achyranthes bidentata* radix is affected by the field, specification and rhizome.

Liang X-l., Yi G-q., He G-x., Gan Li, Li J-y., Li Y-y. (2014) Determination of ajuforrestin A, ecdysterone and 8-O-acetylharpagide in *Ajuga nipponensis* Makino by HPLC. Chinese Traditional Patent Medicine (6), 1241-1244 [in Chinese].

Abstract: AIM: To establish an HPLC method for determining contents of ajuforrestin A, ecdysterone and 8-O-acetylharpagide in *Ajuga nipponensis* Makino. METHODS: The Sino Chrom ODS-BP column (250 mm \times 4.6 mm, 5 μ m) was used at 25 $^{\circ}$ C with the flow-rate of 0.8 mL/min. Ajuforrestin A was detected at detection wavelength of 228 nm with the mobile phase of acetonitrile-water (70 : 30). Ecdysterone was detected at detection wavelength of 243 nm with the mobile phase of acetonitrile-water (18 : 82). 8-O-Acetylharpagide was detected at detection wavelength of 212 nm with the mobile phase of acetonitrile-water (13 : 87). RESULTS: The linear response ranged from 0.24-2.40 μ g for ajuforrestin A ($r = 0.9984$), 0.40-4.00 μ g for ecdysterone ($r = 0.9962$), 0.40-4.00 μ g for 8-O-acetylharpagide ($r = 0.9986$), respectively. Recoveries were 98.90% with RSD 1.74% for ajuforrestin A, 97.36% with RSD 1.83% for ecdysterone, 99.18% with RSD 1.11% for 8-O-acetylharpagide, respectively.

CONCLUSION: The content of 8-O-acetylharpagide in *Ajuga nipponensis* Makino is the highest of the three constituents.

Liang X., Li Y., Fan H., Huang W., Zhang H., Cui Y. and Song X. (2019) Chemical constituents from the roots and rhizomes of *Silene tatarinowii* Regel. *Biochemical Systematics and Ecology* **86**, 103932 (doi.org/10.1016/j.bse.2019.103932).

Abstract: Chemical investigation of the root and rhizome of *Silene tatarinowii* Regel led to the isolation of nine ecdysteroids (1–9) and one sterol (10). All the compounds were determined on the basis of MS and NMR and by comparison with those in the literature. All these compounds were isolated from *S. tatarinowii* for the first time. Furthermore, compounds 7–10 were isolated from *Silene* genus for the first time, of which compounds 7 and 8 were isolated from Caryophyllaceae family members for the first time. This is the first study to report the chemical constituents of *S. tatarinowii* and the chemotaxonomic relationships between *Silene* and other genera of Caryophyllaceae.

Liedtke S., Lorch E., Goedings P. and Wichtl M. (1997) Isolierung von β -Ecdyson und Macranthosid I aus Wurzeln und Rhizomen von *Helleborus niger* subsp. *niger* (Ranunculaceae) [Isolation of β -ecdysone and macranthoside I from roots and rhizomes of *Helleborus niger* subsp. *niger* (Ranunculaceae)]. *Pharmazie* **52**(12), 964-965 [in German].

Liktor-Busa E. (2008) Analysis of the ecdysteroid profile of *Serratula wolffii* roots. Ph.D. Thesis, University of Szeged, Hungary, pp. 13.

Liktor-Busa E., Simon A., Tóth G., Fekete G., Kele Z. and Báthori M. (2007a) Ecdysteroids from *Serratula wolffii* roots. *Journal of Natural Products* **70**, 884-886.

Abstract: Two new natural ecdysteroids, 20,22-didehydrotaxisterone (1) and 1-hydroxy-20,22-didehydrotaxisterone (2), were isolated from the roots of *Serratula wolffii*. Their structures were elucidated by 1D and 2D NMR spectroscopy and mass spectrometry. The biological activities of these compounds were determined via oral aphid (*Acyrtosiphon pisum* (Harris)) tests. Compound 1 was inactive and compound 2 exhibited very low toxicity in the oral aphid test. The activities of these two ecdysteroids were in agreement with those of other 22-deoxyecdysteroids.

Liktor-Busa E., Simon A. and Báthori M. (2007b) *Serratula wolffii*, as a source of new ecdysteroids. *Planta Medica* **73**, DOI: 10.1055/s-2007-987144

Liktor-Busa E., Simon A., Tóth G. and Báthori M. (2008) The first two ecdysteroids containing a furan ring from *Serratula wolffii*. *Tetrahedron Letters* **49** 1738-1740.

Abstract: Two new ecdysteroids, named serfurosterone A and serfurosterone B, were isolated from a methanol extract of the roots of *Serratula wolffii*. Spectroscopic methods revealed that these compounds had previously unknown ecdysteroid structures with acetal functions in the side-chains.

Lin D-z., Wang G-s., Yang X-h. and Xu J-d. (2006) Studies on steroid constituents of *Achyranthes bidentata* Bl. *Zhongguo Yaoxue Zazhi* (Beijing, China) **41**(17), 1295-1297 [in Chinese].

Abstract: The objective of this paper is to investigate the steroid constituents of *Achyranthes bidentata* Bl. Solvent extraction and column chromatog. were used to isolate the non-alkaloid constituent, and physico-chem. constants and spectroscopic anal. were employed for structural elucidation. Three steroid constituents were isolated and their structures were elucidated to be 2 β , 3 β , 5 β , 14 α , 20 β , 22 α , 25-heptahydroxy-cholest-7-en-6-one (Polypodine B, 1); 2 β , 3 β , 14 α , 20 β , 22 α , 25-hexahydroxy-cholest-7-en-6-one (2); 2 β , 3 β , 20 β , 22 α , 25-pentahydroxy-cholesta-8, 14-dien-6-one (3), resp. Compound 3 is a new compound

Ling T-j., Ma W-z. and Wei X-y. (2003) Ecdysteroids from the roots of *Serratula chinensis*. *Redai Yaredai Zhiwu Xuebao* (Journal of Tropical and Subtropical Botany) **11**(2), 143-147 [In English, with a Chinese abstract].

Abstract: Seven ecdysteroids, 20-hydroxyecdysone (1), podecdysone C (2), 3-O-acetyl-20-hydroxyecdysone (3), 20-hydroxyecdysone-20, 22-butylidene acetal (4), shidasterone (5), atrotosterone C (6) and carthamosterone (7), were isolated from the roots of *Serratula chinensis* S. Moore. All compounds except compound 1 were isolated from this plant for the first time, and compound 4 was found to be a new ecdysteroid.

Liu F., Zhang L., Chen B., Li Z., Du Y. and Xiao G. (2014) NaCl solution stress on the bud and seedling growth and the content of β -ecdysone of *Cyanotis arachnoidea*. *Forestry Science Technology* (6), 6-9 [in Chinese]

Liu H., Huang Y., Zhang T., Wang Q. and Chen X. (2006) Studies on chemical constituents of *Paris delavayi* Franch. *Zhongguo Yaoke Daxue Xuebao* **37**(5), 409-412 [in Chinese].

Abstract: The chem. constituents in the rhizome of *Paris delavayi* Franch. were studied. Compounds were isolated by silica gel, Sephadex LH-20, and RP-C₁₈ column chromatog. and the structures were elucidated on the basis of physicochem. data and spectral characteristics. Eight compounds were isolated and identified as six steroidal saponins: paris saponin I (1), paris saponin II (2), 25(R)-diosgenin-3-O- α -L-rhamnopyranosyl(1 \rightarrow 2)- β -D-glucopyranoside (3), 25(R)-diosgenin-3-O- β -D-glucopyranosyl(1 \rightarrow 3)[α -L-rhamnopyranosyl(1 \rightarrow)]- β -D-glucopyranoside (4), 25(R)-pennogenin-3-O- α -L-arabinofuranosyl(1 \rightarrow 4)[α -L-rhamnopyranosyl(1 \rightarrow 2)]- β -D-glucopyranoside (5), 25(R)-pennogenin-3-O- α -L-rhamnopyranosyl(1 \rightarrow 4)- α -L-rhamnopyranosyl(1 \rightarrow 4)[α -L-rhamnopyranosyl(1 \rightarrow 2)]- β -D-glucopyranoside (6); one phytoecdysone: 2 β ,3 β ,14 α ,20 β ,22 α ,25-hexahydroxy-cholest-7-en-6-one (7); and daucosterol (8). All the eight compounds are obtained from this plant for the first time.

Liu H-Y., Ran X-H, Gong N-B., Ni W., Qin X-J., Hou Y-Y., Lu Y. and Chen C-X. (2013) Sesquiterpenoids from *Chloranthus multistachys*. *Phytochemistry* **88**, 112-118.

Abstract: An 8,9-*seco*-lindenane disesquiterpenoid, chloramultiol G, four eudesmane sesquiterpenoids, *ent*-(3*R*)-3-hydroxyatractylenolide III and multistalactones A–C, and four guaiane sesquiterpenoids, (1*R*,4*S*,5*R*,8*S*,10*S*)-zedoalactone A and multistalactones D–F, along with 14 known compounds, were isolated from whole plant tissues of *Chloranthus multistachys*. Their structures were established by extensive NMR experiments in conjunction with mass spectrometry. Except for chloramultiol G, the absolute stereochemistries of the other eight were confirmed by single-crystal X-ray crystallography and CD spectra. Nine compounds were tested for cytotoxicity against five human tumor cell lines and for antifungal activity against four microorganisms *in vitro*, but all were inactive.

Liu J., Zhou Y-q. and Peng C. (2005) Determination of the contents of ecdysterone in *Achyranthes bidentata*. *Journal of Anhui Traditional Chinese Medical College* (3), 43-44 [in Chinese].

Abstract: Objective: To determine the content of moulting steroids in ox-knee plants.

Methods: The content of moulting steroids in ox-knee was determined by high-performance liquid chromatography.

RESULTS: The linear relationship between moulting steroids was good in the range of 0.2 to 1.8 μ g. Conclusion:

This method is fast, simple and accurate, and can be used as a quantitative analysis method of moulting steroids.

Liu T. And Gao Z. (2011) Phytochemical investigation of *Woodwardia japonica*. *Planta Medica* (DOI: 10.1055/s-0031-1273584)

Abstract: The rhizome of *Woodwardia japonica* Smith (Blechnaceae) is a traditional Chinese medicine, named Gouji Guanzhong in Chinese, to cure flu and verminosis [1,2]. Its clinical use is the same as the rhizome of *Dryopteris crassirhizoma*, named as Mianma Guanzhong recorded in Chinese pharmacopoeia [3]. Previously, our group reported the isolation of a new trimeric phloroglucinol together with 12 known compounds from the rhizome of *Dryopteris crassirhizoma* [4]. In this study the phytochemical investigation of the ethanolic extract of *W. japonica* resulted in the isolation of four sterones, including ecdysterone (1), achyranthesterone A (2), ponasterone A (3), ponasteroside A (4), one triterpene, namely, woodwardic acid (5), and two flavonoids, including kaempferol-3-O-rhamnopyranoside-7-O-rhamnopyranoside (6) and kaempferol-3-O-(4-O-acetyl)-rhamnopyranoside-7-O-rhamnopyranoside (7). Their structures were determined on the basis of UV, 1D, 2D-NMR, MS and reported data. The compounds 1,2,3, and 4 are reported for the first time from this species

Liu Y., Liu J., Yu S., Huang X. and Hu Y. (2007) Studies on chemical constituents of *Cynanchum forrestii*. *Zhongguo Zhongyao Zazhi* **32**(6), 500-503 [in Chinese].

Abstract: The chem. constituents of *Cynanchum forrestii* were studied. Chromatog. techniques were applied to isolate the chem. constituents. The structures were identified on the basis of physico-chem. constants and spectroscopic data. Eight compounds were isolated from the 95% ethanol extract of the roots of *C. forrestii* and elucidated as (+)-5'-methoxyisolaricresinol 3a-O- β -D-glucopyranoside (1), hexahydroxycholest-7-en-6-one (2; 20E?), tylophorinidine (3), sucrose (4), palmitic acid (5), β -sitosterol (6), daucosterol (7) and nonanedioic acid (8). Compounds 1-3 from its genus and compounds 4-8 from the plant are obtained for the first time.

Lloyd-Jones J.G., Rees H.H. and Goodwin T.G. (1973) Biosynthesis of ecdysterone from cholesterol in *Taxus baccata*. *Phytochemistry* **12**, 569-572.

Abstract: Biosynthesis of ecdysterone from [4-¹⁴C, 3-³H]cholesterol in *Taxus baccata* does not involve obligatory oxidation at C-3 during the formation of the A/B-*cis* ring junction.

Lomovsky O., Korolyov K. and Kwon Y.S. (2003) Mechanochemical solubilization and mechanochemically assisted extraction of plant bioactive substances. *Science and Technology* **1**, 7-20.

Abstract: Mechanochemical solubilization and mechanochemically assisted extraction of plant bioactive compounds are discussed. The mechanochemical approach advances are illustrated by examples of some phytosterols, phytoecdysteroids and triterpenoid acids. The mechanical treatment of plant raw powder and solid reagents is used for chemical transformation of bioactive substance into soluble chemical form. In some cases it is possible to

increase the extraction yield of bioactive substance more than 50%, to elevate the selectivity of green product and to simplify the following purification, to exclude the organic solvents from technology. Use of powder products of mechanochemical treatment as a commercial product is the most economically effective realization. Utilization of powder containing solubilized steroids and triterpenoid acids as growth regulator and results of biological tests are discussed.

Louden D., Handley A., Taylor S., Lenz E., Miller S., Wilson I.D., Sage A. and Lafont R. (2001) Spectroscopic characterisation and identification of ecdysteroids using high-performance liquid chromatography combined with on-line UV-diode array, FT-infrared and ¹H nuclear magnetic resonance spectroscopy and time of flight mass spectrometry. *Journal of Chromatography A* 910(2), 237-246.

Abstract: A prototype multiply hyphenated reversed-phase HPLC system has been applied to the analysis of a mixture of pure ecdysteroids and an ecdysteroid-containing plant extract. Characterisation was achieved via a combination of diode array UV, ¹H NMR, FT-IR spectroscopy and time of flight (TOF) mass spectrometry. This combination of spectrometers allowed the collection of UV, ¹H NMR, IR and mass spectra for a mixture of pure standards enabling almost complete structural characterisation to be performed. The technique was then applied to a partially purified plant extract in which 20-hydroxyecdysone and polypodine B were identified despite incomplete chromatographic resolution and the presence of co-chromatographing interferents. The experimental difficulties in the use of such a systems for these analytes are described.

Louden D., Handley A., Lafont R., Taylor S., Sinclair I., Lenz E., Orton T. and Wilson I.D. (2002) HPLC analysis of ecdysteroids in plant extracts using superheated deuterium oxide with multiple on-line spectroscopic analysis (UV, IR, ¹H NMR and MS). *Analytical Chemistry* 74, 288-294.

Abstract: HPLC, using superheated D₂O as the mobile phase, combined with on-line characterization via a combination of diode array UV, ¹H NMR, FT-IR spectroscopy, and mass spectrometry has been used for the analysis of a standard of 20-hydroxyecdysone- and ecdysteroid-containing plant extracts. This combination of spectrometers enabled the on-flow collection of UV, ¹H NMR, IR, and mass spectra not only for pure 20-hydroxyecdysone (100-400 microg on column) but also the major ecdysteroids present in crude extracts of *Silene otites*, *Silene nutans*, and *Silene frivaldiskyana*. The ecdysteroids unequivocally identified in these extracts included 20-hydroxyecdysone, polypodine B, and integristerone A.

Lu Z., Zheng J., Wang H., Ye W. and Zhao S. (2009) Chemical constituents in the stem heartwood of *Vitex quinata*. *Jiangsu Pharmaceutical and Clinical Research* (4), 287-289 [in Chinese, with an English abstract].

Lu Z., Zhu Y., Huang R. and Li Y. (2017) Research on quality standards of minority areas specialty herbs medicine *Vitex quinata*. *Journal of Guangxi Medical University* (4), 607-611 [in Chinese].

Abstract: Objective: To establish quality standard for minority areas specialty herbs medicine *Vitex quinata* (Lour.) Will. Methods: Ten batches of *Vitex quinata* were collected. The morphology and microscopic structure were observed, and the pharmacognostic identification, the content and limit of ecdysterone were analysed by thin-layer chromatography (TLC) and high-performance liquid chromatography (HPLC) methods. According to Chinese Pharmacopoeia (2010 edition), the moisture, total ash content, acid-insoluble ashes content, and alcohol-soluble extract were quantified. Results: The character, macroscopical identification and identification *Vitex quinata* by TLC were established. The percentage of moisture in crude drug was ranged from 9.9% to 11.7%, total ash was 1.12%-4.93%, acid-insoluble ash was 0.21%-2.79%, alcohol-soluble extract was 2.3%-8.2%. The content of ecdysterone was 0.14% to 1.27%. Ecdysterone was linear in the range of 0.204-2.04μg, and the average recovery rate was 101.5%. Conclusion: The method was accurate with good reproducibility and could be used to control the quality of *Vitex quinata* (Lour.) Will.

Luo Y-n., Lin H., Lin L-w. (2014) Study on microwave processing technology of *Achyranthes bidentata* Radix by orthogonal design. *Journal of Chinese Medicinal Materials* (8), 1353-1356 [in Chinese].

Abstract: Objective: To study the microwave processing method of *Achyranthes bidentata* Radix processed with wine. Methods: The content of total saponins, oleanolic acid and β-ecdysterone were determined as the indices to get the optimal microwave processing technology by orthogonal design. Results: The best technology of *Achyranthes bidentata* Radix processed with wine was: the amount of vinegar was 20%, moistening time was 60 min, with 60% microwave heating for 3 min. Conclusion: This method is simple, practical, scientific and easy to control.

Ma H., Yang Q., Chen M. and Li Y. (2018) Extraction method of 20-hydroxy]ecdysone from *Pogonotherum paniceum*. *Faming Zhuanli Shenqing/Chinese Patent* CN 108084242 A 20180529 [in Chinese].

Ma Y., Che Z., Bi K., Wang X. and Huang W. (2000) Determination of ecdysterone in the root of *Achyranthes bidentata* Bl. by HPLC. *Acta Pharmaceutica Sinica* (4), 313-315 [in Chinese].

Macek T. and Vaněk T. (1994) *Pteridium aquilinum* (L.) Kuhn (Bracken Fern): *in vitro* culture and the production of ecdysteroids. In: Biotechnology in Agriculture and Forestry 26; Medicinal and Aromatic Plants VI (Ed. Bajaj Y.P.S.) Springer-Verlag, Berlin/Heidelberg, pp. 299-315.

Madhavan S.C., Bose C., Perakathusseril T.M. and Banerji A. (2014) Indian medicinal plant, *Coscinium fenestratum*, a new bio source for the multifunctional bio active molecule ecdysterone. International Journal of Herbal Medicine 2(5) 5-9.

Abstract: Phytochemical investigation on *Coscinium fenestratum* (Gaertn.) Collebr., an important Ayurvedic plant, revealed the presence of significant amounts of ecdysterone in the stem (0.22%) and leaves (0.12%), in addition to berberine. Ecdysterone was characterized using High Performance Liquid Chromatography (HPLC), Infrared Spectroscopy (FT-IR) and Liquid Chromatography-Mass Spectroscopy (LC-MS). Isolation of this multi-functional bioactive compound will throw light on the chemical basis for the various pharmacological effects of *Coscinium* plant extract.

Malakov P.Y., Papanov G.Y., de la Torre M.C. and Rodriguez B. (1998) Constituents of *Ajuga laxmanii*. Fitoterapia 69(6), 552.

Malinski M.P., Kikowska M., Kruszka D., Napierala M., Florek E., Sliwinska E. and Thiem B. (2019) Various *in vitro* systems of Ragged Robin (*Lychnis flos-cuculi* L.): a new potential source of phytoecdysteroids? Plant Cell, Tissue and Organ Culture 139: 39-52.

Abstract: *Lychnis flos-cuculi* L. is a species containing ecdysteroids, triterpenoid saponins, flavonoids, and phenolic acids, and therefore is a plant of potential medicinal value. In the presented research, diverse *in vitro* cultures of this taxon were developed to obtain the uniform material capable of producing ecdysteroids, including micropropagated plantlets, shoot cultures, liquid agitated whole plant cultures with fast-growing roots, and callus. A protocol of micropropagation through axillary bud formation was established using plant growth regulators at different concentrations and combinations. All the variants of plant growth regulator supplementation significantly affected a shoot proliferation rate ranging from 8 to 16 plantlets per explant, depending on the medium; DNA content of all the studied plant materials was similar. The thin-layer chromatography analysis of the extracts revealed the presence of ecdysteroids in every plant material apart from callus. The content of 20-hydroxyecdysone and polygodine B was evaluated by high-performance liquid chromatography. Agitated plantlets were *in vitro* cultures that efficiently formed abundant root biomass with significant concentrations of ecdysteroids. *In vitro*-derived adventitious roots contained twofold higher content of ecdysteroids than those of intact plants. The organs of flowering *in vitro*-propagated plants, transferred to experimental plot, contained twice as much ecdysteroids when compared to the organs of plants from the natural site, among which flowers were the richest in ecdysteroids. The results revealed that adventitious roots from *L. flos-cuculi* agitated cultures can be considered as an alternative biotechnological source of biomass rich in pharmaceutically active ecdysteroids.

Malinski M.P., Budzianowski J., Kikowska M., Derda M., Jaworska M.M., Mlynarczyk D.T., Szukalska M., Florek E. and Thiem B. (2021) Two ecdysteroids isolated from micropropagated *Lychnis flos-cuculi* and the biological activity of plant material. Molecules 26, article 904 (doi: 10.3390/molecules26040904).

Abstract: Genetically uniform plant material, derived from *Lychnis flos-cuculi* propagated *in vitro*, was used for the isolation of 20-hydroxyecdysone and polygodine B and subjected to an evaluation of the antifungal and antiamebic activity. The activity of 80% aqueous methanolic extracts, their fractions, and isolated ecdysteroids were studied against pathogenic *Acanthamoeba castellanii*. Additionally, a Microtox[®] acute toxicity assay was performed. It was found that an 80% methanolic fraction of root extract exerts the most potent amoebicidal activity at IC₅₀ of 0.06 mg/mL at the 3rd day of treatment. Both ecdysteroids show comparable activity at IC₅₀ of 0.07 mg/mL. The acute toxicity of 80% fractions at similar concentrations is significantly higher than that of 40% fractions. Crude extracts exhibited moderate antifungal activity, with a minimum inhibitory concentration (MIC) within the range of 1.25–2.5 mg/mL. To the best of our knowledge, the present report is the first to show the biological activity of *L. flos-cuculi* in terms of the antifungal and antiamebic activities and acute toxicity. It is also the first isolation of the main ecdysteroids from *L. flos-cuculi* micropropagated, ecdysteroid-rich plant material.

Mamadaliyeva N.Z., Ramazanov N.S., Dinan L.N. and Saatov Z. (2000) Phytoecdysteroids of plants of the genus *Silene*. 2-Dehydro-20-hydroxyecdysone 3-*O*-benzoate from *Silene wallichiana*. Khimiya Prirodnykh Soedinenii (5), 405-407 [in Russian]/Chemistry of Natural Compounds 36(5) 513-515 [in English].

Mamadaliyeva N.Z., Saatov Z., Kachala V.V. and Shashkov A.S. (2002a) Phytoecdysteroids of plants of the *Silene* genus. 2-Deoxyecdysterone 25-acetate from *Silene wallichiana*. Khimiya Prirodnykh Soedinenii (2), 149-150 [in Russian]/Chemistry of Natural compounds 38(2), 179-181 [in English].

Abstract: The new ecdysteroid 2-deoxyecdysterone-25-acetate was isolated from roots of *Silene wallichiana* Klotzsch.

Mamadaliyeva N.Z., Zibareva L.N. and Saatov Z. (2002b) Phytoecdysteroids of *Silene linicola*. *Khimiya Prirodnykh Soedinenii* (3), 225-227 [in Russian]/*Chemistry of Natural Compounds* 38(3), 268-271 [in English]

Abstract: Seven ecdysteroids isolated from the aerial part of *Silene linicola* are identified using ¹H and ¹³C NMR methods. Their yield from raw material was determined.

Mamadaliyeva N.Z., Zibareva L.N., Saatov Z. and Lafont R. (2003) Phytoecdysteroids of *Silene viridiflora*. *Khimiya Prirodnykh Soedinenii* (2), 150-153 [in Russian]/*Chemistry of Natural Compounds* 39(2), 199-203 [in English].

Abstract: The known ecdysteroids polipodine B, 2-deoxyecdysterone, ecdysterone, 26-hydroxypolipodine B, integristerone A, and sileneosides A and D were observed in the aerial part of *Silene viridiflora* L. (Caryophyllaceae). 26-Hydroxypolipodine B was observed in this species for the first time.

Mamadaliyeva N.Z., Zibareva L.N., Evrard-Todeschi N., Girault J.P., Maria A., Ramazanov N.S., Saatov Z. and Lafont R. (2004a) New minor ecdysteroids from *Silene viridiflora*. *Collection of Czechoslovak Chemical Communications* 69(8), 1675-1680.

Abstract: Two new minor ecdysteroids, the 2,22- and 3,22-diacetates of 20,26-dihydroxyecdysone have been isolated from the aerial part of *Silene viridiflora* L. (Caryophyllaceae).

Mamadaliyeva N.Z., Zibareva L.N., Lafont R., Dinan L. and Saatov Z. (2004b) Phytoecdysteroids from the *Silene* genus. *Chemistry of Natural Compounds* 40(6), 574-578 [in English]/*Khimiya Prirodnykh Soedinenii* (6), 472-475 [in Russian].

Abstract: Research results on the isolation and identification of ecdysteroids from plants of the *Silene* L. genus were presented.

Mamadaliyeva N.Z., Egamberdieva D., Lafont R. and Girault J.P. (2008) Phytoecdysteroids and antibacterial activity of the plant *Coronaria flos-cuculi*. *Chemistry of Natural Compounds* 44(3), 404-406 [in English]/*Khimiya Prirodnykh Soedinenii* (3), 323-324 (2008) [in Russian].

Abstract: Plants of the genus *Coronaria* (Caryophyllaceae) are potential producers of ecdysteroids [1]. It was shown previously that a representative of this genus, *C. coreacea*, contains 20-hydroxyecdysone and viticosterone E [2]. We investigated the ecdysteroid composition of *C. flos-cuculi* L. that was introduced to the Botanical Garden of the Academy of Sciences of the Republic of Uzbekistan. According to TLC of the methanol extract, the plant contains at least 11 ecdysteroids, 3 of them in large amounts.

Mamadaliyeva N.Z., Janibekov A.A., Girault J.-P. and Lafont R. (2010) Two minor phytoecdysteroids of the plant *Silene viridiflora*. *Natural Product Communications* 5 1-4.

Abstract: Chemical investigations of *Silene viridiflora* (L.) yielded a new ecdysteroid, 20-hydroxyecdysone 20,22-monoacetate-25-acetate (1), and a known ecdysteroid, 2-deoxypolipodine B-3-β-D-glucoside (2). The elucidation of the chemical structures was established by 1D and 2D NMR experiments.

Mamadaliyeva N.Z., El-Readi M.Z., Janibekov A.A., Tahrani A. and Wink M. (2011) Phytoecdysteroids of *Silene guntensis* and their *in vitro* cytotoxic and antioxidant activity. *Zeitschrift für Naturforschung* 66c 215-224.

Abstract: Phytoecdysteroids from aerial parts of *Silene guntensis* B. Fedtsch were investigated and three phytoecdysteroids were isolated: 2,3-diacetate-22-benzoate-20-hydroxyecdysone (1), 2-deoxy-20-hydroxyecdysone (2), and 20-hydroxyecdysone (3). Their chemical structures were elucidated by DEPT, COSY, ¹H and ¹³C NMR spectroscopy. The isolated compounds 1-3 and crude extracts were evaluated for their antiproliferative and antioxidant activities. They exhibited substantial inhibition of cell growth against human cervix adenocarcinoma (HeLa), hepatocellular carcinoma (HepG-2), and breast adenocarcinoma (MCF-7) cells. The chloroform extract showed potent cytotoxic effects [IC₅₀ values (26.58 ± 1.88) μg/mL, (20.99 ± 1.64) μg/mL, and (18.89 ± 2.36) μg/mL, respectively]. The new compound 1 was mildly cytotoxic compared to extracts [(127.97 ± 11.34), (106.76 ± 7.81), and (203.10 ± 19.56) μg/mL, respectively]. Water and n-butanol extracts exhibited good antioxidant activities [IC₅₀ values of (68.90 ± 6.45) μg/mL and (69.12 ± 5.85) μg/mL, respectively].

Mamadaliyeva N.Z., El-Readi M.Z., Ovidi E., Ashour M.L., Hamoud R., Sagdullaev S.S., Azimova S.S., Tiezzi A. and Wink M. (2013) Antiproliferative, antimicrobial and antioxidant activities of the chemical constituents of *Ajuga turkestanica*. *Phytopharmacology* 4(1), 1-18.

Abstract: *Ajuga turkestanica* Rgl. Brigg (Lamiaceae) is a medicinal plant from Uzbekistan. Methanol, chloroform, butanol, and water extracts as well as isolated phytoecdysteroids and iridoids were evaluated for their antioxidant,

cytotoxic and antibacterial activities. Water and butanol extracts exhibited good antioxidant activity with IC 50 values of 7.24 ± 0.82 and 14.57 ± 1.64 $\mu\text{g/mL}$. The chloroform extract showed potent cytotoxic effects against the cancer cell lines HeLa, HepG-2, and MCF-7 with IC 50 values of 7.13 ± 0.85 , 9.03 ± 0.92 , and 10.77 ± 1.44 $\mu\text{g/mL}$, respectively. Compared to the extracts, isolated phytoecdysteroids and iridoids showed weak cytotoxic activity. The chloroform extract has antimicrobial properties even against multiresistant strains like *Staphylococcus aureus* MRSA 1000/93 and *Streptococcus pyogenes* ATCC 12344. The methanol and chloroform extracts of *A. turkestanica* were further investigated for their GLC-volatile components using GLC/FID and GLC/MS. Pregna-4,9 (11)-dien-20-ol-3-on-19-oic acid lactone (19.58%), 20-methyl-pregna-5,17-dien-3 β -ol (12.93%), 3,7-dioxocholan-24-oic acid (10.53%) and betulin (10.18%) were detected as the major compounds.

Mamadaliyeva N.Z., Böhmendorfer S., Zengin G., Bacher M., Potthast A., Akramov D.K., Janibekov A. and Rosenau T. (2019) Phytochemical and biological activities of *Silene viridiflora* extractives. Development and validation of a HPTLC method for quantification of 20-hydroxyecdysone. *Industrial Crops & Products* **129**, 542-548.

Abstract: *Silene viridiflora* (Caryophyllaceae) has adaptogen, tonic, actoprotector and immunomodulator properties that are mainly attributed to ecdysteroids. The aim of the present study is to investigate the chemical constituents of essential oil as well as the 20-hydroxyecdysone content in the methanol extract *S. viridiflora* and its related biological activity. GC-MS analysis showed the essential oil of *S. viridiflora* to have a complex composition, which was dominated by methyl palmitate (8.05%), 3-hexen-1-ol (7.54%), 3-hexenyl benzoate (3.86%), β -myrcene (3.80%), 1,1-diethoxy-2-phenylethane (3.65%), hexahydrofarnesyl acetone (3.20%) and 4-terpineol (2.47%). A high performance thin-layer chromatography (HPTLC) method was developed to analyze the methanolic extracts of *S. viridiflora* and to quantify the main ecdysteroid, 20-hydroxyecdysone. Validation showed that the method was accurate over the entire calibration range and resulted in errors of less than 10% in the 100–815 $\mu\text{g/ml}$ concentration range. Antioxidant and enzyme inhibitory potentials of methanol extract and individual compounds of *S. viridiflora* were evaluated. The results could provide a starting point for designing novel phyto-pharmaceuticals.

Mamatkhanov A.U., Shamsutdinov M.R.E. and Shakirov T.T. (1979) Isolation of ecdysterone from inflorescences of *Rhaponticum integrifolium*. *Khimiya Prirodnykh Soedinenii* (5), 667-668 [in Russian].

Abstract: A method has been developed for isolating ecdysterone from the inflorescences of *Rh. integrifolium*. The yield is 0.15% on the weight of the raw material.

Mamatkhanov A. U., M. R.-I. Shamsutdinov and T. T. Shakirov (1980) Isolation of ecdysterone from the roots of *Rhaponticum carthamoides*. *Khimiya Prirodnykh Soedinenii* (4), 528-529 [in Russian]/*Chemistry of Natural Compounds* (4), 381-382 [in English].

Mamatkhanov A.U., Shamsutdinov M.-R.I. and Shakirov T.T. (1983) Isolation of ecdysterone. *Khimiya Prirodnykh Soedinenii* (5), 601-605 [in Russian]; *Chemistry of Natural Compounds* (5), 565-568 [in English].

Abstract: The kinetics of the extraction of ecdysterone from the roots with rhizomes of *Rhaponticum carthamoides* has been studied. The results of investigations on the selection of the optimum solvent for extracting ecdysterone from the raw material, for the process of sorption purification, and for the recrystallization of the ecdysterone are given.

Mamedov N.A., Mamadaliyeva N.Z., Janibekov A.A. and Craker L.E. (2017) Screening some plants from *Silene* genus for 20-hydroxyecdysone. *International Journal of Secondary Metabolite* **2**, 74-77.

Abstract: Eight *Silene* species include *S. brachuica*, *S. guntensis*, *S. linicola*, *S. oreina*, *S. praemixta*, *S. pseudotites*, *S. viridiflora* and *S. wallichiana* were screened for the main ecdysteroid 20-hydroxyecdysone by HPLC using the UV spectroscopy. HPLC analyses have shown all *Silene* plants except *S. oreina* contain 20-hydroxyecdysone, but in different concentration. Studies have shown that *S. praemixta* and *S. viridiflora* are rich phytoecdysteroids containing plants and the yields of total ecdysteroids are 2.0% and 1.6%, respectively. The results of investigation species of *Silene*: *S. brachuica*, *S. praemixta*, *S. viridiflora*, *S. guntensis*, *S. linicola*, *S. pseudotites* and *S. wallichiana* showed that the yields of 20E of these plants are 0.03, 0.27, 0.35, 0.082, 0.367, 0.071 and 0.08% respectively. The plants of *S. linicola*, *S. praemixta* and *S. viridiflora* suggested to be used for production of ecdysteroids containing preparations.

Mao X., Lu W. and Hu J. (2013) Molting sterol composition of *Diploclisia glaucescens*. *Yaoxue Fuwu Yu Yanjiu* **13**(1), 60-61 [in Chinese].

Abstract: The molting sterol composition of *Diploclisia glaucescens* was analyzed. *Diploclisia glaucescens* was extracted using ethanol. The structure of the isolated components was identified using spectral analysis. The isolated components were calonysterone, stachysterone B, stachysterone C and 20-hydroxyecdysone. Calonysterone, stachysterone B, stachysterone C were isolated from *Diploclisia glaucescens* for the first time.

Marco M-P., Sánchez-Baeza F.J., Camps F. and Coll J. (1993) Phytoecdysteroid analysis by high-performance liquid chromatography-thermospray mass spectrometry. *Journal of Chromatography* **641**, 81-87.

Abstract: The potential of high-performance liquid chromatography-mass spectrometry with thermospray interface to identify and analyse ecdysteroids has been investigated. The response of eight different ecdysteroids and their acetone derivatives is studied by positive- and negative-ion detection as well as with single-ion monitoring and scan mass detection modes. The usefulness of this technique for ecdysteroid identification and quantitation is discussed and, finally, the identification of a new phytoecdysteroid isolated from *Polypodium vulgare* is presented as an application of this technology. A fast and easy procedure for the extraction and purification of phytoecdysteroids is also described.

Marinho BM, Fernandes DN, Chicoti MZ, Ribeiro GJG, Almeida VG, Santos MGD, Guimarães VHD, Marchioretto MS, Martins HR, de Melo GEBA and Gregorio LE. (2021) Phytochemical profile and antiproliferative activity of human lymphocytes of *Gomphrena virgata* Mart. (Amaranthaceae). *Natural Product Research* **25**, 1-7 (doi: 10.1080/14786419.2021.1895151).

Abstract: *Gomphrena virgata* Mart. popularly known as ‘Cangussu-branco’, is used in Brazilian folk medicine to treat inflammations and infections. This work aimed to carry out phytochemical analysis and evaluate the anti-inflammatory potential of *Gomphrena virgata*. In the phytochemical investigation, in addition to the presence of two ecdysteroids, 20 R-dihydroxyecdysone and 20-hydroxyecdysone, identified by HPLC-PDA-MS and NMR, 22 compounds were identified by GC-MS. In the cytotoxicity study, the aqueous extract of the roots of this species did not show in vitro toxicity of PBMCs in the concentrations of 250, 500 and 1000 µg/mL when analyzed by the trypan blue exclusion method. Also, it was effective in reducing lymphocyte proliferation, stimulated with the mitogen PHA, by 26.02%, 48.57% and 50.49% when compared to dexamethasone, respectively. In this work we present information about the phytochemicals of *G. virgata*, showing that the species is promising in obtaining compounds with medicinal potential mainly anti-inflammatory potential.

Martins de Jesus C.C., de Araújo M.H., Simão T.L.B.V., Lasunskaja E.B., Barth T., Muzitano M.F. and Pinto S.C. (2020) Natural products from *Vitex polygama* and their antimycobacterial and anti-inflammatory activity, *Natural Product Research* (DOI: 10.1080/14786419.2020.1868461).

Abstract: Tuberculosis (TB) remains a worldwide public health threat because of the emergence of resistant strains and subsequent inappropriate response to current therapy. We have been studying the *restinga* plants’ antimycobacterial and anti-inflammatory potential. Dichloromethane fraction (DCM) from *Vitex polygama* Cham. showed high activity against *Mycobacterium tuberculosis* (Mtb) H37Rv. In this context, DCM fraction and isolated compounds were investigated against Mtb H37Rv and M299 (MDR strain) and for their immunomodulatory and cytotoxicity actions. Orientin showed the best antimycobacterial effect against Mtb M299 MDR strain (MIC₅₀ 15.4 ± 1.6 µg/mL), capacity of inhibiting NO production by macrophages (IC₅₀ 6.5 ± 1.2 µg/mL) and no significant cytotoxicity. The antimycobacterial effect of orientin was also observed on Mtb H37Rv intracellular growth in RAW 264.7 macrophages (MIC₅₀ 3.5 ± 1.1 and MIC₉₀ 9.1 ± 1.0 µg/mL). This is the first report describing the antimycobacterial effect of orientin, in both extra- and intracellular growth.

Martinussen I., Volodin V., Volodina S. and Uleberg E. (2011) Effect of climate on plant growth and level of adaptogenic compounds in maral root (*Leuzea carthamoides* (Willd.) DC.), crowned saw-wort (*Serratula coronata* L.) and roseroot (*Rhodolia rosea* L.). *The European Journal of Plant Science and Biotechnology* **5** (special issue 1), pp. 6.

Martucciello S, Paoletta G., Muzashvili T., Skhirtladze A., Pizza C., Caputo I. And Piacente S. (2017) Steroids from *Helleborus caucasicus* reduce cancer cell viability inducing apoptosis and GRP78 down-regulation. *Chemico-Biological Interactions* (doi: 10.1016/j.cbi.2017.11.002).

Abstract: *Helleborus caucasicus* (Ranunculaceae) is an endemic plant of the Caucasian flora, widely distributed in West Georgia. Biological activities for the extracts of some *Helleborus* species including *H. caucasicus* have been reported. In this work we found that butanolic extract of the underground parts of *H. caucasicus* and isolated compounds decreased cell viability in vitro on cancer cell line of lung origin (Calu-1) in a concentration-dependent manner, compared to the normal cell line. In particular, we identified that furostanol derivative (25S)-22 α ,25-epoxyfurost-5-ene-3 β ,11 β ,26-triol 26-O- β -d-glucopyranoside (5), 20-hydroxyecdysone (6), and 3 β ,5 β ,14 β -trihydroxy-19-oxo-bufa-20,22-dienolide 3-O- α -l-rhamnopyranoside, known as deglucohellebrin (7) exerted a strong cytotoxic effect on the same cells and on other cancer cell lines (HepG2 and Caco-2) reducing the S-phase entry (compound 6) and inducing cell apoptosis associated with activation of caspase-3 (compound 7). Moreover we demonstrated that 6 and 7 significantly decreased protein expression of GRP78, a general ER-stress marker, suggesting pro-apoptotic functions. These findings indicated that selected compounds from *H. caucasicus* are potential interesting agents in anti-cancer therapy.

Matsumoto T. and Tanaka N. (1991) Production of phytoecdysteroids by hairy root cultures of *Ajuga reptans* var. *atropurpurea*. *Agricultural and Biological Chemistry* **55**(4), 1019-1025.

Abstract: Over 20 'hairy root' clones of *Ajuga reptans* var. *atropurpurea* (Labiatae) were obtained from transformation with *Agrobacterium rhizogenes* MAFF 03-01724. Growth, opine synthesis and production of phytoecdysteroids in the clones have been examined. Mikimopine, a specific opine synthesized in the tissue transformed with strain MAFF 03-01724, was detected in most of the clones, and four phytoecdysteroids, 20-hydroxyecdysone, norcyasterone B, cyasterone and isocyasterone, were detected in all of the clones.

It was found that the production of phytoecdysteroids was closely related to the growth of hairy roots, by monitoring the changes in the culture. One of the rapidly growing and well branching hairy root lines, Ar-4, was selected and used for fermenter experiments. On an air-lift type culture of Ar-4 for 45 days, the weight of the root tissue was increased by 230 times and the content of 20-hydroxyecdysone reached 0.12% on a dry weight basis. This content is 4 times higher than those of the original roots.

Matsuoka T., Imai S., Sakai M. and Kamada M. (1969) Studies on phytoecdysones - a review of our works. *Annual Report of the Takeda Research Laboratory* **28**, 221-271 [in Japanese].

McMorris T.C. and Voeller B. (1971) Ecdysones from gametophytic tissues of a fern. *Phytochemistry* **10**, 3253-3254.

Abstract: Ponasterone A and crustecdysone have been isolated from culture filtrates of the fern gametophyte *Pteridium aquilinum* (L.) Kuhn.

Melé, E., Messegueur, R., Gabarra, J., Tomás, J., Coll, J. and Camps, F. (1992) *In vitro* bioassay for the effect of *Ajuga reptans* phytoecdysteroids on *Trialeurodes vaporariorum* larval development. *Entomologia Experimentalis et Applicata* **62**, 163-168.

Meng D.L., Li X., Wang J.H. and Li W. (2005) A new phytosterone from *Achyranthes bidentata* Bl. *Journal of Asian Natural Products Research* **7**(2), 181-184.

Abstract: A new phytosterone named achyranthesterone A (1), along with the three known compounds stachysterone D (2), beta-ecdysone (3) and polypodine B (4) have been isolated from the roots of *Achyranthes bidentata* Bl. The structure of the new compound was determined as 2beta,3beta,14alpha,20S,21,22R,25-heptahydroxycholest-7-en-6-one on the basis of physico-chemical properties and spectral methods.

Meng Y., Whiting P., Šik V., Rees H.H. and Dinan L. (2001a) Ecdysteroids and bufadienolides from *Helleborus torquatus* (Ranunculaceae). *Phytochemistry* **57**(3), 401-407.

Abstract: Three bufadienolides, hellebortin A (5-[β-D-glucopyranosyloxy]-10,14,16-trihydroxy-19-nor-{5β,10β,14β,16β}-bufa-3,20,22-trienolide [1]), hellebortin B (5-[β-D-glucopyranosyloxy]-3,4-epoxy-14-hydroxy-19-oxo-bufa-20,22-dienolide [2]) and hellebortin C (5-[β-D-glucopyranosyloxy]-3,4-epoxy-10,14-dihydroxy-19-nor-bufa-20,22-dienolide [3]), together with 20-hydroxyecdysone 3-O-β-D-glucoside (4) and 20-hydroxyecdysone (5) have been isolated by bioassay- and RIA-directed HPLC analyses of a methanol extract of the seeds of *Helleborus torquatus*. The structure and relative stereochemistry of the novel bufadienolide hellebortin A (1) and the structures of hellebortin B (2) and hellebortin C (3) were determined unambiguously by comprehensive analyses of their 1D and 2D NMR data. These five compounds are isolated from *Helleborus torquatus* for the first time. The biological activities of compound 1, 4 and 5 as ecdysteroid agonists and antagonists have been assessed.

Three bufadienolides (hellebortins A-C), together with the known ecdysteroids 20-hydroxyecdysone and 20-hydroxyecdysone 3-glucoside have been identified from seeds of *Helleborus torquatus*.

Meng Y., Bourne P.C., Whiting P., Šik V. and Dinan L. (2001b) Identification and ecdysteroid antagonist activity of three oligostilbenes from the seeds of *Carex pendula* (Cyperaceae). *Phytochemistry* **57**(3), 393-400.

Abstract: Methanolic extracts of seeds of several (*Carex* species) were found to antagonise the action of 20-hydroxyecdysone in the *Drosophila melanogaster* microplate-based B(II) cell bioassay. Bioassay-guided HPLC analysis of seeds of *Carex pendula* (drooping sedge) provided one previously unknown tetrastilbene (cis-miyabenol A) and two known oligostilbenes (kobophenol B and cis-miyabenol C) as the biologically active compounds (EC₅₀ values were 31, 37 and 19 microM, respectively, vs. 5 x 10⁻⁸ M 20-hydroxyecdysone). The structures and relative stereochemistries of these compounds were deduced by comprehensive 1D- and 2D-NMR experiments. These compounds are isolated from *Carex pendula* for the first time. *In vitro* experiments with dipteran and lepidopteran ecdysteroid receptor proteins demonstrate that the oligostilbenes are able to compete with radiolabelled ecdysteroid ([³H]ponasterone A) for occupancy of the ligand binding site. IC₅₀/K_i values are similar to the EC₅₀ values obtained in the B(II) bioassay.

Meng Y., Whiting P., Šik V., Rees H.H. and Dinan L. (2001c) Limnantheoside C (20-hydroxyecdysone 3-O-β-D-glucopyranosyl-[1-3]-β-D-xylopyranoside), a phytoecdysteroid from seeds of *Limnanthes alba* (Limnathaceae). *Zeitschrift für Naturforschung* **56c**, 988-994.

Abstract: A new ecdysteroid glycoside, limnantheoside C (20-hydroxyecdysone 3-O-beta-D-glucopyranosyl-[1-3]-beta-D-xylopyranoside [1]), together with limnantheoside A (20-hydroxyecdysone 3-O-beta-D-xylopyranoside [2]) and 20-hydroxyecdysone (3) have been isolated by bioassay/RIA-directed HPLC analyses of a methanol extract of the seedmeal of *Limnanthes alba* Hartw. ex Benth. The structure of the novel ecdysteroid glycoside (1) was determined unambiguously by UV, LSIMS and a combination of 1D- and 2D-NMR experiments. These three compounds are isolated from *Limnanthes alba* for the first time.

Meng Y., Whiting P., Zibareva L., Bertho G., Girault J.-P., Lafont R. and Dinan L. (2001d) Identification and quantitative analysis of the phytoecdysteroids in *Silene* species (Caryophyllaceae) by high-performance liquid chromatography: novel ecdysteroids from *S. pseudotites*. *Journal of Chromatography A* **935**, 309-319.

Abstract: Many species in the genus *Silene* (Caryophyllaceae) have previously been shown to contain ecdysteroids and this genus is recognised as a good source of novel ecdysteroid analogues. We have used ecdysteroid-specific radioimmunoassays and the microplate-based *Drosophila melanogaster* B(II) cell bioassay for ecdysteroid agonist and antagonist activities to identify further phytoecdysteroid-containing species in this genus. The main ecdysteroid components from 10 *Silene* species (*S. antirrhina*, *S. chlorifolia*, *S. cretica*, *S. disticha*, *S. echinata*, *S. italica*, *S. portensis*, *S. pseudotites*, *S. radicata*, *S. regia*) were isolated and identified, mainly by normal-phase and reversed-phase high-performance liquid chromatography. The amount of each ecdysteroid was determined by comparing chromatogram peak areas with those for reference 20-hydroxyecdysone (20E) on reversed-phase HPLC. 20E is the most abundant ecdysteroid in each of the *Silene* extracts. Polypodine B, 2-deoxy-20-hydroxyecdysone and ecdysone are also common ecdysteroids in these *Silene* species, but the proportions of these ecdysteroids vary between the *Silene* species. HPLC proved to be a quick and effective way to screen *Silene* species, determine ecdysteroid profiles and, hence, identify extracts containing novel analogues. An extract of the aerial parts of *S. pseudotites* was found to contain several new ecdysteroids. These have been isolated and identified spectroscopically (by NMR and mass spectrometry) as 2-deoxyecdysone 22beta-D-glucoside, 2-deoxy-20,26-dihydroxyecdysone and 2-deoxypolypodine B 3beta-D-glucoside. Additionally, (5alpha-H)-2-deoxyintegristerone A (5alpha-2H 91%, 5alpha-1H 9%) was isolated as an artefact. This study contributes to the understanding of ecdysteroid distribution in *Silene* species and provides further information on the chemotaxonomic significance of ecdysteroids in *Silene* species.

Messeguer J., Melé E., Reixach N., Irurre-Santilari J and Casas J. (1998) *Polypodium vulgare* L. (wood fern): *in vitro* cultures and the production of phytoecdysteroids. *Biotechnology in Agriculture and Forestry* **41** (Medicinal and Aromatic Plants X, Ed, Bajaj Y.P.S.), Springer-Verlag, Berlin/Heidelberg, pp. 333-348.

Abstract: The wood fern *Polypodium vulgare* L. (Fig. 1) belongs to the family Poly-podiaceae, and is the most widely distributed fern species in the world, appearing mostly in warm, humid forests (Bonnier 1934). The name *Polypodium* arises from the particular shape of its rhizomes, branching like feet. The plant size ranges from 10 to 50 cm; the fronds have a very long petiole and the blade is deeply divided in 20 to 40 alternate lobules. The large sori are present almost through entire year and are placed on two parallel lines among the median vein on the underside of the lobes. The peduncled sporangia open transversely throughout a longitudinal ring called the annulus (Strasburger et al. 1983). It is a very popular fern in Europe and some purgative and vermifugal properties are attributed to its rhizomes (Volák et al. 1990).

Miladera K., Saatov Z., Kholodova Y.D., Gorovits M.B., Shashkov A.S. and Abubakirov N.K. (1992) Phytoecdysteroids of plants of the genus *Serratula*. Ajugasterone C 20,22-monoacetonide from *Serratula wolffii*. *Khimiya Prirodnykh Soedinenii* (1), 71-76 [in Russian]/*Chemistry of Natural Compounds* **28**, 59-63 [in English].

Abstract: The phytoecdysteroids of the epigeal part of the plant *Serratula wolffii* Andrae have been investigated. The structure of a new ecdysteroid — ajugasterone C 20,22-monoacetonide has been established. Seven known ecdysteroids have been isolated and identified.

Miliauskas G., van Beek T.A., de Waard P., Venskutonis R.P. and Sudhölter E.J.R. (2005) Identification of radical scavenging compounds in *Rhaponticum carthamoides* by means of LC-DAD-SPE-NMR. *Journal of Natural Products* **68**(2), 168-172.

Abstract: A hyphenated LC-DAD-SPE-NMR setup in combination with on-line radical scavenging detection has been applied for the identification of radical scavenging compounds in extracts of *Rhaponticum carthamoides*. After NMR measurements, the pure compounds were infused into a mass spectrometer. The technique enabled selective detection and identification of individual radical scavenging compounds without any prior off-line chromatographic steps. Seven compounds, namely, quercetagenin-7-beta-glucopyranoside (1), quercetagenin-7-(6"-acetyl-beta-glucopyranoside) (3), 6-hydroxykaempferol-7-beta-glucopyranoside (2), 6-methoxykaempferol-3-beta-glucopyranoside (4), 6-hydroxykaempferol-7-(6"-acetyl-beta-glucopyranoside) (5), chlorogenic acid (6), and beta-

ecdysone (7), were identified in ethanol or aqueous extracts. Compound 5 is a new natural compound. Its radical scavenging activity was tested against DPPH radical and was found to be weaker than that of the reference antioxidants rosmarinic acid and Trolox.

Miller R.W., Clardy J., Kozłowski J., Mikolajczak K.L., Plattner R.D., Powell R.G., Smith C.R., Weisleder D and Zheng Q.-T. (1985). *Planta Medica* **46** (1), 40- 42.

Abstract: Five phytoecdysteroids were isolated from the seeds of *Diploclisia glaucescens* and identified by spectrometric methods (¹H-NMR, ¹³C-NMR and CIMS). One of them, 24-*epi*-makisterone A, has not been reported before. The other four - 20-hydroxyecdysone, makisterone A, 24(28)-dehydromakisterone A and pterosterone - were known previously. The ¹³C-NMR spectrum of 24(28)-dehydromakisterone A is presented for the first time.

Mohammed R., El-Hawary S.S. and Abo-youssef A.M. (2012) Biological investigation of some wild Aizoaceae and Chenopodiaceae species growing in Egypt. *Journal of Natural Products* **5**, 193-206.

Abstract: Six plants belonging to Aizoaceae and Chenopodiaceae families from Egypt were investigated for their potential use as antimicrobials against several Gram positive, Gram negative bacteria and fungi. The n-hexane extract of the tested plants showed inhibition zone of 27-22mm against *Bacillus subtilis*, *Staphylococcus aureus*. None of the tested extracts showed any activity against *Pseudomonas aeruginosa* or *Botrytis cinerea*. The polar extracts did not show any remarkable inhibition for the tested microorganisms. The nonpolar and polar extracts of *Atriplex lindleyi* Moq. susp. inflata Fam. Chenopodiaceae were further phytochemically screened for their secondary metabolites. Eight compounds were isolated from the whole plant. Compounds 2, 3, 4 and 8 are first reported from *Atriplex lindleyi*. The polar fractions of *Atriplex lindleyi* Moq. susp. inflata significantly decreased fasting blood glucose level to 93.33±10.43 and 94.60±8.55mg/dl as compared to the diabetic control value. The screening of crude extracts obtained from some Aizoaceae and Chenopodiaceae species growing in Egypt has shown that some of them were potentially rich sources for antifungal, antibacterial and antidiabetic agents.

Moriyama H. and Nakanishi K. (1968) Insect hormones, VI. Confirmation of the skeletal structure of ponasterone A. *Tetrahedron Letters* (9), 1111-1112.

No Abstract.

Mu H., Li H., Chen J. and Li P. (2014) Simultaneous determination of three steroids in *Achyranthes bidentata* Radix by RP-HPLC. *Journal of China Pharmaceutical University* (2), 210-212 [in Chinese].

Abstract: An HPLC method was established for the simultaneous determination of β-ecdysterone, R-inokosterone and S-inokosterone in the *Achyranthes bidentata* Radix to compare the quality from different habitats. The three phytoecdysones were separated with a Venusil XBP C18 column by isocratic elution using 0.1% formic acid in water and acetonitrile (84 : 16) as the mobile phase, while the UV detector wavelength was set at 250 nm. Calibration curves of β-ecdysterone, R-inokosterone and S-inokosterone were linear over the range of 3.07-1470.00, 0.52-246.00, and 0.47-225.00 μg/mL, respectively. Average recoveries were 101.4%, 101.1% and 101.1%, respectively. The established method will be helpful in the quality control of *Achyranthes bidentata* Radix.

Mu L., Yang S.-c., Guan D.-j., Yang T., Wen G.-s. and Zhang W.-m. (2011) A study on accumulation regularity of content of β-ecdysone and optimal harvest time in *Cyanotis arachnoidea* C.B. Clarke. *Journal of Yunnan Agricultural University (Natural Science)* (2), 194-198 + 204

Muchate N.S., Kadam N.S., Rajurkar N.S. and Nikam T.D. (2017) High-performance thin-layer chromatography and indirect TLC-HRMS-based determination of 20-hydroxyecdysone in *Sesuvium portulacastrum*. *Journal of Planar Chromatography* **30** (3), 193-198.

Abstract: 20-Hydroxyecdysone (20E), an imperative phytoecdysteroid, regulates several biochemical and physiological processes during the different developmental stages in insects. The aim of the present work was to validate an analytical method for the detection and identification of the 20E content in *Sesuvium portulacastrum* using high-performance thin-layer chromatography (HPTLC) combined with image analysis and using high-resolution mass spectrometry (HRMS). Using HPTLC, the better separation of 20E was achieved on TLC plate prewashed with ethanol—ethyl acetate—water (8:2:0.5) and using the solvent system consisting of chloroform—methanol—benzene (12.5:2.5:1.5) at R_F value of 0.30. The bands were documented using a TLC visualizer and scanned at 254 nm in absorption mode. The method was validated for specificity, linearity, precision, robustness, stability, accuracy, limit of detection (LOD), and limit of quantification (LOQ) as per the International Conference on Harmonization (ICH) guidelines. The linear calibration curve was achieved in the range of 50–500 ng band⁻¹ with the most significant correlation coefficient (*r* = 0.997). The relative standard deviations for intra-day and inter-day precisions were found to be 1.897 and 3.125 at 300 ng band⁻¹, and the limit of detection and limit of quantification were 4.67 and 14.16 ng band⁻¹, respectively. Furthermore, 20-hydroxyecdysone isolated from standard and plant sample bands was identified using indirect TLC—HRMS analysis. The HPTLC method and the indirect

TLC—HRMS analysis were observed to be simple, precise, accurate, and suitable for the determination of 20E in *S. portulacastrum*. In the phytopharmaceutical industry, the present method can be applied for the screening and routine quality control of 20E in plant materials and formulations.

Muchate N.S., Rajurkar N.S., Suprasanna P. and Nikam T.D. (2018) Evaluation of *Spinacia oleracea* (L.) for phytodesalination and augmented production of bioactive metabolite, 20-hydroxyecdysone. *International Journal of Phytoremediation* 20(10), 981-994.

Abstract: In this study, adaptive features of *Spinacia oleracea* to different levels of salinity, its use in desalination and production of 20-Hydroxyecdysone were studied. Plants showed survival up to EC 12 dS/m with reduced growth as compared with control. Net photosynthesis rate, transpiration, stomatal conductance, and water use efficiency of salt treated plants declines with increasing salinity stress. Higher antioxidant enzyme activities and compatible solutes accumulation were observed in salt treated plants as function of osmotic adjustment. Significant Na C sequestration and Na/K ratio were noted with increase in salt stress in comparison to the control. Since the plant accumulates a bioactive, secondary metabolite 20-Hydroxyecdysone (20E), we observed significant 20E content in plants grown at EC 4-12 dS/m in comparison to control. Furthermore, a preliminary field experiment, showed significant reduction in the soil electrical conductivity by 1.8 ds/m after 90 days of plant growth with Na C sequestration in plant biomass. Subsequent to this growth period, the phytodesalinated soil supported the significant growth of a glycophyte (rice). Our results suggest that *S. oleracea* can adapt to saline conditions with antioxidant defense and osmotic adjustment. The plant can be used as a potential candidate for desalination and also for enhanced production of 20-Hydroxyecdysone.

Muchate N.S., Rajurkar N.S., Suprasanna P. and Nikam T.D. (2019) NaCl induced salt adaptive changes and enhanced accumulation of 20-hydroxyecdysone in the in vitro shoot cultures of *Spinacia oleracea* (L.). *Scientific Reports* 9, 12522 (<https://doi.org/10.1038/s41598-019-48737-6>).

Abstract: Spinach (*Spinacia oleracea* L.) is a vegetable plant with high nutritional properties. In the present work, we studied responses of in vitro shoot cultures to salt stress (0 (control), 100, 200 and 300 mM NaCl) and salt stress-induced accumulation of 20-hydroxyecdysone (20E). Our results revealed that effect of low to moderate level of salinity stress (100-200 mM) was less pronounced on growth and tissue water content (TWC) of shoot cultures compared to higher salinity level (300 mM). The salt treated shoot cultures showed better osmotic adjustment in terms of significant accumulation of compatible solutes and total soluble sugars and also higher antioxidant enzyme activity. As the NaCl stress was increased, there was a corresponding linear raise in the Na⁺ accumulation while the contents of both K⁺ and Ca²⁺ decreased significantly. We also studied salt-stress induced accumulation of a bioactive compound; 20E and results showed that 200 mM salt treated shoot cultures accumulated significantly 2.9 fold higher 20E as compared to untreated shoot cultures. The results suggest that *Spinacia oleracea* exhibits considerable salt tolerance with better osmotic adjustment and can be considered a suitable candidate for the production of bioactive secondary metabolite.

Murakami T., Wada H., Tanaka N., Yamagishi T., Saiki Y. and Chen C-M. (1980) Chemische und chemotaxonomische Untersuchungen von Filices. XXXII. Chemische Untersuchungen der Inhaltstoffe von *Plenasium banksiifolium* (Pr.) Pr. *Chemical and Pharmaceutical Bulletin* 28(10), 3137-3139.

Abstract: The fronds of *Plenasium banksiifolium* (PR.) PR. contain stigmastan-3 β , 5 α -diol-6-one and its C28-homolog, which are previously unreported in nature, together with the known compounds, ecdysone, ecdysterone, astragalin, stigmastan-3 β , 5 α , 6 β -triol and its C28-homolog.

Munkhjargal N. (2013) Study of seasonal dynamics of phytoecdysteroids in *Silene repens*. *Mongolian Medical Science* 164 (2), 98-100.

Muzashvili T.C. and Kemertelidze E.P. (2015) Phytochemical investigation of *Helleborus caucasicus* seeds and flowers. Abstract for the International Scientific and Practical Conference: 'Achievements and Prospects for the Development of Phytochemistry', Karaganda, Kazakhstan, April 10-11th, 2015, p. 111

Nagakari M., Kushiro T., Matsumoto T., Tanaka N., Kakinuma K. and Fujimoto Y. (1994a) Incorporation of acetate and cholesterol into 20-hydroxyecdysone by hairy root clone of *Ajuga reptans* var. *atropurpurea*. *Phytochemistry* 36(4), 907-910.

Abstract: [2-¹³C]Acetate and [26,27-¹³C₂]cholesterol were incubated with a hairy root culture of *Ajuga reptans* var. *atropurpurea*. The C¹³ NMR analysis of the biosynthesized 20-hydroxyecdysone revealed that both substrates were incorporated into the phytoecdysone in an appreciable yield. In contrast, acetate, but not cholesterol, was incorporated into cyasterone and 29-norcyasterone. These results have demonstrated that this plant tissue culture is an excellent experimental system for the study of ecdysteroid biosynthesis

Nagakari M., Kushiuro T., Yagi T., Tanaka N., Matsumoto T., Kakinuma K and Fujimoto Y. (1994b) 3 β -Hydroxy-5 β -cholest-7-en-6-one as an intermediate of 20-hydroxyecdysone biosynthesis in a hairy root culture of *Ajuga reptans* var. *atropurpurea*. Journal of the Chemical Society, Chemical Communications 1761-1762.

Abstract: [3 α -2H]-, [4 α -2H]- and [4 β -2H]-Cholesterols and [3 α -2H]- and [5-2H]-3 β -hydroxy-5 β -cholest-7-en-6-ones were converted with a hairy root culture of *Ajuga reptans* var. *atropurpurea* into 20-hydroxyecdysone, in which the deuterium atoms retained their original positions, thus strongly suggesting that 3 β -hydroxy-5 β -cholest-7-en-6-one is an obligatory int...

Nair K.S., Babu C.M., Trivedy K. and Chinya P.K. (2010) Ecdysteroid extract from common catchfly *Silene gallica* L. for rearing management of silkworm, *Bombyx mori* L. and stabilized cocoon crop. Journal of Biopesticides 3 (1 Special Issue), 217-221.

Abstract: The cocoon spinning process takes 24 to 72 h depending on the seasons as the maturation process in a silkworm colony is not uniform. To hasten the larval maturation process and to shorten the mounting duration, administration of phytoecdysteroid was planned. 20-hydroxyecdysone (20E) equivalent was extracted through a Soxhlet apparatus from the dry powder of high altitude herb, *Silene gallica* known as common catchfly, partially purified through liquid partitioning and thin layer chromatography and the active principle was quantified through HPLC using pure 20E as reference. The active principle was made to a concentration of 25 ppm in water and administered to silkworm at the onset of maturation process. In winter months, about 80 % of the treated silkworms were ripe by 24 hours whereas it took 48 h for 80 % maturation in control. In rainy season > 80 % of treated larvae took about 18 hours for ripening while that in control took 30 h. In summer though, the threshold of 80 % maturation reached by 18 h in treated and by 36 h in control larvae. Though phytoecdysteroid is a known and potential biopesticide, it is used in the present work as a beneficial compound. The physiological and economical implications of such a use are discussed.

Najafi B., Sadati S.N. and Khanavi M. (2015) Ajugalide E, an phytoecdysteroid from larvicidal fraction of *Ajuga chamaecistus* ssp. *tomentella* on malaria vector *Anopheles stephensi*. Iranian Journal of Pharmaceutical Sciences 11(1) 7-8.

Abstract:

Introduction: The genus *Ajuga*, belongs to Lamiaceae family, is one of the exclusive subspecies in the flora of Iran. The plants of this genus are used traditionally for treatment of joints pain, gout, jaundice, and as insecticide. According to the previous results, among different fractions of *Ajuga chamaecistus* subsp *tomentella* (Boiss) Rech. F, hexane fraction showed the most larvicidal activity against malaria vector *Anopheles stephensi*

Methods: The hexane fraction of methanolic extract (80%) was chromatographed on silica gel and RP-C18 columns using different solvent systems to give compound 1. ¹³C-, ¹H-NMR, and IR spectroscopic methods were employed for identification of the isolated compound.

Results: The structure of compound 1, the main phytoecdysteroid separated from hexane fraction, was determined to be ajugalide-E.

Conclusion: The results of this study suggest that ajugalide-E is the main constituents of the hexane fraction of *Ajuga chamaecistus* ssp. *tomentella*.

Nakagawa T, Hara N and Fujimoto Y (1997) Biosynthesis of 20-hydroxyecdysone in *Ajuga* hairy roots: stereochemistry of C-25 hydroxylation. Tetrahedron Letters 38(15), 2701-2704.

Abstract: Feeding of [¹³C₂]acetate to hairy roots of *Ajuga reptans* var. *atropurpurea* followed by ¹³C-NMR analysis of the biosynthesized cholesterol and 20-hydroxyecdysone indicated that C-25 hydroxylation in 20-hydroxyecdysone biosynthesis proceeds both in retention and inversion mechanisms. Feeding studies of [26-¹³C]- and [27-¹³C]cholesterols established that the ratio of retention and inversion mechanisms is ca. 3:1.

Nakamura O., Mimaki Y., Sashida Y., Nikaido T. and Ohmoto T. (1994) Three new furostanol saponins from the bulbs of *Ipheion uniflorum*. Chemical and Pharmaceutical Bulletin 42(5), 1116-1112.

Abstract: Phytochemical screening of the bulbs of *Ipheion uniflorum* (Liliaceae) has led to the isolation of three new furostanol saponins (2-4) along with a known phytoecdysteroid, ecdysterone (1). The structures of the new compounds were determined by two-dimensional NMR techniques, 1H-1H correlation spectroscopy (COSY), homonuclear Hartmann-Hahn (HOHAHA), phase-sensitive nuclear Overhauser effect spectroscopy (NOESY), heteronuclear multiple quantum coherence (HMQC) and heteronuclear multiple-bond correlation (HMBC) spectra, and hydrolysis to be 3 beta-hydroxy-22 alpha-methoxy-26-O-beta-D- glucopyranosyloxy-5 alpha-furost-25(27)-en-2-one 3-O-[O-alpha-L-rhamnopyranosyl-(1-->2)-O-[O-alpha-L-arabinopyranosyl+ +- (1-->2)-O-[beta-D-xylopyranosyl-(1-->3)]-beta-D-glucopyranosyl-(1-->4)]- beta-D-galactopyranoside) (2) and its 25(27)-dihydro derivatives (3: 25S; 4: 25R). The inhibitory activity exhibited by compounds 1-4 and the corresponding spirostanol saponins (2a and 3a) of 2 and 3 on cAMP phosphodiesterase was assayed as a primary screening test to identify new compounds with medicinal potential.

Nakamura S., Chen G., Nakashima S., Matsuda H., Pei Y., and Yoshikawa M. (2010) Brazilian Natural Medicines. IV. New noroleanane-type triterpene and ecdysterone-type sterol glycosides and melanogenesis inhibitors from the roots of *Pfaffia glomerata*. Chemical and Pharmaceutical Bulletin. 58(5) 690-695.

Abstract: The ethyl acetate and 1-butanol soluble fractions of the roots of *Pfaffia glomerata* were found to show inhibitory effects on melanogenesis in theophylline-stimulated B16 melanoma 4A5 cells. From the ethyl acetate and 1-butanol soluble fractions, we isolated a new noroleanane-type triterpene, pfaffianol A, its glycosides, pfaffiaglycosides A and B, and ecdysterone-type sterol glycosides, pfaffiaglycosides C, D, and E, together with eight known constituents. The structures of new constituents were determined on the basis of physicochemical and chemical evidence. Among them, pfaffianol A (IC₅₀=44 microM) and pfaffoside C (IC₅₀=92 microM) substantially inhibited melanogenesis without cytotoxic effects. The inhibitory effects were stronger than that of reference compound, arbutin (IC₅₀=174 microM).

Nakanishi K. (1969) Ponasterones, compounds with moulting hormone activity. Bulletin de la Société Chimique de la France (10), 3475-3485.

Nakanishi K., Koreeda M., Sasaki S., Chang M.L. and Hsu H.Y. (1966) Insect hormones. The structure of ponasterone A, an insect-moulting hormone from the leaves of *Podocarpus nakaii* Hay. Journal of the Chemical Society, Chemical Communications (24), 915-917.

No Abstract.

Nakanishi K., Koreeda M., Chang M.L. and Hsu H.Y. (1968) Insect hormones, V. The structures of ponasterones B and C. Tetrahedron Letters (9), 1105-1110.

No Abstract.

Nakano K., Murakami K., Nohara T., Tomimatsu T. and Kawasaki T. (1981) The constituents of *Paris verticillata*, M.v. Bieb. Chemical and Pharmaceutical Bulletin 29, 1445-1451.

Abstract: Nine compounds (1-9) have been isolated from the whole plants of *Paris verticillata* M. v. BIEB. (Liliaceae) and their structures characterized. They can be divided into four groups ; phytosteryl derivatives (1, phytosteryl (6'-palmitoyl)- β -D-glucopyranoside ; 2, the despalmitate of 1), phytoecdysones (3, ecdysone ; 4, ajugasterone A ; 5, ecdysterone), pennogenin glycosides (6, pennogenin tetraglycoside (T-g) ; 7, prototype glycoside of 6), and kaempferol glycosides (8, kaempferol 3-O- α -L-rhamnopyranosyl-(1 \rightarrow 2)- β -D-glucopyranoside ; 9, 7-O- β -D-glucopyranosyl kaempferol 3-O- α -L-rhamnopyranosyl-(1 \rightarrow 2)- β -D-glucopyranoside)

Nakano K., Nohara T., Tomimatsu T. and Nishikawa M. (1982) A phytoecdysteroid, taxisterone, from *Taxus cuspidata*. Phytochemistry 21(11), 2749-2751.

Namuna A. (2019) Bioecological characteristics and natural resources of *Rhaponticum integrifolium* in Uzbekistan. American Journal of Plant Sciences 10, 850-865.

Abstract: The large life cycle *Rhaponticum integrifolium* in natural populations (Qashqadaryo, Uzbekistan) was studied. Duration of before generative, generative and senile periods of *Rhaponticum integrifolium* is 2 - 5, 8 - 10 and 2 - 4 years respectively. The beginning of phenophases depends on the height of the terrain above sea level: raising the terrain from 400 to 1500 m leads to a delay in the onset of phenophases. Age states of *Rhaponticum integrifolium* in different ecological and phylogenetic conditions were revealed. Updated distribution of detected participation of vegetation and the raw *Rhaponticum integrifolium* in Uzbekistan, describes the dynamics of the contents ecdysteroids in the periods of vegetation. At the same time, a map of the species area in Central Asia was made and the term of its phytomass recovery under different operating modes of thickets was determined.

Nawrot J., Budzianowski J., Nowak G., Micek I., Budzianowaka A. and Gornowicz-Porowska J. (2021) Biologically active compounds in *Stizolophus balsamita* inflorescences: isolation, phytochemical characterization and effects on the skin biophysical parameters. International Journal of Molecular Sciences 22, 4428, pp. 16 (doi: 10.3390/ijms22094428).

Abstract: Three germacranolides, as well as five flavonoids, natural steroid and simple phenolic compounds, were isolated from the inflorescence of *Stizolophus balsamita* growing in Iran. The paper presents active compounds found for the first time in the inflorescence of this species. The flavonoids, simple phenolic compounds and natural steroids have been isolated for the first time in the genus *Stizolophus*. The MTT assay was employed to study in vitro cytotoxic effects of the taxifolin against human fibroblasts. We also evaluate the possible biological properties/cosmetic effects of *Stizolophus balsamita* extract and taxifolin on the human skin. Sixty healthy Caucasian adult females with no dermatological diseases were investigated. We evaluate the effects of *S. balsamita* extract and taxifolin on skin hydration and transepidermal water loss (TEWL). It was revealed that *S. balsamita* extract might

decrease TEWL level and fixed the barrier function of the epidermis. The presence of bioactive phytochemical constituents in *S. balsamita* inflorescences makes them a valuable and safe source for creating new cosmetics and medicines.

Nejma A.B., Ngair A., Jannet H.B., Hamza M.A., Daich A., Othman M. and Lawson A.M. (2015) New septanoside and 20-hydroxyecdysone septanoside derivative from *Atriplex portulacoides* roots with preliminary biological activities. *Bioorganic & Medicinal Chemistry Letters* **25** 1665–1670

Abstract: *Atriplex portulacoides* is a perennial, shrubby halophytic plant. Widespread in salt marshes along the coasts of Europe, North Africa and SouthWest Asia, it belongs to Chenopodiaceae family (goosefoot family), that is largely distributed throughout the world especially in arid and saline regions. 1 This family is consisted of 104 genus and more than 1400 species, the majority growing naturally in saline soils. 2 Different species of *Atriplex* (*A. hortensis*, *A. fruticosa*, *A. inflata*, *A. parvifolia*, *A. semibaccata*, *A. undulata*, *A. vestita*) including *A. portulacoides* were reported as sources of interesting biological effects (antifungal, antiviral, antioxidant, cytotoxic, antimicrobial, etc.) through their extracts or their chemical constituents. 3 These literature data prompted us to choose the species *A. portulacoides* to contribute to its phytochemical and biological studies. The phytochemical investigation of this plant led to the isolation of two new compounds designated as portulasoid (2) and septanoecdysone (3) along with the known 20-hydroxyecdysone (20HE) (1). Their chemical structures were elucidated on the basis of extensive spectroscopic methods including ES-HRMS, 1D and 2D-NMR. The isolated compounds were finally tested for their antioxidant activity by using DPPH • , ABTS +• and Fe 3+ and also for their antibacterial and anticholinesterase activities.

Nguyen T.K.T., Ninh K.B., Do T.T., Tran M.L., Vu H.G., Nguyen X.N. and Phan V.K. (2018) Ecdysteroids from leaves of *Vitex trifolia*. *Vietnam Journal of Chemistry* **56**(2), 162-166.

Abstract: Four known ecdysteroids, ecdysone (1), 20-hydroxyecdysone (2), 20-hydroxyecdysone 2,3-monoacetonide (3), and turkesterone (4) were isolated from leaves of *Vitex trifolia*. The structure of these compounds was elucidated by means of 1D- and 2D-NMR spectra and was compared with those reported in literature. Compound 3 was reported from *Vitex* genus for the first time; compounds 1, 2, and 4 from *V. trifolia* for the first time.

Nie R.L. and Qiu M.H. (1987) The phytoecdysones in residue of mother liquid of the moulting hormone produced in factory from *Cyanotis arachnoidea*. *Acta Botanica Yunnanica* **9**(2), 253-256 [in Chinese].

Nie R. and Yue Y. (1983) The dermination of β -ecdysone from cultivated *Cyanotis arachnoidea*. *Acta Botanica Yunnanica* **5**(3), 317-318 [in Chinese].

Nien S-l., Hsu H-y., Ho M. and Yo Y-c. (1978) The isolation and identification of phytoecdysones from *Cyanotis arachnoidea*. *Acta Chimica Sinica* **36**(5), 137-141 [in Chinese, with an English abstract].

Abstract: Two insect moulting hormones isolated from *Cyanotis arachnoidea* C.B.Clarke. (*Commelmaceae*) were identified as β -ecdysone and its 2-acetate. yields of phytoecdysones were 1.2%(from whole dry herbs) and 2.9%(dry roots) respectively.

Nikolaeva I.G., Tsybiktarova L.P., Garmaeva L.L., Nikolaeva G.G., Olennikov D.N. and Matkhanov I.E. (2017) Determination of ecdysteroids in *Fornicium uniflorum* (L.) and *Serratula centauroides* (L.) raw materials by chromatography-UV spectrophotometry. *Journal of Analytical Chemistry* **72**(8), 854-861 [in English]/*Zhurnal Analiticheskoi* **72**(8), 733-741 [in Russian].

Abstract: A chromatography–spectrophotometry method is developed for determining total ecdysteroids in terms of 20-hydroxyecdysone in *Serratula centauroides* (L.) and *Fornicium uniflorum* (L.) raw materials, based on the purification of extracts from the plant raw materials with aluminum oxide and subsequent spectrophotometric determination relative to a 20-hydroxyecdysone reference sample. Ecdysteroids were determined in plants by HPLC; rhapontisterone and turkesterone are found in *F. uniflorum* for the first time.

Nishimoto N., Shiobara I Y., Inoue I S-s., Fujino I M., Takemoto T., Yeoh C.L. and Hashimoto G. (1986) Ecdysterone from *Pfaffia tuberosa* (Spreng.) Hicken. *Revista Brasileira Farmacognosia* **1**(2) 188-191.

Abstract: In this paper, we wish to report the isolation and identification of ecdysterone from *Pfaffia tuberosa* (Spreng.) Hicken. The roots of *Pfaffia tuberosa* (Spreng.) Hicken were collected in the Mato Grosso State - Brazil. The dried and sliced roots (197g) were extracted with hot MeOH. A suspension of the resulting MeOH extract in the water was treated with n-BuOH. The n-BuOH layer was concentrated in vacuo to give a brown viscous oil (5g), which was chromatographed on silicagel (65g) using CHCl₃ - MeOH - H₂O (80:20:10) lower phase, and 15ml fractions were collected. Fractions 11-13 were concentrated and the residual solid was recrystallized from EtOH to afford oleanolic acid (65mg). The crystalline residue obtained from fractions 44-56 was recrystallized from EtOAc-MeOH

to give ecdysterone (87mg). Each compound was identified by direct comparison with an authentic sample. Further HPLC quantitative analysis showed lower content of ecdysterone (0,23%) compared with that of *Pfaffia iresinoides* Spreng.

Nishimoto N., Shiobara Y., Fujino M., Inoue S-S., Takemoto T., de Oliveira F., Akisue G., Akisue M.K., Hashimoto G., Tanaka O., Kasai R. and Matsuura H. (1987) Ecdysteroids from *Pfaffia iresinoides* and reassignment of some ¹³C NMR chemical shifts. *Phytochemistry* 26, 2505-2507.

Abstract: From the crude drug 'Brazil ginseng', the roots of *Pfaffia iresinoides*, a large amount of ecdysterone has been isolated together with polypodine B and pterosterone. The ecdysterone content of each part of the plant was determined by high pressure liquid chromatography quantitative analysis. A triterpenoid saponin, chikusetsusaponin IVa has also been isolated.

Nishimoto N., Shiobara Y., Inoue S-S., Fujino M., Takemoto T., Yeoh C.L., de Oliveira F., Akisue G., Akisue M.K. and Hashimoto G. (1988) Three ecdysteroid glycosides from *Pfaffia iresinoides*. *Phytochemistry* 27(6), 1665-1668.

Nohara T., Ito Y., Seike H., Komori T., Moriyama M., Gomita Y. and Kawasaki T. (1982) Study on the constituents of *Paris quadrifolia* L. *Chemical and Pharmaceutical Bulletin* 30(5), 1851-1856.

Abstract: Seven compounds have been isolated from whole plants of *Paris quadrifolia* L. and their chemical structures characterized as follows ; pennogenin (1), pennogenin 3-O- α -L-rhamnopyranosyl-(1 \rightarrow 2)-[α -L-rhamnopyranosyl-(1 \rightarrow 4)]- β -D-glucopyranoside (2), pennogenin 3-O- α -L-rhamnopyranosyl-(1 \rightarrow 4)- α -L-rhamnopyranosyl-(1 \rightarrow 4)-[α -L-rhamno-pyranosyl-(1 \rightarrow 2)]- β -D-glucopyranoside (3), 22-hydroxyfurostanol 3, 26-O-bisglycoside corresponding to 3 (4), 1-dehydrotrillenogenin (5), ecdysterone (10) and kaempferol 3-O- β -D-glucopyranosyl-(1 \rightarrow 4)- β -D-glucopyranoside (11). In addition, a stimulative effect of 3, one of the major components, on isolated bull frog heart was observed.

Nomura K, Fujimoto Y (2000) Mechanism of C-2 hydroxylation during the biosynthesis of 20-hydroxyecdysone in *Ajuga hairy* roots. *Chemical and Pharmaceutical Bulletin* 48(3), 344-348.

Abstract: Feeding synthetic [2 β -²H]- and [2 α -²H]-cholesterols to the hairy roots of *Ajuga reptans* var. *atropurpurea* and ²H-NMR analysis of the biosynthesized 20-hydroxyecdysone revealed that hydroxylation at C-2 proceeds with retention of configuration. Feeding [2 α , 3 α -²H-₂]cholesterol followed by ²H-NMR analysis of the 2, 3, 22-triacetate of the resulting 20-hydroxyecdysone ruled out a mechanism which involves a partial loss of the 2 α -hydrogen. The steric course of C-2 hydroxylation in *Ajuga hairy* roots is identical with that reported in the insect, *Schistocerca gregaria*.

Nomura K., Nakamura A., Fujimoto Y., Yamada J., Takahashi K and Morisaki M. (2000) Clerosterol as a common sterol precursor of cyasterone and related ecdysteroids in *Ajuga hairy* roots. 2000 International Chemical Congress of Pacific Basin Societies, Honolulu, Hawaii, abstract 377.

Novoselskaya N.L., Gorovits M.B. and Abubakirov N.K. (1975) Phytoecdysones of *Serratula*. IV. Sogdisterone. *Khimiya Prirodnikh Soedinenii* 429-430 [in Russian].

Novoselskaya I.L., Gorovits M.B. and Abubakirov N.K. (1981a) Ecdysterone and polypodine B from *Paris quadrifolia*. *Khimiya Prirodnikh Soedinenii* (3), 402-403 [in Russian].

Novoselskaya I.L., Gorovits M.B. and Abubakirov N.K. (1981b) Phytoecdysteroids from *Serratula coronata*. *Khimiya Prirodnikh Soedinenii* 668-669 [in Russian].

Novozhilova E., Rybin V., Gorovoy P., Gavrilenko I., Doudkin R. (2014) Phytoecdysteroids in aerial parts of the Far Eastern species of *Caryophyllaceae*. *Turczaninowa* 17(2), 42-48 [in Russian, with an abstract in English].

Abstract: Distribution of integristerone A, 20-hydroxyecdysone, ecdysone, 2-deoxy-20-hydroxyecdysone has been analyzed during blossom phase in East Asian species of *Lychnis*, *Silene*, *Melandrium* and *Sagina* (*Caryophyllaceae*). The prevailing compound in a blossoming phase at *Lychnis* and *Silene* is 20-hydroxyecdysone. The maximum contents of the 20-hydroxyecdysone in investigated species is observed in the flowers.

Novozhilova E., Rybin V., Gorovoy P., Gavrilenko I., Doudkin R. (2015) Phytoecdysteroids of the East Asian *Caryophyllaceae*. *Pharmacognosy Magazine* 11(42, Supplement 1) S225-S230.

Abstract: Occurrence of integristerone A (1), 20-hydroxyecdysone (2), ecdysone (3), 2-deoxy-20-hydroxyecdysone (4) has been analyzed in 64 species of the East Asian *Caryophyllaceae*. Ecdysteroid content was determinate by high-performance liquid chromatography (HPLC). HPLC with a high-resolution mass spectrometry was performed

on Shimadzu LCMS-IT-TOF (Japan) system equipped with a LC-20A Prominence liquid chromatograph, a photodiode array detector SPD-M20A and ion-trap/time-of-flight mass spectrometer. New sources of phytoecdysteroids: *Melandrium sachalinense* and *Melandrium firmum* have been revealed. It is the 1(st) time that two has been identified in *M. sachalinense* and *M. firmum*; 1 in the species: *Lychnis fulgens*, *Silene repens*, *Silene foliosa*, *Silene stenophylla*, *Silene jensseensis* and *M. sachalinense*; 3 in *Lychnis cognata*; 4 in *L. fulgens*, *S. stenophylla* and *S. jensseensis* (the tribe Lychnideae, the subfamily Caryophylloideae). Ecdysteroid-negative taxa are *Spergularia rubra* of the tribe Sperguleae; species of the genera *Minuartia*, *Honckenya*, *Eremogone*, *Arenaria*, *Moehringia*, *Pseudostellaria*, *Fimbripetalum*, *Stellaria* and *Cerastium* of the tribe Alsineae; *Scleranthus annuus* of the tribe Scleranthae, as well as the East Asian representatives of the genera *Gypsophila*, *Psammophilola*, *Dianthus* and *Saponaria* of the tribe Diantheae; *Oberna* and *Agrostemma* of the tribe Lychnideae. This investigation shows the most promising sources of ecdysteroids are species of genera *Silene* and *Lychnis*.

Nowak G., Urbanska M., Nawrot J., Bernard M.K. and Dawid-Pac R. (2013) Color and chemical reactions of selected sesquiterpene lactones and ecdysones from Asteraceae on TLC plates. *Journal of Planar Chromatography* 26(3), 289-293.

Abstract: The significance of sesquiterpene lactones occurring in the Asteraceae family is well known. These compounds are thought to be useful chemotaxonomic markers, especially in genera and species of Centaureinae subtribe. A relatively simple and fast technique like the comparative thin-layer chromatography of plant extracts would be very helpful for establishing the correct natural division of subtribe. Furthermore, some sesquiterpene lactones have improved pharmacological activities, useful for instance in the treatment of inflammatory states (helenalin in *Arnicae anthodium*, matricin in *Chamomillae anthodium*), migraine (parthenolide in *Parthenii folium*), and digestive ailments (cnicin in *Cnici benedicti herba*, cynaropicrin in *Cynarae folium*). Ecdysones, in turn, are used as tonic agents that decrease the cholesterol level and improve the condition of the skin. Phytosteroids of the ecdysones group: 20-hydroxyecdysone and polypodine B, often occur in the species of family Asteraceae [1].

Nsimba R.Y., Kikuzaki H. And Konishi Y. (2008) Ecdysteroids act as inhibitors of calf skin collagenase and oxidative stress. *Journal of Biochemistry and Molecular Toxicology* 22(4), 240-250.

Abstract: Three new phytoecdysteroids have been isolated from the seeds of *Chenopodium quinoa* and structurally identified as 20,26-dihydroxy, 28-methyl ecdysone, 20,26-dihydroxy, 24(28)-dehydro ecdysone, and 20-hydroxyecdysone 22-glycolate using serial chromatographic and spectroscopic methods. Both previously reported compounds and newly identified phytoecdysteroids were evaluated for their inhibitory effect on calf skin collagenase, as this enzyme is involved in aging skin diseases. Their effectiveness on scavenging 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radicals, as well as in chelating the iron metal ions was also investigated. All isolated compounds demonstrated a higher potency to inhibit this matrix metalloproteinase and to chelate the iron ion, both with respect to the number of carbonyl groups bearing their carbon skeleton, suggesting that their mechanism of action involves their ability to coordinate both ions (either the zinc ion of the catalytic domain of collagenase or the iron ion), acting as an electron donor. The DPPH-scavenging ability was hydroxyl dependent. Satisfactory results obtained from the enzyme in vitro experiment were further supported by the gel electrophoresis. These results indicate that ecdysteroids might be considered as potent chemical agents to prevent or delay both collagenase-related skin damages and oxidative stress.

Nugroho A., Park K-S., Seo D-J. and Park H-J. (2018) Identification and HPLC quantification of a phytoecdysone and three phenolic glycosides in *Lamium takesimense* Nakai. *Natural Product Science* 24(4), 241-246.

Abstract: The herbs of *Lamium takesimense* Nakai (Lamiaceae) is used to treat spasmodic and inflammatory disease. The four polar compounds, ecdysterone, isoacteoside, rutin and lamiuside C, were isolated and identified from the BuOH fraction of the *L. takesimense* MeOH extract. HPLC quantification was performed on a Capcell Pak C18 column (5 μ m, 4.6 mm \times 250 mm) with a gradient elution of H₂O and 0.05% acetic acid in MeOH. The HPLC method was validated in terms of linearity, sensitivity, stability, precision, and accuracy. The quantitative level in plant material was determined as the following order: lamiuside C (4, 3.75 mg/g dry weight) > ecdysterone (1, 1.93 mg/g) > isoacteoside (2, 1.32 mg/g) > rutin (3, 0.97 mg/g).

Ochieng C.O., Ishola I.O., Oplyo S.A., Manguro L.A.O., Owuor P.O. and Wong K-C. Wong (2013) Phytoecdysteroids from the stem bark of *Vitex donania* and their anti-inflammatory effects. *Planta Medica* 79, 52-59.

Abstract: With reference to the ethnopharmacological significance of *Vitex doniana* Sweet (Lamiaceae) leaves in the treatment of stomach and rheumatic pains as well as inflammatory disorders, biological studies on its stem bark extracts have also reported anti-inflammatory and analgesic activities, with no attempt to identify the active components. Chromatographic and spectroscopic procedures identified three new phytoecdysteroids: 21-hydroxyshidasterone (1), 11 β -hydroxy-20-deoxyshidasterone (2), and 2,3-acetonide-24-hydroxyecdysone (3) from the stem bark methanol extracts along with known ecdysteroids shidasterone (4), ajugasterone C (5), 24-hydroxyecdysone (6), and 11 β ,24-hydroxyecdysone (7). The compounds (1-7) showed significant ($p \leq 0.05$)

inhibitory effect at 100 mg/kg dose on rat paw oedema development due to carrageenan-induced inflammation in Sprague Dawley rats. These results suggest a possible contribution of ecdysteroids to the anti-inflammatory effect of some *V. doniana* stem bark extracts.

Odinokov V.N., Galyautdinov I.V., Fatykhov A.A. and Khalilov L.M. (2000) A new phytoecdysteroid. Russian Chemical Bulletin 49(11), 1923-1924 [in English]/ Izvestiya Akademii Nauk. Seriya Khimicheskaya (11), 1954-1955 [in Russian].

Abstract: A new phytoecdysteroid, viz, 2-deoxy-3-epi-4 β , 20-dihydroxyecdysone ((20R,22R)-3 α ,4 β 14 α ,20,22,25-hexahydroxy-5 β -cholest-7-en-6-one), named coronatasteronc, was isolated from *Serratula coronata* and identified by NMR spectroscopy.

Odnikov V.N., Galyautdinov I.V., Nedopekin D.V., Khalilov L.M., Shashkov A.S., Kachala V.V., Dinan L. and Lafont R. (2002) Phytoecdysteroids from the juice of *Serratula coronata* L. (Asteraceae). Insect Biochemistry and Molecular Biology 32, 161-165.

Abstract: Seven phytoecdysteroids have been isolated from *Serratula coronata* L.. One of them is a new phytoecdysteroid, 3-epi-20-hydroxyecdysone. Two further ecdysteroids, 20-hydroxyecdysone 22-acetate and taxisterone, are isolated from this species for the first time in addition to the typical *S. coronata* ecdysteroids, 20-hydroxyecdysone, ecdysone, ajugasterone C and polypodine B. The juice squeezed from aerial parts of fresh plants of *S. coronata* was extracted with ethyl acetate. The ecdysteroids were isolated by a combination of chromatographic techniques (mainly HPLC) and identified by 1D and 2D ¹H and ¹³C NMR experiments and mass-spectrometry. The biological activities of 3-epi-20-hydroxyecdysone ($EC_{50}=1.6\times 10^{-7}$ M), taxisterone ($EC_{50}=9.5\times 10^{-8}$ M) and ajugasterone C ($EC_{50}=6.2\times 10^{-8}$ M) have been determined in the *Drosophila melanogaster* BII bioassay for ecdysteroid agonist activity.

Odnikov V.N., Kumpun S., Galyautdinov I.V., Evrard-Todeschi N., Veskina N.A., Khalilov L.M., Girault J.-P., Dinan L., Maria A. and Lafont R (2005) Low-polarity phytoecdysteroids from the juice of *Serratula coronata* L. (Asteraceae). Collection of Czechoslovak Chemical Communications 70, 2038-2052.

Ogawa S., Nishimoto N., Okamoto N. and Takemoto T. (1971) Studies on the constituents of *Achyranthis radix*. VIII. The insect-moulting substances in *Achyranthes* genus (Supplement 2). Yakugaku Zasshi 91, 916-920 [in Japanese, with an English abstract].

Ogawa S., Yoshida A. and Kato R. (1977) Analytical studies on the active constituents in crude drugs III. High-speed liquid chromatographic determination of ecdysterone and inokosterone in *Achyranthis radix*. Chemical and Pharmaceutical Bulletin 25(5), 904-908.

Abstract: A simple, rapid, and accurate high-speed liquid chromatographic method was established for the determination of ecdysterone (I) and inokosterone (II) in *Achyranthis radix*. I and II, extracted from *Achyranthis radix*, were directly injected onto column and separated in approximately 15 min using a 2 m Permaphase ETH column with a mobile phase of 10% ethanol in n-hexane at 50°. A working relative standard deviation was less than 2%. The extraction methods were also investigated and it was concluded that the Soxhlet extraction with methanol was the most effective. Moreover, II was separated into two epimeric isomers on a Permaphase octadecyl silane column with a mobile phase of methanol in water.

Ohta S., Guo J-R., Hiraga Y and Suga T. (1996) 24-Epi-pterosterone: a novel phytoecdysone from the roots of *Athyrium yokoscense*. Phytochemistry 41(3), 745-747.

Abstract: A novel phytoecdysone, 24-epi-pterosterone, was isolated from a water extract of the roots of *Athyrium yokoscense* and the structure was determined to be (20R,22R,24R)-2fl,3fl,14~,20,22,24-hexahydroxy-5fl-cholest-7-en-6-one by a combination of spectroscopic methods and single-crystal X-ray analysis. 24-Epi-pterosterone is the first example of an ecdysteroid possessing a 24R-hydroxyl group.

Ohyama K., Kushiro T., Nakamura K. and Fujimoto Y. (1999) Biosynthesis of 20-hydroxyecdysone in *Ajuga hairy* roots: fate of 6 α - and 6 β -hydrogens of lathosterol. Bioorganic and Medicinal Chemistry 7, 2925-2930.

Abstract: The fate of 6 α - and 6 β -hydrogens of lathosterol during the transformation into 20-hydroxyecdysone was chased by feeding [3 α ,6 β -2H₂]- and [3 α ,6 α -2H₂]-lathosterols to hairy roots of *Ajuga reptans* var. *atropurpurea*. The behavior of 6 β -hydrogen, which mostly migrated to the C-5 position of 20-hydroxyecdysone, was in agreement with that of C-6 hydrogen of cholesterol. The results strongly supported the view that cholesterol and lathosterol are first metabolized into 7-dehydrocholesterol, which is then converted into 20-hydroxyecdysone via 7-dehydrocholesterol 5 α ,6 α -epoxide in the hairy roots.

Okuzumi K., Hara N., Fujimoto Y., Yamada J., Nakamura A., Takahashi K. and Morisaki M. (2003) Biosynthesis of phytoecdysteroids in *Ajuga hairy roots*: clerosterol as a precursor of cyasterone, isocyasterone and 29-norcyasterone. *Tetrahedron Letters* **44**(2), 323-326.

Abstract: Feeding studies of six ¹³C-labeled sterols, including clerosterol, to hairy roots of *Ajuga reptans* var. *atropurpurea* have established that clerosterol is a precursor of three phytoecdysteroids, cyasterone, isocyasterone and 29-norcyasterone.

Okuzumi K., Hara N., Uekusa H. and Fujimoto Y. (2005) Structure elucidation of cyasterone stereoisomers isolated from *Cyathula officinalis*. *Organic and Biomolecular Chemistry* **3**, 1227-1232.

Abstract: Chemical investigation of ecdysteroidal constituents of the roots and stems of *Cyathula officinalis* led to the isolation of two cyasterone stereoisomers, 2 and 3, together with the known cyasterone 1. The structures of compounds 2 and 3 were determined to be 28-epi-cyasterone and 25-epi-28-epi-cyasterone, respectively, by means of spectroscopic analysis. X-Ray structures of 1 and 2 confirmed the 24S,25S,28R configuration for 1 and 24S,25S,28S for 2.

Oleznikov D.N. (2018a) Phytoecdysteroids and flavonoids from *Gastrolychnis tristis*. *Chemistry of Natural Compounds* **54**(1), 204-206 [in English]/*Khimiya Prirodnykh Soedinenii* (1) 170-171 [in Russian].

Abstract: *Gastrolychnis tristis* (Bunge) Czerep. [syn. *Lychnis tristis* Bunge, *Melandrium triste* (Bunge) Fenzl, *Silene bungei* Bocquet] (Caryophyllaceae) is distributed in Central Asia and eastern Siberia and occurs in alpine glades and meadows in the foothills of Central Asia, Altai, and Siberia [1]. The genus *Gastrolychnis* is placed in the tribe *Lychnideae* and is probably a source of phytoecdysteroids and flavonoid C-glycosides that were observed earlier in the systematically similar genera *Lychnis* [2, 3] and *Silene* [4], including *S. uralensis* ssp. *apetala* (L.) Bocquet, which was earlier placed in the genus *Gastrolychnis* [*G. apetala* (L.) Tolm. & Kozhanch.] [5]. Information on the components of *G. tristis* is currently limited to a report on the presence of 20-hydroxyecdysone [6]. Therefore, we studied the chemistry of this species.

Oleznikov D.N. (2018b) Minor ecdysteroids from *Rhaponticum uniflorum* leaves from Eastern Siberia. *Chemistry of Natural Compounds* **54**(4), 798-800 [in English]/ *Khimiya Prirodnykh Soedinenii* (4), 674-676 [in Russian].

No Abstract.

Oleznikov D.N. (2018c) Makisterone C-20,22-acetonide from *Rhaponticum uniflorum*. *Chemistry of Natural Compounds* **54**(5), 930-933 [in English]/*Khimiya Prirodnykh Soedinenii* (5), 788-790 [in Russian].

Abstract: A new ecdysteroid that was identified using UV and NMR spectroscopy and mass spectrometry as makisterone C-20,22-acetonide (1) in addition to the known compound polygodin B-20,22-acetonide were isolated from leaves of *Rhaponticum uniflorum* (L.) DC. (Asteraceae). Flowers of *R. uniflorum* also contained 1 and seven known compounds.

Oleznikov D.N. and Kashchenko N.I. (2017) Phytoecdysteroids from *Silene jenseensis*. *Chemistry of Natural Compounds* **53**(6), 1199-1201 [in English]/*Khimiya Prirodnykh Soedinenii* (6) 1016-1017 [in Russian].

No Abstract.

Oleznikov D.N. and Kashchenko N.I. (2018) Method of the rapid analysis of 20-hydroxyecdysone content in plants and ferns using solid-phase extraction on polyamide and microcolumn HPLC-UV. *Khimiya Rastitel'nogo Sr'ya* (3), 41-52 [in Russian].

Abstract: A method for the rapid quantitative analysis of 20-hydroxyecdysone in plants was developed using the microcolumn reversed-phase HPLC with UV detection (246 nm) on the ProntoSIL-120-5-C18 column (2×75 mm) and the gradient eluent system LiClO₄/HClO₄-acetonitrile. The chromatographic stage of the analysis was 5 minutes long, which made it possible to characterize the technique as the fastest. For preliminary purification of the plant extracts, solid phase extraction on polyamide was used which led to an increase in the sensitivity of the analysis. Validation studies showed that the technique characterized by satisfactory metrological data. The values of the limit of detection (LOD) and the limit of quantification (LOQ) of 20-hydroxyecdysone were 3.3 and 10 µg/ml, respectively. The accuracy indices for various levels of 20-hydroxyecdysone content (80–120%) did not exceed 98.57–101.38%. The method applied to the analysis of 20-hydroxyecdysone in 359 species of flowering plants and 12 ferns species growing on the territory of the Buryatia Republic. The presence of 20-hydroxyecdysone was established in 22 species including 18 flowering plants and 4 ferns. The concentration levels of 20-hydroxyecdysone in plants varied from trace (*Athyrium filix-femina*, *Diplazium sibiricum*, *Pteridium aquilinum*) to very high (25.40 mg/g in *Rhaponticum uniflorum* and 25.87 mg/g in *Silene jenseensis*). The presence of 20-hydroxyecdysone was firstly detected in three species including *Gastrolychnis gracilis*, *G. saxatilis* and *Silene violascens*. The developed technique is fast, simple, sensitive and stable and can be recommended for searching purpose to evaluate the new plant sources of 20-hydroxyecdysone.

Olennikov D.N. and Kaschenko N.I. (2019a) New flavonoids and turkesterone-2-*O*-cinnamate from leaves of *Rhaponticum uniflorum*. Chemistry of Natural Compounds **55**(2), 256-264 [in English]/Khimiya Prirodnykh Soedinenii (2), 220-227 [in Russian].

Abstract: Leaves of *Rhaponticum uniflorum* (L.) DC. (Asteraceae) afforded 46 compounds including seven new flavonoids that were identified using UV, IR, and NMR spectroscopy and mass spectrometry as 6-hydroxyluteolin-7-*O*-(2'-*O*-caffeoyl)- β -D-glucopyranoside (rhaunoside A, 1), 6-hydroxyluteolin-7-*O*-(6''-*O*-cinnamoyl)- β -D-glucopyranoside (rhaunoside B, 2), 6-hydroxyluteolin-4'-*O*- β -D-glucopyranoside (rhaunoside C, 3), nepetin-7-*O*-(6''-*O*-caffeoyl)- β -D-glucopyranoside (rhaunoside D, 4), nepetin-7-*O*-(6''-*O*-cinnamoyl)- β -D-glucopyranoside (rhaunoside E, 5), nepetin-3'-*O*- β -D-glucopyranoside (rhaunoside F, 6), and luteolin-7-*O*-(2''-*O*-caffeoyl)- β -D-glucopyranoside (rhaunoside G, 7) and the new ecdysteroid turkesterone-2-*O*-cinnamate (8).

Olennikov D.N. and Kashchenko N.I. (2019b) Ecdysteroids of *Silene italica*: glycosidic and nonglycosidic components and HPLC-DAD-ESI-MS profile. Khimiya Rastitel'nogo Syr'ya (4), 135-147 [in Russian, with an English abstract].

Olennikov D.N. and Kaschenko N.I. (2019c) Phytoecdysteroids of *Serratula centauroides* herb from Cisbaikalia. Russian Journal of Bioorganic Chemistry **45**(7), 913-919.

Abstract: *Serratula centauroides* L. is a plant species of the Asteraceae family common in Cisbaiklia and containing phytoecdysteroids. As a result of chromatographic separation of the aerial part of *S. centauroides* growing in Buryatia Republic, nine compounds were isolated and identified based on UV, NMR and mass spectrometry data. The presence of six compounds including isovitexirone, 24(28)-dehydromakisterone, 20-hydroxyecdysone 20,22-monoacetone, 20-hydroxyecdysone 22-acetate, inokosterone and makisterone C was detected for the first time in this species. Three previously detected compounds were confirmed in *S. centauroides* like integristerone A, 20-hydroxyecdysone and 2-deoxy-20-hydroxyecdysone. According to HPLC data, the highest content of phytoecdysteroids was observed in the leaves of *S. centauroides* (20.64 mg/g) and the lowest in tubular flowers (3.75 mg/g). The main component of the phytoecdysteroids sum was 20-hydroxyecdysone which concentration in the leaves reached 15.07 mg/g. Quantitative analysis of the aqueous medicinal preparations of *S. centauroides* (decoction, infusion) showed a high phytoecdysteroid content (0.77–1.01 mg/mL) and can be used as a source of this group of compounds.

Olennikov D.N. and Kashchenko N.I. (2019d) New ecdysteroids and lignan glycosides from *Rhaponticum uniflorum* (Compositae). XIIIth International Symposium on the Chemistry of Natural Compounds, October 16th-19th, 2019, Shanghai, China, p172 [conference abstract].

Olennikov D.N. and Kashchenko N.I. (2020) Ecdysteroids and glycosylflavones of *Silene sibirica* (Caryophyllaceae). Khimiya Rastitel'nogo Syr'ya (4), 109-119 [in Russian].

Abstract: The present work realized the chemical study of *Silene sibirica* (L.) Pers. (Caryophyllaceae) high-performance liquid chromatography with diode array and mass spectrometric detection (electrospray ionization) (HPLC-DAD-ESI-MS). As a result, 25 compounds were found, including nine ecdysteroids and sixteen flavonoids. Ecdysteroids components were podecdysone C, integristerone A, turkesterone, polypodine B, 20-hydroxyecdysone and its 2-*O*-cinnamate, ecdysone, 2-deoxy-20-hydroxyecdysone, and 2-deoxyecdysone. Flavonoids were the glycosylflavones and derivatives of luteolin and apigenin. Luteolin glycosides included luteolin-7-*O*-rutinoside, lucenin-2, carlinoside, isoorientin and its 2''-*O*-arabinoside, as well as isoscoparin. The largest group of apigenin glycosides included *O*-glycosides as 7-*O*-glucoside and 7-*O*-rutinoside, *C*-glycosides as isovitexin, shaftoside, vicenin-2, and mixed *C,O*-glycosides as isovitexin-2''-*O*-arabinoside and 2''-*O*-rhamnoside. Three unidentified derivatives of apigenin were pre studied and their structural features discussed. Quantitative data about the content of selected compounds indicated that ecdysteroids accumulated in *S. sibirica* flowers (7.14–14.92 mg/g) and glycosylflavones were found predominantly in leaves (7.88–18.55 mg/g). The major ecdysteroid compound was 20-hydroxyecdysone, while flavonoids predominants were shaftoside and isovitexin-2''-*O*-ramnoside. A comparative analysis of the chemical composition of wild-growing and cultivated *S. sibirica* samples showed the stability of the metabolic profile of the plants during the introduction. The biological studies revealed the antiradical and antiglycosidase activity of the extracts. Thus, it was shown that the studied plant species (*S. sibirica*) is a source of ecdysteroids and glycosylflavones, and *S. sibirica* extracts have biological potency.

Oliviera da Silva T.F., Yamaguchi C.S., Ribeiro S.T.C., da Silva Avicola A., Pilau E.J., Porto C., Braz de Oliveira A.J. and Goncalves R.A.C. (2021) Adventitious root culture of *Pfaffia glomerata* (Spreng.) Pedersen in a roller bottle system: an alternative source of b-ecdysone. Phytochemistry Letters **43**, 1-7.

Abstract: β -ecdysone is the main compound of commercial interest produced by the roots of *Pfaffia glomerata* (Spreng.) Pedersen, and is used in herbal medicines and dietary supplements. *In-vitro* root culture

facilitates the continuous production of high-quality compounds, and uses sustainable production techniques. Adventitious roots of *P. glomerata* were cultivated in a roller bottle system and an orbital gyratory system, both in the absence of light. The quantification of β -ecdysone by Liquid Chromatography-Mass Spectrometry/Mass Spectrometry (LC-MS/MS) revealed that the highest accumulation of metabolites occurred after 8 weeks in the roller bottle culture system, wherein 1.81 μ g of β -ecdysone was recovered from 30 mg of dry root. These results suggest that adventitious root culture *in-vitro* may be an alternative strategy for β -ecdysone production, and demonstrates the potential use of bioreactors for practical applications.

Ono M., Takamura C., Sugita F., Masuoka C., Yoshimitsu H., Ikeda T. and Nohara T. (2007) Two new steroid glycosides and a new sesquiterpenoid glycoside from the underground parts of *Trillium kamtschaticum*. Chemical and Pharmaceutical Bulletin **55**(4), 551-556.

Abstract: Two new steroid glycosides, named trikamsterosides A and B, and a new sesquiterpenoid glycoside named trikamsesquiside A were isolated from the underground parts of *Trillium kamtschaticum* PALL. along with 18 known compounds comprising 12 steroids, one sesquiterpenoid glycoside, one phenylpropanoid, one flavonoid glycoside, and three phenylpropanoid sucrose esters. Their chemical structures were determined on the basis of spectroscopic data and chemical evidence. Among them, one phenylpropanoid sucrose ester showed almost the same radical-scavenging effect on 1,1-diphenyl-2-picrylhydrazyl as that of α -tocopherol.

Orlova I.V., Nosov A.M., Volodin V.V., Ivanov A.L. and Butenko R.G. (1993) The potential of cell cultures of *Rhaponticum carthamoides* for the production of ecdysteroids. Scientific Reports of the Komi Research Centre, Issue 306, 16 pp. [in Russian, with an English abstract].

Orlova I.V., Nosov A.M., Luksha V.G. and Volodin V.V. (1994) Ecdysteroid biosynthesis in intact plants and cell cultures of *Rhaponticum carthamoides* Willd. (Iljin) [*Leuzea carthamoides* Willd.]. Fisiologiya Rastenii **41**, 915-920 [in Russian]/Russian Journal of Plant Physiology **41**, 799-804 [in English].

Ou Y., Luo Z., Cheng S., Zhang Y., Tang M., Tang C., Tang D., Xiao S., He Y. and Xiong G., Li S. (2018) Study on the ecdysterone constituents of *Achyranthes aspera* L. Journal of Traditional Chinese Medicine University of Hunan (10), 1129-1132 [in Chinese].

Abstract: Objective: To study the chemical composition of *A. aspera* (ox-knee), a special medicinal herbs of Xiang production. Methods: The initial separation of ethanol extraction and solvent extraction method was adopted, the compound separation and purification of silica and inverse silica column layering and solvent recrystallising were carried out, and the isolated compounds were structurally identified by spectral data, physical properties and literature control. Results: 7 moulting ketones were obtained from soil knee ethanol extract, namely: β -moulting ketones (1), ox knee steroids (2), ketones B (3), pterosterone (1) 4), 24 (28)-ecdysterone (5), achyranthesterone A (6), rubrosterone (7). Conclusion: Compounds 4-7 are all isolated from the plant for the first time.

Ouyang W., Luo Y-f., Li Z., Wang X-l., Zhang Y-k., Zhou H-r., Tang C-y. and Li S-x. (2020) Study on the isolation, identification and content determination of anti-inflammatory components in Tunixi. Natural Product Research and Development (7), 1171-1181 [in Chinese].

Abstract: The inflammation macrophages (RAW 264.7) model induced by lipopolysaccharide (LPS) with nitric oxide (NO) release as an index was established to investigate the anti-inflammatory components in different varieties of Tunixi and column chromatography was used to isolate the active compounds from *Achyranthes aspera* Linnaeus methanol extract. The chemical structures of the active components were deduced based on the nuclear magnetic resonance (NMR) and mass spectrometry (MS) data analysis and comparing with reference substances. The methanol extracts of *A. aspera* showed the best anti-inflammatory effect, furthermore, its 50%, 70% and 100% methanol eluates featured low cytotoxicity and high anti-inflammatory activity. Nine pure compounds isolated from the 50% and 70% methanol eluates were identified as β -ecdysterone (1), inokosterone (2), polypodine B (3), N-trans-feruloyl-tyramine (4), N-cis-feruloyl-tyramine (5), N-cis-feruloyl-3-methoxytyramine (6), N-trans-feruloyl-3-methoxytyramine (7), chikusetsusaponin Iva (8), 5,2'-dimethoxy-6-(methoxymethyl)-7-hydroxy-isoflavonol (10), meanwhile, oleanolic acid 28-O- β -D-glucopyranosyl ester (9) was elucidated as the main component in its 100% methanol eluate. At the concentration of LPS + 25 μ M, the anti-inflammatory activity order was 4 > 1 > 5 > 2 > 6 > 9 > 10 > 3 > 7 > 8. The β -ecdysterone content in the underground rhizomes of wild *A. bidentata* Blume and *A. longifolia* (Makino) Makino changed obviously with seasons and reached the highest level in August at 0.914 \pm 0.016 and 1.412 \pm 0.038 mg/g, respectively. However, there is no obvious seasonal variation observed in *A. aspera*. Moreover, feruloyl tyramine alkaloids with anti-inflammatory activity were firstly reported from *A. aspera* in this research.

Pan Q., Mu C-x. and Zhang J-j. (2015) Comparative study of content of oleanolic acid and ecdysterone from radix *Achyranthes bidentata* in different region. China Modern Medicine (12), 10-12 [in Chinese].

Abstract: Objective: To determine the content of saponin (oleanolic acid) and ecdysterone of *Achyranthes bidentata* from different habitat order to compare the difference of content of saponin (oleanolic acid) and ecdysterone of *Achyranthes bidentata* from different habitat. Methods: High-performance liquid chromatography (HPLC) was used and Octadecylsilane chemically-bonded silica was selected as chromatographic column. Methanol was selected as mobile phase in determination of oleanolic acid and acetonitrile as mobile phase determination of ecdysterone. The ultraviolet detection wavelength in oleanolic acid and ecdysterone was 280 nm and 250 nm, respectively. The sample size was both 10 μ L. Results: The content of saponin (oleanolic acid) and ecdysterone of *Achyranthes bidentata* was the highest with the best quality produced in Qinyang of Henan province, and the content of saponin (oleanolic acid) and ecdysterone of *Achyranthes bidentata* was higher produced in Anyang city of Henan province, Taihe county of Anhui province, Qujing city of Yunnan province, and Shenyang city which all met the standard of State Pharmacopoeia. Conclusion: The quality of *Achyranthes bidentata* produced in Qinyang city of Henan province is best and in other regions like Shenyang city is better which all meet the criteria of China Pharmacopoeia and are suitable for medication.

Pandit S.S., Naik S.D., Jathar V.S. and Kulkarni A.B. (1976) Insect moulting hormone, ecdysterone from *Sida carpinifolia* Linn. Indian Journal of Chemistry 14B, 907-908.

Pardede A., Adfa M., Kusnanda A.J., Ninomiya M. and Koketsu M. (2018) Isolation of secondary metabolites from *Stenochlaena palustris* stems and structure-activity relationships of 20-hydroxyecdysone derivatives on antitermite activity. Wood Research and Technology/Holzforschung 72(10), 899-

Abstract: *Stenochlaena palustris* is an edible fern, belonging to the Blechnaceae family and is called kelakai in South Kalimantan, Indonesia. The effects of fractions of *S. palustris* stems and isolated constituents on termite mortality and paper disc consumption by *Coptotermes curvignathus* have been studied. Treatment of the ethyl acetate (EtOAc) fraction on paper discs greatly induced death of *C. curvignathus* within 6 days. Our phytochemical investigation of *S. palustris* stems led to the isolation of major constituents and 20-hydroxyecdysone exerted the highest termiticidal activity, followed by stenopalustroside A and ajugasterone C. Moreover, the structure-activity relationships (SAR) of synthetic derivatives from 20-hydroxyecdysone and ajugasterone C suggested that a 2,3-diol has considerable effect on their antitermite properties against *C. curvignathus*.

Pathak A.K., Agarwal P.K., Jain D.C., Sharma R.P. and Howarth O.W. (1995) NMR studies of 20 β -hydroxyecdysone, a steroid; isolated from *Tinospora cordifolia*. Indian Journal of Chemistry 34B, 674-676.

Abstract: By using ID and 2D NMR spectra, including ¹H-¹H and ¹H-¹³C long- and one-bond shift correlation, and KOE difference spectra the structure of a steroid, isolated from the ethyl acetate extract of the aerial parts of *Tinospora cordifolia* has been established as 2 β , 3 β , 14 α , 20 α , 22 α , 25-hexahydroxy-5 α -cholest-7-en-6-one (20 β -hydroxyecdysone) (1). All ¹H and ¹³C resonances have been unequivocally assigned,

Pavlik M., Pavlikova D., Szakova J., Vasickova S. and Tlustos P. (2004) Binding forms of risk elements in root fractions of *Leuzea carthamoides* (Willd.) DC. International Biodeterioration and Biodegradation 54(2-3), 239-243.

Abstract: The aim of this study was to confirm that the content of elements posing a health risk and their distribution in root extracts of *Leuzea carthamoides* (Willd.) DC, a medicinal plant containing therapeutic compounds (ecdysteroids, flavonoids and *N*-feruloylserotonin isomers), should not be a factor limiting its use in therapeutic practice. The arsenic, cadmium, copper and lead content was determined for root dry matter and individual fractions of root dry matter produced by sequential extraction. Compounds with biological and therapeutic activity are present in petroleum ether, ethyl acetate, butanol and methanol fractions, which were found to contain minor concentrations of the four elements, i.e. 3.7% of the total arsenic in the root dry matter, and respectively 8.9%, 10.1% and 2.2% of the cadmium, copper and lead. The remaining fractions containing the major proportion of these elements are not used in the pharmaceutical industry. Since only small amounts were found in the root fractions for pharmaceutical use, it can be assumed that *L. carthamoides* can be grown at sites contaminated by these elements.

Pavliková D., Pavlík M., Vašíčková S., Száková J., Tlustoš P., Vokáč K. and Balík J. (2004) Separation of organic compounds binding trace elements in seeds of *Leuzea carthamoides* (Willd.) DC. Applied Organometallic Chemistry 18, 619-625.

Abstract: The distribution of trace elements into important groups of compounds in seeds was investigated using a seven-step sequential extraction of seed biomass (solvents used: petroleum ether, ethyl acetate, butanol, methanol, methanol + H₂O (1 + 1; v/v), H₂O, methanol + H₂O + HCl (49.3 + 49.3 + 1.4; v/v/v)). Isolated fractions were partially characterized using IR spectroscopy. Results of sequential analysis showed different portions of the elements investigated in individual fractions. The dominant portions of cadmium (60.6% of total content), lead (41%), zinc (77.8%) and copper (33.9%) were found in the methanol + H₂O + HCl fractions (compounds isolated from cell walls and cytoskeleton after hydrolysis—phytic acid and its salts, proteins). The second most significant fractions for cadmium, zinc and lead were in the water fractions (pectin, phytin) and for copper in the methanol

fraction (acids of citric cycle). The ethyl acetate fraction, mainly containing lignans and phospholipids, had the highest portion of arsenic (34.2%). Lignans are common compounds for seeds of *Leuzea carthamoides*. Therapeutic compounds of *L. carthamoides* (20-hydroxyecdysone, N-feruloylserotonin isomers) were confirmed in the first four fractions by thin-layer chromatography

Petruk A.A., Vysochina G.I. and Ershova E.A. (2013) The dynamics of astragalalin, isoquercitrin and 20-hydroxyecdysone in fronds of *Pteridium aquilinum* and *Matteuccia struthiopteris*, growing in the vicinity of Novosibirsk. *Khimiya Rastitel'nogo Syr'ya* (4), 151-157.

Abstract: Research is devoted to studying by methods of a highly performance liquid chromatography (HPLC) of the dynamics of the content flavonol glycosides astragalalin, isoquercitrin and ecdysteroid 20-hydroxyecdysone in fronds *Pteridium aquilinum* and *Matteuccia struthiopteris*, growing in the vicinity of Novosibirsk (Akademgorodok, CSBG SB RAS). The analysis carried out on the analytical HPLC-system consisting of the liquid chromatograph «Agilent 1200» with the with diode array detector. In *P. aquilinum* found all of the compounds, in *M. struthiopteris* – only astragalalin. At the beginning of June the content of astragalalin at both species is greatest, isoquercitrin in bracken – trace. Astragalalin at bracken remains at the level of 0,2% until the end of summer, at ostrich fern we observe some increase – to 0,14%. The quantity of isoquercitrin by the end of summer at bracken also increased – to 0,42%. The maximum content of 20-hydroxyecdysone – 0,91% – at *P. aquilinum* recorded in the middle of August, the minimum – in the middle of May in a phase of the beginning of growth

Pinheiro M.L.B., Filho W., da Rocha A.I., Porter B. and Wenker E. (1983) Abutasterone, an ecdysone from *Abuta velutina*. *Phytochemistry* 22(10), 2320-2321.

Abstract: The ecdysone abutasterone has been isolated from the Amazonian plant *Abuta velutina* and its structure elucidated by spectral means.

Píš J., Buděšinský M., Vokáč K., Laudová V. and Harmatha J. (1994) Ecdysteroids from roots of *Leuzea carthamoides*. *Phytochemistry* 37(3), 707-711.

Abstract: Three new ecdysteroids: polypodine B 20,22-acetonide, 20-hydroxy-ecdysone 2,3;20,22-diacetonide and isovitexirone along with 20-hydroxyecdysone, 20-hydroxyecdysone 2,3-acetonide, 20-hydroxyecdysone 20,22-acetonide, ajugasterone C, makisterone A and polypodine B were isolated from the roots of *Leuzea carthamoides*.

Pomilio A.B., González M.D. and Eceizabarrena C.C. (1996) 7,8-Dihydroajugasterone C, norhygrine and other constituents of *Nierembergia hippomanica*. *Phytochemistry* 41(5), 1393-1398.

Pongrácz Z., Báthori M., Tóth G., Simon A., Mák M. and Máthé I. (2003a) 9 α ,20-Dihydroxyecdysone, a new natural ecdysteroid from *Silene italica* ssp. *nemoralis*. *Journal of Natural Products* 66, 450-451.

Abstract: A new natural compound, 9 α ,20-dihydroxyecdysone (1), and two known related compounds, 20-hydroxyecdysone and ecdysone, were isolated from the herb *Silene italica* ssp. *nemoralis*. Compound 1 is the first C-9 hydroxylated ecdysteroid with a cis-fused A/B ring junction to have been isolated from a plant source, and its structure was determined using a combination of spectroscopic techniques.

Pongracz Z., Bathori M., Mathé I., Janisak G. and Miklossy V.V. (2003b) Ecdysteroids as varying chemical constituents of *Silene* species growing in Hungary. *Proceedings of the International Conference on MAP* (Eds. Bernath J. et al.) *Acta Hort.* 597, 131-135.

Abstract: 48 *Silene* species (Caryophyllaceae family) were tested by TLC/densitometry for ecdysteroids. It turned out that 50 % of the species contained 20-hydroxyecdysone (β -ecdysone) justifying that this compound is of widespread occurrence in this genus. *Silene italica*, *S. nutans*, *S. otites* contain, beside 20-hydroxyecdysone, integristeron A, both in varying quantities. *Silene otites* produced the highest ecdysteroid concentration in June and July, when *S. nutans* contained the least ecdysteroids. If the organs were compared with each other, in the case of both species, there is a decrease in the yield in the order inflorescence, leaf, root and stem.

Poyser J.P., Poyser K.A., Silva M. and Sammes P.G. (1973) Some components of *Podocarpus andina* Peopp. ex Endl. *Rev. Latinamer. Quím.* 4, 157-160.

Pradhan P., Gangan V.D., Sipahimalani A.T. and Banerji A. (1997) Two phytoecdysones from *Tinospora cordifolia*: structural assignment by 2D nmr spectroscopy. *Indian Journal of Chemistry B (Organic Chemistry including Medicinal Chemistry)* 36(10), 958-962.

Abstract: Two phytoecdysones viz. ecdysterone 1 and makisterone A 2 have been isolated as their acetates from the n-BuOH extracts of the medicinal plant *Tinospora cordifolia* stems. The structures and complete NMR assignments of the phytoecdysones have been achieved by extensive 1D and 2D NMR studies.

Prakash A. and Ghosal S. (1979) Phytoecdysones. *Journal of Scientific and Industrial Research (India)* **38**, 632-647.

Preston-Mafham J. and Dinan L. (2002) Phytoecdysteroid levels and distribution during development in *Limnanthes alba* Hartw. ex Benth. (Limanthaceae). *Zeitschrift für Naturforschung* **57c**, 144-152.

Abstract: Phytoecdysteroid (PE) production and accumulation in *Limnanthes alba* Hartw. ex Benth. is associated with flowering. PE content per plant remains fairly constant during the primary growth phase of the plant and only begins to increase significantly above amounts found in the seed once the development of the flower stalk has begun. Both content and concentration increase concomitantly from this point. Distributions in individual plants also associated the highest levels of PE accumulation with the reproductive tissues. This substantiates the association of PE with tissues of greatest fitness value and therefore the hypothesis that they contribute to defence. Analysis of extracts of *L. alba* tissues by reversed-phase HPLC coupled with ecdysteroid-specific RIA was used to monitor ecdysteroid profiles. RIA-positive peaks co-chromatographing with 20-hydroxyecdysone, ecdysone and ponasterone A were detected and several tissues also contain PE conjugates. Seedmeal of *L. alba* appears to be a convenient and promising source for the commercial isolation of the potent PE ponasterone A.

Punegov V.V. and Savinovskaya N.S. (2001) The internal standard method for the determination of ecdysteroids in herbs and preparations by HPLC analysis. *Rastitelny Resursy* **37**(1), 97-102 [in Russian, with an English abstract].

Pylina Y.I., Volodina S.O., Bacharov D.S. and Volodin V.V. (2010) Ecdysteroids of *Serratula quinquefolia* Bieb. ex Willd. 2nd Annual Russian-Korean Conference "Current issues of natural products chemistry and biotechnology", March 15-18, 2010, Novosibirsk, Russia, Poster Presentation, p. 116

Qian J.-j., Li X., Yang Y.-y., Lin J. and Chi D.-f. (2015) β -Ecdysterone accumulation and regulation in *Ajuga lobata* suspension culture. *Journal of the Beijing forestry University* (9), 91-100 [in Chinese, with an English abstract].

Abstract: Ecdysterone is a naturally occurring steroid hormone secreted by arthropods, including insects, to regulate the moulting process of larvae. Ecdysterone also has many pharmacological functions such as promoting cell growth and inducing human epidermal cell differentiation. Suspension culture of *Ajuga lobata* D. Don cells provides a method of synthesis of the phytoecdysteroid β -ecdysterone (β -EC). In this study, we tried to characterize the culture conditions to optimize β -EC production. Growth of *A. lobata* cells fit the logistic equation curve, with a growth cycle of 19 days and the stationary phase of 11 to 17 days. Medium conductivity was negatively correlated with dry cell weight and β -EC accumulation, thus could be used to determine the optimal time for cell harvest. Continuous subculture reduced β -EC synthesis, but supplementing medium with β -EC precursors mevalonic (MVA), metabolic inhibitors α -pinene and elicitor NO could significantly promote cell growth and influence β -EC accumulation. Combination of α -pinene, MVA and SNP significantly elevated β -EC accumulation, thus may synergistically enhance β -EC synthesis in *A. lobata*. The optimal concentrations of α -pinene, MVA and NO donor SNP in the suspension culture were 50 μ L/L, 10 mg/L, 80 μ mol/L, respectively.

Qian J., Yang Y., Li X. and Chi D. (2016) 20-Hydroxyecdysone accumulation and regulation in *Ajuga lobata* D. Don suspension culture. *Bioscience, Biotechnology and Biochemistry* **80**(3), 591-599.

Abstract: Suspension culture of *Ajuga lobata* D. Don cells provides a method of synthesis of the phytoecdysteroid 20-hydroxyecdysone (20E) which can regulate the molting process of larvae. We characterized the culture conditions to optimize 20E production. Growth of *A. lobata* D. Don cells fits the logistic equation curve with a growth cycle of 19 days. Medium conductivity was negatively correlated with dry cell weight and 20E accumulation, thus could be used to determine the optimal time for cell harvest. Continuous subculture reduced 20E synthesis, but supplementing medium with 20E precursors mevalonic (MVA), α -Pinene, and nitric oxide (NO) can significantly promote cell growth and influence 20E accumulation. Combination of α -Pinene, MVA, and SNP significantly elevated 20E accumulation, thus may synergistically enhance 20E synthesis in *A. lobata* D. Don. The optimal concentrations of α -Pinene, MVA, and NO donor SNP in suspension culture were 50 μ L L⁻¹, 10 mg L⁻¹, and 80 μ mol L⁻¹. Continuous culture of *A. lobata* D. Don will reduce the β -EC harvest. Supplemented with MVA, α -Pinene, and SNP to improve the content for saving cost.

Quan T.D., Hau D.V., Tam N.T., Thien D.D., Loc T.V., Sung T.V., Thang L.Q., Nhung L.T.H. and Thuy N.T. (2018) Ecdysteroid compounds and an abietane diterpenoid from the Lady Pine (*Dacrycarpus imbricatus*) collected in Lom Dong Province, Vietnam. *Vietnam Journal of Chemistry* **56**(3), 285-289 [in Vietnamese, with an English abstract].

Qiu J., Wang X., Wang Y. and Ding X. (2008) Research on chemical constitution of *Morus alba* leaves. *Zhongchengyao* **30**(9), S1-S2 [in Chinese].

Abstract: The chem. constitution of *Morus alba* leaves was separated via silica gel column chromatog. and Sephadex LH-20 column chromatog. The chem. constitution of *Morus alba* leaves was identified by ¹H NMR, ¹³C

NMR, infra-red spectrometry, TLC. The chem. constitution comprises azelaic acid, aesculetin, rutin, quercetin, isoquercitrin, kaempferol-7-O- β -D-glucopyranoside, 2,4-dihydroxy-benzoic acid, ecdysterone.

Rahman S.U., Adhikari A., Ismail M., Shah M.R., Khurram M., Anis I. and Ali F. (2017) A new trihydroxylated fatty acid and phytoecdysteroids from rhizomes of *Trillium govanianum*. Records of Natural Products 11(3), 323-327.

Abstract: A crude hydro-methanolic extract of *Trillium govanianum* ex D. Don (Melanthiaceae, Trilliaceae) rhizomes and its subsequent solvents soluble fractions were tested against different fungal strains i.e. *Trichophyton rubrum* ATCC 40051, *Aspergillus niger* ATCC 16888, *Candida albicans* ATCC 18804, *Microsporium canis* ATCC 32903 and *Fusarium lini* ATCC 16888. The hydro-methanolic extract showed significant activity against *T. rubrum* and *M. canis* with 80 and 75% inhibitions respectively. Among the fractions, chloroform soluble fraction showed 90% inhibition, with minimum inhibitory concentration (MIC) of 10 μ g/mL against *T. rubrum* followed by ethyl acetate, butanol and n-hexane fractions. The bio-activity guided isolation of chloroform soluble fraction leads to a new trihydroxylated fatty acid, named govanic acid (1) along with two known phytoecdysteroids i.e. 20-hydroxyecdysone (2) and 5, 20-dihydroxyecdysone (3). The structures of isolated compounds were elucidated through 1D, 2D-NMR spectroscopic data analysis. All compounds (1-3) in *T. govanianum* are reported herein for the first time. Compound 1 showed significant activity against *T. rubrum* with 70% inhibition and MIC value of 5 μ g/mL, but lack of activity against the other test strains.

Ramazanov N.S. (2004) Ecdysteroids and iridoidal glycosides from *Vitex agnus-castus*. Chemistry of Natural Compounds 40(3), 299-300 [in English]/*Khimiya Prirodnikh Soedinenii* (3), 251 (2004) [in Russian].
No Abstract.

Ramazanov N.S. (2005) Phytoecdysteroids and other biologically active compounds from plants of the genus *Ajuga*. Chemistry of Natural Compounds 41(4), 361-369 [in English]/*Khimiya Prirodnikh Soedinenii* (4), 293-299 [in Russian].

Abstract: Literature data on the structures of phytoecdysteroids and other biologically active compounds and their biological activities were reviewed.

Ramazanov N.S. (2005b) Phytoecdysteroids from *Serratula coronata* and *Silene longicalycina*. Chemistry of Natural Compounds 41(3), 359 [in English]/*Khimiya Prirodnikh Soedinenii* (3), 289 (2005) [in Russian].
No Abstract.

Ramazanov N.S., Maksimov E.S., Saatov Z and Abdullaev N.D. (1995) Phytoecdysteroids of the genus *Silene*. XVII. Tomentesterone A from *Silene tomentella*. *Khimiya Prirodnikh Soedinenii* (5), 714-719 [in Russian].

Ramazanov N.S., Maksimov E.S., Saatov Z. and Abdullaev N.D. (1996) Phytoecdysteroids of *Silene* plants. 18. Tomentesterone-B from *Silene tomentella*. *Khimiya Prirodnikh Soedinenii* (1), 59-61 [in Russian]/Chemistry of Natural Products 32(1), 47-49 [in English].

Ramazanov N.S., Maksimov E.S., Saatov Z. Mamatkhanov A.U. and Abdullaev N.D. (1997a) Phytoecdysteroids of *Rhaponticum* plants. 1. Carthamosterone A from *Rhaponticum carthamoides*. *Khimiya Prirodnikh Soedinenii* (3), 392-394 [in Russian]/Chemistry of Natural Compounds 33(3), 301-302 [in English].

Ramazanov N.S., Maksimov E.S., Saatov Z. and Abdullaev N.D. (1997b) Phytoecdysteroids of *Rhaponticum* plants. 2. Carthamosterone B from *Rhaponticum carthamoides*. *Khimiya Prirodnikh Soedinenii* (3), 395-396 [in Russian]/Chemistry of Natural Compounds 33(3), 303-304 [in English].

Ramazanov N.S., Mamadaliyeva N.Z. and Bobaev I.D. (2007) Phytoecdysteroids from five species of the genus *Silene*. Chemistry of Natural Compounds 43(1), 117-118 [in English]/*Khimiya Prirodnikh Soedinenii* (1), 97-98 (2007) [in Russian].
No Abstract.

Ramazanov N.S., Sultanov S.A., Saatov Z. and Nigmantullaeva N.M. (1997c) Phytoecdysteroids of *Silene* plants and dynamics of their content. *Khimiya Prirodnikh Soedinenii* (5), 718-723 [in Russian]/Chemistry of Natural Compounds 33(5), 558-562 [in English].

Ramazanov N.S., Bobayev I.D., Yusupova U.Y., Aliyeva N.K., Egamova F.R., Yuldasheva N.K. and Syrov V.N. (2017) Phytoecdysteroids-containing extract from *Stachys hissarica* plant and its wound-healing activity. Natural Product Research 31(5), 593-597 (DOI.org/10.1080/14786419.2016.1205058).

Abstract: A number of phytoecdysteroid compounds, such as ecdysterone, polipodin V, 2-deoxy-20-hydroxyecdysone, integristeron A and 2-deoxydizon were isolated from *Stachys hissarica* plant and their structures were confirmed by NMR, mass and IR spectroscopy. In addition, the biological activity of the *S. hissarica* plant's extract was tested on rats for wound healing activity. It was shown that the extract at repeated oral (per os) administration at a dose of 10 mg/kg speeds up the healing process of linear skin wounds in rats. The wound-healing activity of *S. hissarica* extract is confirmed to be effective and exceeds known drug methyluracil (2,4-dioxo-6-methyl-1,2,3,4-tetrahydropyrimidine), especially in case of alloxan induced diabetic animals.

Ramazonov N., Yusupova U., Egamova F. and Syrov V. (2020) Anti-stress activity of phytoecdysteroids isolated from aerial part of *Silene claviformis*. Chemical Science International Journal 29(7) 9-17 (article CSIJ.60497).

Abstract: Aims: The aim of present study was to isolate the phytoecdysteroids from aerial part of *Silene claviformis* (Caryophyllaceae) and investigate their biological activity. Place and Duration of Study: The investigation were carried out during 2019 and 2020 at laboratory of the chemistry of glycosides and department of the pharmacological and toxicology of Institute of the Chemistry of Plant Substances AS RUz, Tashkent, Uzbekistan. Methodology: The phytoecdysteroids were isolated from aerial part of *Silene claviformis* using chromatographic methods. Thin-layer chromatography made on Silufol UV-254 and Merck plates, Fluka Analytical Germany, by spraying with alcohol solution of vanillin and heating for 1-2 minutes for 90-1000, UV lamp light at 254 nm and 365 nm. Their structures were confirmed by NMR and IR spectroscopy. Sum of phytoecdysteroids was administered at a dose of 10 mg/kg orally. The data obtained during the experiments were processed and analyzed by the method of variation statistics using the Student t-criterion. Results: *Silene claviformis* contains 2-deoxyecdysterone (1), polypodine B (2), 20-hydroxyecdysone (3), ecdysterone-20,22-acetalisovaleric aldehyde (4), integristeron A (5), cyasterone (6), ecdysterone-20,22-acetalisovalerian (7), 2-deoxy- α -ecdysone (8). The compounds 2 and 6 are reported for the first time from this genus. The biological activity (stress-protective effect) of the mentioned phytoecdysteroids studied for the first time. These compounds were tested on male mice for their biological activities and results showed that, the adrenal gland mass increased in relation to the adrenal gland mass of intact animals by 42.6%, they showed a significant decrease in the content of ascorbic acid and cholesterol by 56.5 and 49.1%, respectively. A significant decrease in glycogen content (by 30.1%) and a noticeable activation of lipid peroxidation processes were noted in the liver, as indicated by an increase of 69.2% in the content of malondialdehyde in the organ. Conclusion: This is the first study reporting an orally biological investigation for *Silene claviformis*. The sum of phytoecdysteroids showed potent stress-protective activity. The stress-protective effect of phytoecdysteroids was more pronounced in compared the eleutherococcus extract.

Rao K.V. and Johnson J.H. (1998) Occurrence of 2,6-dimethoxy cinnamaldehyde in *Taxus floridana* and structural revision of taxiflorine to taxchinin M. Phytochemistry 49(5), 1361-1364.

Abstract: Taxiflorine, originally isolated from the needles of *Taxus floridana* and described previously, has its structure revised to that of taxchinin M. Four other known taxanes also isolated were: 1-deoxy baccatin IV, 1-hydroxy baccatin 1, 10-deacetyl paclitaxel-7-xyloside and 13-deacetyl taxiflorine (taxchinin L), together with: trans-2,6-dimethoxy cinnamaldehyde, rhododendrol, ponasterone A and α -conidendrin.

Rastrelli L., de Tommasi N. and Ramos I. (1996) Ecdysteroids in *Chenopodium pallidicaule* seeds. Biochemical Systematics and Ecology 24(4), 353.

Ravishankar G.A. and Mehta A.R. (1979) Control of ecdysterone biogenesis in tissue cultures of *Trianthema portulacastrum*. Journal of Natural Products 42, 152-158.

Abstract: Seedling callus of *Trianthema portulacastrum* was established and screened for phytoecdysone by a bioassay on the larvae of house-fly *Musca domestica*. Methods for detection, extraction, separation, and estimation of ecdysterone are described. Effects of various phytohormones and different sucrose levels on growth and ecdysterone production by callus tissues were examined. Naphthaleneacetic acid supported maximum growth of callus and 2,4-dichlorophenoxyacetic acid (2,4-D) yielded maximum ecdysterone. Increasing kinetin concentrations were inhibitory to ecdysterone biogenesis. A combination of 2,4-D with kinetin enhanced growth and ecdysterone production. Gibberellic acid was antagonistic to callus growth enhanced by 2,4-D and kinetin; however, it stimulated ecdysterone biogenesis. The present study indicates the feasibility for increased ecdysterone production in callus cultures as compared to the intact plant.

Raynor M.W., Kithinji J.P., Bartle K.D., Games D.E., Mylechreest I.C., Lafont R., Morgan E.D. and Wilson I.D. (1989) Packed column supercritical fluid chromatography and linked supercritical fluid chromatography-mass spectrometry for the analysis of phytoecdysteroids from *Silene nutans* and *Silene otites*. Journal of Chromatography 464, 292-298.

No Abstract.

Read H., Wilson I.D. and Lafont R. (1990) A note on overpressure thin-layer chromatography of ecdysteroids. In: Chromatography and isolation of insect hormones and pheromones (Eds. McCaffery A.R. and Wilson I.D.), Plenum Press, New York, pp. 127-130.

Reixach N., Irurre-Santilari J., Casas J., Mele E., Messeguer J. and Camps F. (1996) Biosynthesis of phytoecdysteroids in *in vitro* prothalli cultures of *Polypodium vulgare*. *Phytochemistry* **43**(3), 597-602.

Abstract The concentration of ecdysteroids in *Polypodium vulgare* has been studied in different parts of the wild plant as well as in the two *in vitro* micropropagated tissues. The ecdysteroid composition (abutasterone, polypodine B, 20-hydroxyecdysone, inokosterone, 24-hydroxyecdysone, pterosterone and ecdysone) was the same for all the sources investigated, but quantitative differences were found among them, with the exception of spores that did not produce these compounds. In contrast, the *in vitro* culture of spores originated the formation of haploid (prothalli) and diploid (sporophytes) tissues with a common feature: a proportion of 25-deoxyecdysone derivatives higher than that found in the wild plant. Prothalli micropropagated *in vitro*, generated from spores collected in different European areas, exhibited characteristic phytoecdysteroid contents that were constant over the four-year period studied. On the other hand, results on incorporation of radiolabelled ecdysteroid precursors, such as mevalonate, cholesterol or ecdysone, indicated that our *in vitro* system is adequate for biosynthetic studies. Labelled mevalonate and cholesterol were incorporated into all identified ecdysteroids, and labelled ecdysone was biotransformed into other 25-hydroxyecdysteroids present in this plant. Based on the different rates of incorporation of these precursors, a biosynthetic pathway for the last steps of ecdysteroid biosynthesis in *P. vulgare* was proposed.

Reixach N., Irurre-Santilari J., Camps F., Mele E., Messeguer J. and Casas J. (1997) Phytoecdysteroid overproduction in *Polypodium vulgare* prothalli. *Phytochemistry* **46**(7), 1183-1187.

Reixach N., Lafont R., Camps F. and Casas J. (1999) Biotransformations of putative phytoecdysteroid biosynthetic precursors in tissue cultures of *Polypodium vulgare*. *European Journal of Biochemistry* **266**, 608-615.

Abstract: Incubation of calli and prothalli of *Polypodium vulgare* with different tritium-labelled ecdysteroids has led to modification of some previous assumptions about the biosynthesis of ecdysteroids in plants. Thus, 25-deoxy-20-hydroxyecdysone was transformed efficiently in both tissues into 20-hydroxyecdysone (20E), but no 25-deoxyecdysteroids such as pterosterone and inokosterone were formed. Likewise, incubation of 2-deoxyecdysone (2dE) produced exclusively ecdysone (E) and 20E, indicating a high 2-hydroxylase activity in both tissues, despite calli not producing phytoecdysteroids. This 2-hydroxylation was also evident in the transformation of 2,22-dideoxyecdysone (2,22dE) into 22-deoxyecdysone (22dE). Different ecdysteroids that do not occur in *P. vulgare* were formed in the incubation of 3-dehydro-2,22,25-trideoxyecdysone (3D2,22,25dE) by 3 α -reduction and 3 β -reduction and 25-hydroxylation processes. The fact that 22,25-dideoxyecdysone and 22dE were the only 2-hydroxylated products formed in this case suggests that only compounds bearing a 3 β -hydroxyl group are substrates for the 2-hydroxylase. Surprisingly, 22-hydroxylation was never observed with either 2,22dE or 3D2,22,25dE, raising the possibility that it could occur at an early step in the biosynthetic pathway. In this respect, labelled 22R-hydroxycholesterol was efficiently converted into E and 20E, whereas 22S-hydroxycholesterol was not transformed into ecdysteroids, because of its unsuitable configuration at C22. Finally, the conversion of 25-hydroxycholesterol into E and 20E was greatly enhanced after thermal treatment of prothalli which induces the release of previously stored ecdysteroids. Thus, *P. vulgare* prothalli and calli appear to be particularly suitable models for the study of ecdysteroid biosynthesis and its regulation in plants.

Rele S., Banerji A., Chintalwar G., Kumar V. and Yadava V. (2003) A new conformer of 20-hydroxyecdysone from *Sesuvium portulacastrum*: an X-ray crystallographic study. *Natural Product Research* **17**(2), 103-108.

Abstract: X-ray single crystal analysis of 20-hydroxyecdysone (20E, 2), an important insect moulting hormone, isolated from *Sesuvium portulacastrum* Linn. was found to exhibit a unique stereochemical configuration revealing it to be a different conformer (polymorph) possessing the aliphatic chain atoms in a trans configuration. Moreover, the analysis also revealed the presence of three molecules of water of crystallisation thereby restricting the freedom of the aliphatic side chain.

Revina T.A. and Gureeva I.I. (1985) Ecdysterone content in ferns of mountain regions of South Siberia. *Rastitelny Resursy* **21**(1), 75-78 [in Russian].

Revina T.A., Karnachuk R.A., Tajlasheva T.J. (1986): Dynamics of 20-hydroxyecdysone in overground part of *Serratula coronata* L. and influence of different spectral composition on it. *Rastitelny Resursy* **22**, 70-72.

Revina T. A., Revushkina A. S. and Rakitin A. V. (1988) Ecdysteroid-containing species in flora of the Altai mountains. *Rastitelny Resursy* **34**(4), 565-570 [in Russian].

Rimpler H. (1967) Steroide mit ecdysonartiger Wirkung aus *Vitex megapotamica* [Steroids with ecdysone-like action from *Vitex megapotamica*]. Pharmazeutische Zeitung 48, 1799-1800 [in German].

Rimpler H. (1969) Pterosteron, Polypodin B und ein neues ecdysonartiges Steroid (Viticosteron E) aus *Vitex megapotamica* [Pterosterone, polypodine B and a new ecdysone-like steroid (viticosterone E) from *Vitex megapotamica*]. Tetrahedron Letters 10(5), 329-333 [in German].

No Abstract.

Rimpler H. (1972a) Iridoids and ecdysones from *Vitex* species. Phytochemistry 11, 2653-2654.

Rimpler H. (1972b) Phytoecdysone und Iridoide aus *Vitex megapotamica* [Phytoecdysones and iridoids from *Vitex megapotamica*]. Archiv der Pharmazie 305(10) 746-751 [in German, with an English abstract].

Rimpler H. and Schultz G. (1967) Vorkommen von 20-Hydroxyecdysone in *Vitex megapotamica* [The presence of 20-hydroxyecdysone in *Vitex megapotamica*]. Tetrahedron Letters 8(22), 2033-2035 [in German].

Ripa P.V., Martin E.A., Cocclione C.M. and Adler J.H. (1990) Fluctuation of phytoecdysteroids in developing shoots of *Taxus cuspidata*. Phytochemistry 29(2), 425-427.

Abstract Phytoecdysteroids from the vegetative shoots of *Taxus cuspidata*, an evergreen shrub, were found to fluctuate at different developmental stages during shoot growth. The quantity of ecdysterone (2 β ,3 β ,14 α ,20 R,22 R,25-hexahydroxy-5 β -cholest-7-en-6-one) was 47 \pm 5 mg/kg fresh weight, while the quantity of ponasterone A (2 β ,3 β ,14 α ,20 R,22 R-pentahydroxy-5 β -cholest-7-en-6-one) was 23 mg/kg from leaves of one- to two-year-(52-104 week) old shoots. One-, four-, 18- and 37-week-old leaves and soft shoots contained, on a fresh weight basis, respectively, 56, 52, 114 and 127 mg/kg of ecdysterone and 2, 2, 23 and 15 mg/kg of ponasterone A. These results indicate that ecdysteroid accumulation is dynamic and possibly driven by cycles of synthesis, transport, and degradation.

Rosa H.S., Salgueiro A.F.C., Noronha D.S., Silva M.P., Pereira G.C., Mendez A.S.L. and Folmer V. (2014) Protective effect against *in vitro* lipid peroxidation and chemical composition of *Sida tuberculata* extracts. XLIII Annual Meeting of the Sociedade Brasileira de Bioquímica e Biologia Molecular, Foz do Iguaçu, PR, Brazil, May 17-20th, 2014.

Rostagno M.A., Debien I.C.N., Vardanega R., Nogueira G.C., Barbero G.F. and Meireles M.A.A. (2014) Fast analysis of β -ecdysone in Brazilian ginseng (*Pfaffia glomerata*) extracts by high-performance liquid chromatography using a fused-core column. Analytical Methods 6, 2452-2459.

Abstract: The recent development of fused-core technology in HPLC columns is enabling faster and highly efficient separations. This technology was evaluated for the development of a fast analysis method for β -ecdysone in extracts of *Pfaffia glomerata*. A step-by-step strategy was used to optimize temperature (30–55 °C), flow rate (1.0–2.0 mL min⁻¹), mobile phase composition (mixtures of water and methanol or acetonitrile) and equilibration time (1–5 min). A gradient method has been developed using two solvents: 0.1% acetic acid in water and 0.1% acetic acid in acetonitrile. Optimized conditions provided a method for the separation of β -ecdysone in approximately 2 min with a total analysis time (sample-to-sample) of 9 min, including the return to initial conditions and the re-equilibration of the column. Evaluation of chromatographic performance revealed excellent intraday and interday reproducibility (>99.5%), resolution (2.78), selectivity (1.13), and peak symmetry (1.09) while presenting low limits of detection (0.20 mg L⁻¹) and quantitation (0.67 mg L⁻¹). The robustness of the method has also been calculated according to the concentration/dilution of the sample. Several sample solvents were evaluated and the best chromatographic results were obtained using 80% methanol in water. Finally, the developed method was validated with different extracts of *Pfaffia glomerata* samples.

Roth U., König M and Seifert K. (1995) Ecdysteroids from *Penstemon venustus*. Phytochemistry 39(4), 941-942.

Abstract: For the first time ecdysteroids have been found in the genus *Penstemon*. In addition to the known ecdysteroids taxisterone, 20-hydroxyecdysone, makisterone A and C, the new ecdysteroid venustone has been isolated from the roots of *Penstemon venustus*. This ecdysteroid was shown to be 22-O-[(3R)-3-hydroxybutanoyl]-20-hydroxyecdysone. The structure has been determined primarily on the basis of NMR spectroscopy. The assignments of the NMR signals were performed by means of ¹H-¹H COSY – 45° and ¹H-¹³C COSY experiments.

Ruan H, Miao J., Ma D. and Liu S. (2012) Determination of gentiopicrin and ecdysterone in *Gentiana macrophylla* Pall and *Achyranthes bidentata* BI by HPLC. Journal of Heilongjiang Bayi Agricultural University (6), 33-35 and 40 [in Chinese].

Abstract: Established an RP-HPLC system to measure the content of pyrethroids [?] in the qin and ox-knee. Using the Kromasil C18 column (4.6mm×250mm, 5µm); 1% acetic acid aqueous solution (B) for gradient elution [?]; 0mL min⁻¹ [?]; Detection wavelength: 254nm; column temperature: 25°C. The sample concentrations of glyphosate [?] and moulting steroids were 0.80~20.00, 0.40~10.00µg·mL⁻¹. The linear relationship in the range is good, and the average recovery rate (n=6) is 100.02% and 99.95%. Conclusion: This method is simple, accurate, reproducible and has a strong application value. [Problems with translation program?]

Rudel D., Bathori M., Gharbi J., Girault J-P., Racz I., Melis K., Szendrei K. and Lafont R. (1992) New ecdysteroids from *Serratula tinctoria*. *Planta Medica* **58**, 358-364.

Abstract: Six new ecdysteroids have been isolated from *SERRATULA TINCTORIA*; these are: the 2,22- and 3,22-diacetates of 20-hydroxyecdysone, 5β-hydroxyrubrosterone, 3-epi-poststerone, 3-epi-rubrosterone, and 22-oxo-20-hydroxyecdysone. These minor compounds were found together with the known ecdysteroids, 20-hydroxyecdysone, its 2-, 3-, and 22-monoacetates, rubrosterone, poststerone, polypodine B (5β,20-dihydroxyecdysone), pterosterone (25-deoxy-20,24-dihydroxyecdysone), and makisterone C (24-ethyl-20-hydroxyecdysone). All these ecdysteroids were isolated by a combination of several chromatographic techniques (liquid chromatography on alumina, DCCC, and HPLC), then identified using standard mass spectrometric and 2D (1)H-NMR techniques.

Rufai Z.H., Munshi N.A., Sharma R.K., Khurshed A., Malik G.N. and Raja T.A. (2011) Occurrence of insect moulting hormone (β-ecdysone) in some locally available plants. *International Journal of Advanced Biological Research* **1**(1), 104-107.

Russell G.B. (1972) Phytoecdysones from *Phymatodes novae-zelandiae*. *Phytochemistry* **11**, 1496.
No Abstract.

Russell G.B. and Fenemore P.G. (1970) Insect moulting hormone activity of some New Zealand Gymnosperms. *New Zealand Journal of Science* **13**, 61-68.

Russell G.B. and Fenemore P.G. (1971) Insect moulting hormone activity in some New Zealand ferns. *New Zealand Journal of Science* **14**, 31-35.

Russell G.B. and Fraser J.G. (1973) Insect moulting hormones: dacrysterone, a new phytoecdysone from *Dacrydium intermedium*. *Australian Journal of Chemistry* **26**, 1805-1807.

Abstract: Dacrysterone, a new phytoecdysone, has been isolated from the bark of *Dacrydium intermedium* and shown to be 5β-hydroxymakisterone A (6). Makisterone A also occurs in this plant.

Russell G.B., Horn D.H.S. and Middleton E.J. (1971) New phytoecdysones from *Dacrydium intermedium*. *Journal of the Chemical Society, Chemical Communications* **71**.

Abstract: Two new phytoecdysones from *Dacrydium intermedium* are shown to be crustecdysone 2-cinnamate (I) and polypodine B 2-cinnamate (II).

Russell G.B., Fenemore P.G., Horn D.H.S. and Middleton E.J. (1972) Insect moulting hormones: the phytoecdysones of *Dacrydium intermedium*. *Australian Journal of Chemistry* **25**, 1935-1941.

Abstract: The 2-cinnamate esters of P-ecdysone, polypodine B, and ponasterone C and the 3-p-ourmarate ester of p-ecdysone have been isolated from the bark of *Dacrydium intermedium* as new natural ecdysone esters. P-Ecdysone, polypodine B, ponasterone C, and pterosterone also occur in this plant.

Russell G.B., Greenwood D.R., Lane G.A., Blunt J.W. and Munro M.H.G. (1981) 2-Deoxy-3-epiecdysone from the fern *Blechnum vulcanicum*. *Phytochemistry* **20**(10), 2407-2410.

Abstract: 2-Deoxy-3-epiecdysone and ecdysone were isolated from fronds of the fern *Blechnum vulcanicum*. The structure and stereochemistry of the epiecdystero

Russo H.M., Queiroz E.F., Marcourt L., Rutz A., Allard P-M., De Almeida R.F., Carvahlo N.M., Wolfender J-L. and da Silva Bolzani V. (2020) Phytochemical analysis of the methanolic leaves extract of *Niederzuehlla multiglandulosa* (Malpighiaceae), a plant species toxic to cattle in Brazil. *Phytochemistry Letters* **37**, 10-16.

Abstract: *Niederzuehlla multiglandulosa* (Malpighiaceae) is a neotropical liana responsible for cattle intoxication outbreaks in Brazil that may lead to abortion and even death of the animals, causing substantial losses in Brazilian trade balance. A phytochemical study of *N. multiglandulosa* leaves was performed in order to obtain chemical information about secondary metabolites of this poorly studied species. The methanolic extract was purified by medium pressure liquid chromatography (MPLC) combined with semi preparative HPLC-PDA leading to the

isolation of 16 compounds, including a new steroid named 5-hydroxypodecdysone B (13). The structure of the isolated compounds was determined by classical spectroscopic methods such as NMR and HRMS. Complementary 19F NMR and UPLC-HRMS analysis did not detect the presence of monofluoroacetate (MFA), a toxic compound previously described from this species.

Rybin V., Boltenkov E. and Novozhilova E. (2007) Application of high-performance liquid chromatography for simultaneous identification of integristerone A, 20-hydroxyecdysone, ecdysone and 2-deoxy-20-hydroxyecdysone. *Natural Product Communications* 2(11), 1101-1104.

Abstract: Conditions for separation and identification of integristerone A (1), 20-hydroxyecdysone (2), ecdysone (3) and 2-deoxy-20-hydroxyecdysone (4) by reversed-phase HPLC with UV and mass-selective detection at atmospheric pressure chemical ionization have been developed. Correlation between quasi-molecular ions fragmentation and structure peculiarities of the studied ecdysteroids has been shown. Application of these conditions for the analysis of extract from the inflorescences of *Serratula komarovii* made it possible to reveal and to identify compounds 1, 2 and 4.

Saad M.L., Kovalenko P.G., Medvedeva T.V., Korniets G.V., Shuman N.V., Kholodova Y.D. and Galkin A.P. (1992) Cell and tissue culture of *Serratula wolffii* as a source of biologically active phytoecdysteroids. *Fiziologiya i Biokhimiya Kul'turnykh Rastenii* 24(6), 611-615 [in Russian].

Saatov Z., Usmanov B.Z. and Abubakirov N.K. (1979a) Phytoecdysones of *Silene praemixta*. I. Silenosterone. *Khimiya Prirodnykh Soedinenii* (6), 793-797 [in Russian].

Abstract: In addition to known ecdysteroids (2-deoxy- α -ecdysone and 2-deoxyecdysterone) from *Silene praemixta* (Caryophyllaceae) we have isolated new ones — premixisterone and selenosterone (I), C₂₇H₄₂O₇, mp 115–117°C (from MeOH), $[\alpha]_{D28} +86.9 \pm 2^\circ$ (c 0.92, MeOH), yield 0.003%. The acetylation of (I) with (CH₃CO)₂O in Py gave 22-acetyl-selenosterone (II), C₂₉H₄₄O₆, mp 210–212°C (MeOH-C₆H₁₄), $[\alpha]_{D27} +45.5 \pm 3^\circ$ (c 0.16; MeOH). On the basis of physicochemical and spectral characteristics it has been established that (I) has the structure of 14 α ,22R,25-trihydroxy-5 β -cholest-7-ene-3,6-dione. The IR, PMR, and mass spectra of (I) and (II) are presented.

Saatov Z., Usmanov B.Z. and Abubakirov N.K. (1979b) Phytoecdysones of *Silene praemixta*. II. Praemixisterone. *Khimiya Prirodnykh Soedinenii* (6), 797-799 [in Russian]/*Chemistry of Natural Compounds* (6), 703-705 [in English].

Saatov Z., Gorovits M.B., Abdullaev N.D., Usmanov B.Z. and Abubakirov N.K. (1981) Phytoecdysteroids of *Silene* plants. III. Silenoside A - a new ecdysteroid glycoside from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (6), 738-744 [in Russian].

Saatov Z., Gorovits M.B., Abdullaev N.D., Usmanov B.Z. and Abubakirov N.K. (1982) Phytoecdysteroids of *Silene* plants. V. Silenoside B - digalactoside of ecdysterone from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (5), 611-615 [in Russian].

Saatov Z., Gorovits M.B., Abdullaev N.D., Usmanov B.Z. and Abubakirov N.K. (1982b) Phytoecdysteroids of *Silene* plants. IV. Silenoside C - integristerone A galactoside from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (2), 211-214 [in Russian].

Saatov Z., Abdullaev N.D., Gorovits M.B. and Abubakirov N.K. (1984a) Phytoecdysteroids of *Silene* plants. VI. ecdysone 22-sulphate - a new ecdysteroid from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (4), 467-470 [in Russian].

Saatov Z., Abdullaev N.D., Horowitz M.B. and Abubakirov N.K. (1984b) Phytoecdysteroids of *Silene* plants. VII. Silenoside D - 3-O- α -D-galactopyranoside of ecdysterone from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (6), 741-744 [in Russian]/*Chemistry of Natural Compounds* 700-703 (1985) [in English].

Saatov Z., Gorovits M.B., Abdullaev N.D., Usmanov B.Z. and Abubakirov N.K. (1985) Phytoecdysteroids from *Silene* plants. VIII. 2-Deoxyecdysterone 3-acetate from *Silene praemixta*. *Khimiya Prirodnykh Soedinenii* (1), 60-62 [in Russian].

Saatov Z., Gorovits M.B., Melibaev S. and Abubakirov N.K. (1986a) Phytoecdysteroids of *Silene* plants. IX. Ecdysterone 22-benzoate from *Silene scabrifolia*. *Khimiya Prirodnykh Soedinenii* (1), 77-80 [in Russian].

Abstract: An ecdysteroid which has proved to be ecdysterone 22-O-benzoate has been isolated from the epigeal organs of *Silene scabrifolia* Kom.

- Saatov Z., Abdullaev N.D., Horowitz M.B. and Abubakirov N.K. (1986b) Phytoecdysteroids of *Silene* plants. X. Silenoside E - 2-deoxyecdysone-3-O- β -D-glucopyranoside from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (3), 323-326 [in Russian].
- Saatov Z., Horowitz M.B. and Abubakirov N.K. (1986c) Phytoecdysteroids from *Silene* plants. XI. 2-Deoxyecdysone 3-acetate from *Silene scabrifolia*. *Khimiya Prirodnykh Soedinenii* (4), 439-441 [in Russian].
- Saatov Z., Gorovits M.B., Abdullaev N.D. and Abubakirov N.K. (1987a) Phytoecdysteroids of plants of the genus *Silene*. XII. 5 α -Ecdysterone 22-O-benzoate from *Silene scabrifolia*. *Khimiya Prirodnykh Soedinenii* 678-681 [in Russian].
- Saatov Z., Gorovits M.B. and Abubakirov N.K. (1987b) Phytoecdysteroids of plants of the genus *Silene*. XIII. Ecdysterone 20,22-monoacetone from *Silene scabrifolia*. *Khimiya Prirodnykh Soedinenii* (5), 767-768 [in Russian].
- Saatov Z., Gorovits M.B. and Abubakirov N.K. (1987c) Phytoecdysteroids in plants of the genus *Silene*. XV. 2-Deoxyecdysone 22-O-benzoate from *Silene wallichiana*. *Khimiya Prirodnykh Soedinenii* (6), 852-855 [in Russian]/*Chemistry of Natural Compounds* (6), 708-711 [in English].
- Saatov Z., Gorovits M.B. and Abubakirov N.K. (1988) Phytoecdysteroids of the *Silene* genus. XVI. Viticosterone E 22-O-benzoate from *Silene wallichiana*. *Khimiya Prirodnykh Soedinenii* (4), 546-549 [in Russian].
Abstract: A new phytoecdysteroid, viticosterone E 22-O-benzoate (I), C₃₆H₅₀O₆, mp 147- 149°C (methanol-water), [a]_D²⁰ +63.20 (methanol), has been isolated from the epigeal organs of *Silene wallichiana* Klotzsch. The alkaline hydrolysis of (I) led to viticosterone E and benzoic acid. The acetylation of (I) gave the 2,3-diacetate (II), C₄₀H₅₄O₁₁, mp 152-153°C (methanol-water), [a]_D²⁰ +65.5° (methanol). Details of the IR, PMR, and mass spectra of (I) and (II) are given.
- Saatov Z., Abdullaev N.D., Gorovits M.B. and Abubakirov N.K. (1990a) Phytoecdysteroids of *Silene* plants. XVII. Ecdysterone 22,25-di-O-benzoate from *Silene scabrifolia*. *Khimiya Prirodnykh Soedinenii* (3), 363-366 [in Russian].
- Saatov Z., Gorovits M.B. and Abubakirov N.K. (1990b) Phytoecdysteroids of the plant *Dianthus hoeltzeri*. *Khimiya Prirodnykh Soedinenii* (5), 837 [in Russian].
- Saatov, Z., Umarova, R. U., Gorovits, M. B., & Abubakirov, N. K. (1990c) Phytoecdysteroids from plants of the genus *Melandrium*. 1. Polypodine B 22-acetate from *Melandrium turkestanicum*, N. K. *Khimiya Prirodnykh Soedinenii* 480-483 [in Russian].
- Saatov Z., Gorovits M.B. and Abubakirov N.K. (1991) Phytoecdysteroids in plants of the genus *Melandrium* II. melandrioid A - viticosterone E galactoside from *Melandrium turkestanicum*. *Khimiya Prirodnykh Soedinenii* (4), 517-520 [in Russian].
- Saatov Z., Gorovits M.B. and Abubakirov N.K. (1993) Phytoecdysteroids in plants of the genus *Silene*. *Khimiya Prirodnykh Soedinenii* (5), 627-635 [in Russian]/*Chemistry of Natural Products* 29(5), 551-557 (1994) [in English].
- Saatov, Z., Syrov, V. N., Mamatkhanov, A. V., & Abubakirov, N. K. (1994) Phytoecdysteroids of *Ajuga* and biological activity 1. Occurrence and chemical structure of isolated compounds. *Khimiya Prirodnykh Soedinenii* (2), 152-160 [in Russian].
- Saatov Z, Agzamkhodzhaeva DA, Syrov VN (1999) Distribution of phytoecdysteroids in plants of Uzbekistan and the possibility of using drugs based on them in neurological practice. *Khimiya Prirodnykh Soedinenii* (2), 209-215 [in Russian]/*Chemistry of Natural Compounds* 35(2), 186-191 [in English].
Abstract: The distribution of phytoecdysteroids in plants of the families Labiatae, Compositae and Caryophyllaceae growing in Uzbekistan has been analysed. It has been shown that some of them possess a capacity for lowering the levels of urea and residual nitrogen in the blood and for improving the functional state of the kidneys in various pathological states. The dependence of biological activity on the structure of the compounds is discussed. The possibility of broadening the indications for the use of the drug ekdyisten, the first to have been created from compounds of this class, in complications affecting the eyes of patients suffering from chronic glomerulonephritis has been substantiated experimentally and clinically.

Sabri N.N., Asaad A. and Khafagy S.M. (1981) Isolation of four ecdysones from *Ajuga iva* roots and a rapid semiquantitative method for ecdysone determination. *Planta Medica* 42, 293-295.

Abstract: The ethereal extract of *Ajuga iva* (L.) SCHREB. roots, yielded 4 ecdysones. Three were proved to be cyasterone, makisterone A and ecdysterone, by using UV, IR, MS spectral methods and comparison with standard samples. Spectral data indicate that the fourth ecdysone is similar to cyasterone but having an additional hydroxy group in the side-chain. The chromatographic pattern of ecdysone content in roots, stems and leaves is presented as well as a rapid and simple semiquantitative method for their estimation. The roots had the highest total content (0.381 %).

Sadati N., Ostad S.N., Karimian Z., Shams Ardekani M.R., Akbarzadeh T., Hadjiakhoondi A. and Khanavi M. (2012) Phytochemical study and *in vitro* cytotoxic effect of *Ajuga chamaecistus* ssp. *tomentella*. *Asian Journal of Chemistry* 24(7), 2871-2874.

Abstract: In this study total methanolic extract (80 %), n-hexane, diethyl ether, hydroalcoholic fractions and three major compounds from aerial parts of *Ajuga chamaecistus* ssp. *tomentella* were investigated for *in vitro* cytotoxic effect against cancer (HT-29, Caco-2, T47D) and normal (NIH 3T3) cell lines by the MTT assay. The n-hexane fraction showed cytotoxicity against all cell lines with IC 50 \geq 200 μ g/mL and the diethyl ether fraction exhibited medium cytotoxic effect against HT-29 cell line (IC 50 311.01 \pm 9.0 μ g/mL). The diethyl ether fraction was chromatographed on silica gel using a chloroform-ethyl acetate-methanol gradient system to give compound 1, 2 and 3. The structure of compound 1, 2 and 3 were determined to be 20-hydroxyecdysone, cyasterone and 8-acetylharpagide, respectively, by means of spectroscopic analysis. These three major compounds were inactive in cytotoxicity evaluation (IC 50 > 400 and 800 μ g/mL), suggesting little correlation between the degree of cytotoxic effect of the diethyl ether fraction and the isolated compounds.

Sadati S.N., Jahantab S., Ziyarati P., Khanavi M. and Shams-Ardekani M.R. (2016) Quantification of 20-hydroxyecdysone, a major phytoecdysteroid, in *Ajuga chamaecistus* ssp. *tomentella* using high-performance liquid chromatography. *Traditional and Integrative Medicine* 1(3), 96-100.

Abstract: The genus *Ajuga* is used for the treatment of joint pain, gout, jaundice, and wound healing in Iranian traditional medicine. *Ajuga chamaecistus* ssp. *tomentella* is an exclusive subspecies of *Ajuga chamaecistus* in the flora of Iran. Plants belong to *Ajuga* species are advantageous sources of phytoecdysteroids. 20-hydroxyecdysone (20E) is an important phytoecdysteroid with anabolic property. This study aimed to determine and quantify 20E in methanolic extract of aerial parts of *Ajuga chamaecistus* ssp. *tomentella*. The standard reference of 20E was isolated from n-butanolic fraction of aerial parts of *Ajuga chamaecistus* ssp. *tomentella* using high-performance liquid chromatography (HPLC). The analysis was carried out on an ODSA (C18) column with isocratic elution using water-acetonitrile (75:25). The standard calibration curve represented good linearity ($r^2 = 0.9997$). The limit of quantification (S/N = 10) and detection (S/N = 3) were determined as 7.93 and 2.38 μ g/ml, respectively. The content of 20E in methanolic extract of *Ajuga chamaecistus* ssp. *tomentella* was determined to be 2.58% (w/w) (0.46% in dry plant). The quantitative proportion of the 20E found in *Ajuga chamaecistus* ssp. *tomentella* extract proposes the possible uses of this plant in commercial formulations or as a source of 20E.

Sadikov S.T. and Saatov Z. (1998) Phytoecdysteroids of plants of the genus *Silene* XIX. Structure of silenoid G. *Khimiya Prirodnykh Soedinenii* (5), 663-665 [in Russian].

Sadikov Z.T. and Saatov Z. (1999) Phytoecdysteroids of plants of the genus *Silene* - XX. Integristerone A 25-acetate from *Silene brahuica*. *Khimiya Prirodnykh Soedinenii* (4), 492-493 [in Russian]/*Chemistry of Natural Compounds* 35(4), 440-441 [in English].

Sadikov Z.T., Saatov Z., Girault J.-P. and Lafont R. (2000) Silenoid H, a new phytoecdysteroid from *Silene brahuica*. *Journal of Natural Products* 63(7), 987-988.

Abstract: Silenoid H (1), a new phytoecdysteroid, has been isolated from the roots of *Silene brahuica* and identified as 22-O- α -D-galactosylintegristerone A 25-acetate by MS and NMR analysis.

Sadikov Z., Saatov Z., Garcia M., Girault J.-P. and Lafont R. (2001) Ecdysteroids from *Silene claviformis*. *Khimiya Prirodnykh Soedinenii* (3), 223-225 [in English]. Correction: *Chemistry of Natural Compounds* 37(6), 580 (2001).

Sadykov Z.T., Ramazanov N.S. and Saatov Z. (1997) Phytoecdysteroids of *Rhaponticum* plants - polypodine B-22-O-benzoate from *Rhaponticum carthamoides*. *Khimiya Prirodnykh Soedinenii* (6), 851-853 [in Russian].

Saeng-ngam S., Juntawong N., Vajaroathai S. and Visetson S. (2004) Comparative study of moulting hormone content in different plant species. Proceedings of the 42nd Kasetsart University Annual Conference, Kasetsart,

Thailand, 3-6th February, 2004, p. 284-290 (on-line at: <http://kucon.lib.ku.ac.th/FullText/KC4201035.pdf>) [in Thai, with English Abstract].

Sagdullaev S.S., Sultanov S.A., Nigmatullaev A.I., Saatov Z. and Mamatkhanov A.U. (1999) *Rhaponticum integrifolium* as a producer of ecdysteroids. *Khimiya Prirodnykh Soedinenii* (2), 245-247 [in Russian]/*Chemistry of Natural Compounds* 35(2), 219-220 [in English].

Sakimbai A.D., Baizhigit Y.A., Mazhikenova G.M., Tyanakh S., Kozhanova A.M., Minayeva Y.V., Tulenov B.I. and Adekenov S.M. (2019) *Kopia scoparia* is a new source of 2-deoxyecdysone. XIIIth International Symposium on the Chemistry of Natural Compounds, October 16th-19th, 2019, Shanghai, China, p191 [abstract].

Saleem M., Musaddiq S., Riaz N., Zubair M., Ashraf M., Nasar R. and Jabbar A. (2013) Ecdysteroids from the flowers of *Aerva javanica*. *Steroids* 78 1098–1102.

Abstract: Four new ecdysteroids (1-4), along with three known steroids, β -ecdysone (5), 5- β -2-deoxyintegristerone A (6) and 24-epi-makisterone A (7) (Fig. 1), were isolated from the methanolic extract of the flowers of *Aerva javanica* by using normal and reverse phase chromatography. The structures of the new compounds (1-4) were determined due to 1D ((1)H and (13)C), 2D NMR (HSQC, HMBC, COSY, NOESY) techniques and high resolution mass spectrometry (HREIMS). The known compounds (5-7) were characterized based on the 1D NMR spectroscopy and mass spectrometry and by comparison with the literature values. All isolates were evaluated for their inhibitory activities against enzymes acetylcholinesterase (AChE), butyrylcholinesterase (BChE) and lipoxygenase (LOX).

Sánchez W.E., Brown K.S., Nishida T., Durham L.J. and Duffield A.M. (1970) Hydrophilic chemical constituents of *Podocarpus sellowii* Klotzsch. *An. Acad. brasil Ciénc.* 42 (Supplement), 77-85.

Santos A.C., Chua M.T., Eufemio N. and Abela C. (1970) Isolation of commisterone, a new phytoecdysone from *Cyanotis vaga*. *Experientia* 26(10), 1053-1054.
No abstract.

Santos A.C., Chua M.T., Eufemio N., Abela C., Hikino H. and Takemoto T. (1972) Identity of commisterone with ecdysterone. *Planta Medica* 21(3), 279-281.

Abstract: An insect moulting hormone commisterone which has been isolated from the leaves of *Cyanotis vaga* (*Commelinaceae*) has been identified as ecdysterone.

Sardini D. and Krepinsky J. (1974) La determinazione densitometrica degli ecdisoni [Densitometric determination of ecdysones]. *Il Farmaco Edizione Pratica* 29(12), 723-731 [in Italian, with an English abstract].

Sarker S.D., Girault J.-P., Lafont R. and Dinan L.N. (1996a) Ecdysteroids from *Gomphrena haageana* (Amaranthaceae). *Biochemical Systematics and Ecology* 24, 177-178.

Sarker S.D., Dinan L.N., Girault J.-P., Lafont R. and Waterman P.G. (1996b) Punisterone ([20R,24S]-25-deoxy-11 α ,20,24-trihydroxyecdysone): a new phytoecdysteroid from *Blandfordia punicea*. *Journal of Natural Products* 59, 789-793.

Abstract: A new phytoecdysteroid, punisterone [(20R,24S)-25-deoxy-11 α ,20,24-trihydroxyecdysone] (1), together with six known ones, ecdysone, 20-hydroxyecdysone, 5 β ,20-dihydroxyecdysone, ponasterone C, pterosterone, and turkesterone, have been isolated from the seeds of *Blandfordia punicea*.

Sarker S.D., Savchenko T., Whiting P., Lafont R. and Dinan L.N. (1996c) The genus *Ourisia* (Scrophulariaceae): a potential source of phytoecdysteroids. *Biochemical Systematics and Ecology* 24(7/8), 803-804.

Sarker S.D., Savchenko T., Whiting P., Šik V., Lafont R. and Dinan L. (1997a) Occurrence of ecdysteroids in the genus *Centaurea* (Compositae): 20-hydroxyecdysone from *Centaurea moschata*. *Biochemical Systematics and Ecology* 25(4), 367-368.

Sarker S.D., Girault J.-P., Lafont R. and Dinan L.N. (1997b) Ecdysteroid xylosides from *Limnanthes douglasii*. *Phytochemistry* 44(3), 513-521.

Abstract: Bioassay/RIA-guided phytochemical examination of seeds of members of the genus *Limnanthes* demonstrates the presence of moderate to high levels of phytoecdysteroids. *Limnanthes douglasii* has afforded two novel ecdysteroid glycosides; limnantheoside A [20-hydroxyecdysone-3- β -D-xylopyranoside] and limnantheoside B [ponasterone A-3- β -D-xylopyranoside], together with 20-hydroxyecdysone and ponasterone A. HPLC/RIA data support the additional presence of small amounts of ecdysone and an analogous

ecdysone glycoside. The levels of ecdysteroids in individual seeds and plants of *L. douglasii* are very variable. The HPLC behaviour of ecdysteroid glycosides, including these two new xylosides, in both RP- and NP-systems using different mobile phases is also discussed.

Sarker S.D., Savchenko T., Whiting P., Šik V., Rees H.H. and Dinan L. (1997c) Analysis of species of the Ranunculaceae for ecdysteroid agonists and antagonists I: ecdysteroids in the genus *Pulsatilla*. *Biochemical Systematics and Ecology* 25(5), 473-474.

Sarker S.D., Savchenko T., Šik V., Rees H.H. and Dinan L. (1998a) 20-Hydroxyecdysone and its glucosides from *Trisetum flavescens*. *Biochemical Systematics and Ecology* 26(1), 135-137.

Sarker S.D., Savchenko T., Whiting P., Šik V. and Dinan L. (1998b) Ecdysteroids from the seeds of *Trianthema turgidifolia* and *T. pilosa* (Aizoaceae). *Biochemical Systematics and Ecology* 26, 691-693.

Sarker S.D., Šik V., Rees H.H. and Dinan L. (1998c) 1 α ,20R-Dihydroxyecdysone from *Axyris amaranthoides*. *Phytochemistry* 49(8), 2305-2310.

Abstract: Bioassay/RIA-directed phytochemical examination of the seeds of *Axyris amaranthoides* afforded a new ecdysteroid: 1 alpha,20R-dihydroxyecdysone [1-epi-integristerone A], together with 20-hydroxyecdysone and polypodine B. The structure of 1 alpha,20R-dihydroxyecdysone was determined unequivocally by UV, LSIMS, and a combination of 1D and 2D NMR techniques.

Sarker S.D., Šik V., Rees H.H. and Dinan L. (1998d) 2-Dehydro-3-*epi*-20-hydroxyecdysone from *Froehlichia floridana*. *Phytochemistry* 49(8) 2311-2314.

Abstract: A new phytoecdysteroid, 2-dehydro-3-*epi*-20-hydroxyecdysone, together with 20-hydroxyecdysone have been isolated by bioassay/RIA-directed HPLC analyses of a methanol extract of the seeds of *Froehlichia floridana*. The structure of the novel ecdysteroid was determined unambiguously by UV, LSIMS, and a combination of 1D and 2D NMR techniques. The biological activity in the *Drosophila melanogaster* B₁₁ cell bioassay (ED₅₀ = 4.0 x 10⁻⁷ M) is considerably lower than that of 20-hydroxyecdysone (ED₅₀ = 7.5 x 10⁻⁹ M).

Sarpong F.M. (2012) Chemical constituent(s), anti-inflammatory, antioxidant and anti-nociceptive activities of the roots of *Palisota hirsuta* K. Schum (Commelinaceae). Ph.D. Thesis, Kwame Nkrumah University of Science and Technology, Kumasi, pp 156.

Sarpong F.M., Armah F.A., Amponsah I.K. and Atchgolo P.K. (2016) Antinociceptive ecdysteroids and other constituents of *Palisota hirsuta* K. Schum (Comelinaceae). *Journal of Applied Pharmaceutical Science* 6(10), 147-153.

Abstract: The root of *Palisota hirsuta* is used in Ghana and other West African countries for the treatment of various disease conditions such as rheumatoid arthritis (also in other inflammatory and painful conditions), infertility in females, anaemia, and dysentery. The current study sought to evaluate the anti-nociceptive effect of the hydroethanolic root extract and compounds isolated from *Palisota hirsuta*. Mice were used for studying the antinociceptive activity of *P. hirsuta* extracts and isolates at doses of 10-300 mg/kg-p.o and 10-30 mg/kg⁻¹ respectively using the Formalin Induced paw licking model. The total crude extract, methanolic and petroleum ether fractions showed analgesic activity in a dose-dependent manner for both the early and late phases. Three isolates, a fatty acid mixture, 20-hydroxyecdysone and an uncharacterized ecdysteroid (PH V) were obtained from the methanol fractions with significant activity. 20-hydroxyecdysone exhibited a dose-dependent inhibition of the nociception for both the early and late phases; 71.39±9.19% and 89.19±3.81% respectively. PH V showed significant activity between the early and late phases of inhibition as compared to the reference drug morphine. The present study has given scientific credence to the use of the roots of *P. hirsuta* for the mitigation of pain and established its antinociceptive constituents.

Sauer H.H., Bennett R.D. and Heftmann E. (1968) Ecdysterone biosynthesis in *Podocarpus elata*. *Phytochemistry* 7, 2027-2030.

Abstract: The insect molting hormone ecdysterone was isolated in radioactive form after administration of cholesterol-4-¹⁴C to *Podocarpus elata* seedlings. Cholestenone-4-¹⁴C, however, was not significantly incorporated into ecdysterone by this plant. The biosynthetic implications of these results are discussed.

Sautour M., Canon F., Miyamoto T., Dongmo A. and Lacaille-Dubois M.A. (2007) Ecdysteroids from *Dioscorea dumetorum*. *Planta Medica* 73, DOI: 10.1055/s-2007/987198.

Abstract: Among the yam species commonly grown and consumed in Cameroon, *Dioscorea dumetorum* (Kunth) Pax (Dioscoreaceae) is the most important one [1]. The tuber of this plant is used in traditional medicine for the

treatment of diabetes and crude extracts of *D. dumetorum* have been shown to possess a hypoglycemic effect in fasted normal rats and rabbits [2]. As a part of our ongoing studies of the plants from Dioscoreaceae [3,4], a phytochemical investigation of the rhizome of *D. dumetorum* has led to the isolation of a new ecdysteroid, 5,11,20-trihydroxyecdysone (**1**), and two known ecdysteroids, kerkesterone (**2**) and ajugasterone C (**3**), by several chromatographic steps on normal and reversed phase silica gel. Their structures were determined by spectroscopic methods including 1D- and 2D-NMR (COSY, TOCSY, HSQC and HMBC). This is the first report on the occurrence of phytoecdysteroids in the Dioscoreaceae family. These compounds were devoid of antifungal activity against three *Candida* species (*C. albicans*, *C. glabrata* and *C. tropicalis*, MIC >200 µg/ml).

Sautour M., Canon F., Miyamoto T., Dongmo A. and Lacaille-Dubois M.-A. (2008) A new ecdysteroid and other constituents from two *Dioscorea* species. *Biochemical Systematics and Ecology* doi: 10.1016/j.bse.2008.03.002.

Abstract: Phytochemical investigation of the rhizome of *Dioscorea dumetorum* has led to the isolation by several chromatographic steps on normal and reversed phase silica gel of a new ecdysteroid, (20R)-5 β ,11 α ,20-trihydroxyecdysone (**1**), and two known ecdysteroids, ajugasterone C (**2**) and herkesterone (**3**). Their structures were determined by spectroscopic methods including 1D- and 2D-NMR (COSY, TOCSY, HSQC and HMBC). This is the first report on the occurrence of phytoecdysteroids in the Dioscoreaceae family. These compounds were devoid of antifungal activity against three *Candida* species (*Candida albicans*, *Candida glabrata* and *Candida tropicalis*, MIC > 200 µg/ml).

Savchenko T., Whiting P., Sarker S.D. and Dinan L. (1997) Phytoecdysteroids in the genus *Agapanthus* (Alliaceae). *Biochemical Systematics and Ecology* 25(7), 623-629.

Abstract: Bioassay/RIA-based analysis of seeds of 15 species, subspecies and varieties of *Agapanthus* revealed the presence of phytoecdysteroids in most of them. However, the concentrations varied markedly between samples. The accumulation of phytoecdysteroids was highest in a sample of *A. campanulatus* ssp. *patens* and a sample of *A. inapertus* ssp. *hollandii* (c. 1.5 mg ecdysone equivalents per g seed, using the DBL-1 antiserum). Moderate levels were present in a sample of *A. inapertus* ssp. *inapertus*. Phytoecdysteroid levels were almost undetectable in samples of *A. coddii*, *A. caulescens* ssp. *angustifolius*, *A. minor* var. *alba* and *A. praecox* ssp. *orientalis*. RP-HPLC separation coupled with monitoring by RIA and bioassay revealed that the samples contain significantly different ecdysteroid profiles. The major phytoecdysteroid in several of the extracts has been identified as 20-hydroxyecdysone on the basis of chromatographic evidence. It is concluded that ecdysteroid levels and profiles vary considerably in seeds of the genus *Agapanthus*, even between seed samples of the same species.

Savchenko T., Whiting P., Sarker S.D. and Dinan L. (1998a) Analysis of species of the Ranunculaceae for ecdysteroid agonists and antagonists - II. Ecdysteroids in the genus *Anemone*. *Biochemical Systematics and Ecology* 26(1), 131-134.

No Abstract.

Savchenko T., Whiting P., Sarker S.D. and Dinan L. (1998b) Distribution and identity of phytoecdysteroids in *Gomphrena* spp. (Amaranthaceae). *Biochemical Systematics and Ecology* 26(3), 337-346.

Savchenko T., Whiting P., Šik V., Underwood E., Sarker S.D. and Dinan L. (1998c) Distribution and identities of phytoecdysteroids in the genus *Briza* (Gramineae). *Biochemical Systematics and Ecology* 26(7), 781-791.

Abstract: Within the genus *Briza*, phytoecdysteroids are predominantly associated with native Eurasian species, while none of the native South American species tested was found to accumulate detectable levels of ecdysteroids in their seeds. Bioassay/RIA-guided HPLC analysis led to the isolation of six known ecdysteroids (abutasterone, ecdysone, 20-hydroxyecdysone, polypodine B, pterosterone and sidisterone) from the seeds of *Briza maxima*. The structures of these compounds were elucidated by UV, EIMS and a series of 1D and 2D NMR experiments, notably ¹H NMR, ¹³C PENDANT, COSY 45, HMBC, HMQC and NOESY. The distributions of ecdysteroids present in different plant parts of *Briza* spp. were analysed by RIA. Those species containing detectable levels of ecdysteroids in the seeds (*B. elatior*, *B. maxima*, *B. media* and *B. minima*) possessed even higher levels of ecdysteroids in the plants, while the other species, with undetectable ecdysteroid levels in the seeds, possessed very low (probably biologically insignificant) levels in the plants. The highest levels of ecdysteroids are associated with meristematic region at the root/aerial interface in plants of *B. elatior*.

Savchenko T., Whiting P., Germade A. and Dinan L. (2000) Ecdysteroid agonist and antagonist activities in species of the Solanaceae. *Biochemical Systematics and Ecology* 28(5), 403-419.

Abstract: Previously, it has been shown that certain withanolides from *Ichroma gesnerioides* (Solanaceae) possess ecdysteroid antagonistic activity. Phytoecdysteroids (agonists) are widely distributed in the plant world, but solanaceous species have not been extensively examined for their presence. We have now surveyed 128 species of solanaceous plants for the presence of ecdysteroid agonist and antagonist activities using the *Drosophila*

melanogaster B_{II} cell line bioassay. Only weak antagonistic activity was associated with a few of the methanolic extracts, including those from species known to contain high levels of withanolides. Therefore, the major withanolides are inactive per se, but they may be activated after ingestion by invertebrate predators. Several extracts possessed ecdysteroid agonist activity as a consequence of the presence of phytoecdysteroids. Phytoecdysteroid-accumulating species are at least as common in the Solanaceae as they are in plants in general. Preliminary characterization of the identities of the phytoecdysteroids present in the most active extracts has been performed by hplc separations on normal- and reversed-phase systems in conjunction with ecdysteroid-specific radioimmunoassay and bioassay. Each of the phytoecdysteroid-accumulating species examined (*Browallia speciosa*, *Nierembergia hippomanica* var *violacea*, *N. solanacea* and *Solanum nigrum*) contain a cocktail of ecdysteroids, of which 20-hydroxyecdysone and polypodine B (5 β ,20-dihydroxyecdysone) are major components.

Savchenko T., Blackford M., Sarker S.D. and Dinan L. (2001) Phytoecdysteroids from *Lamium* spp: identification and distribution within plants. *Biochemical Systematics and Ecology* **29**(9), 891-900.

Abstract: Bioassay/radioimmunoassay (RIA) analysis of the seeds of four *Lamium* species, *L. album*, *L. galeobdolon*, *L. maculatum* and *L. purpureum* revealed the presence of phytoecdysteroids in all of them. Bioassay/RIA-guided and photo-diode array-monitored HPLC analysis of the aerial parts of *L. album* and *L. purpureum* led to the isolation of four known ecdysteroids (abutasterone, inokosterone, polypodine B and pterosterone) from the former, and 20-hydroxyecdysone from the latter. Distribution and identities of ecdysteroids in different parts of these two species and also in the seed extract of *L. maculatum* have been analysed by RIA and bioassay.

Schmelz E.A. (1999) The role of phytoecdysteroids in spinach (*Spinacia oleracea*): physiological responses to below ground herbivory support a plant defense hypothesis. Ph.D. Thesis, University of Arizona.

Schmelz E.A., Grebenok R.J., Galbraith D.W. and Bowers W.S. (1998) Damage-induced accumulation of phytoecdysteroids in spinach: rapid root response involving the octadecanoic acid pathway. *Journal of Chemical Ecology* **24**(2), 339-360.

Abstract: Some plant defenses are known to be rapidly induced following attack by phytophagous insects. Plant-produced insect molting hormones, termed phytoecdysteroids, are believed to aid plant resistance; however, their dynamics are poorly understood. Using spinach (*Spinacia oleracea*) as a model system, we examined the inducibility of phytoecdysteroids, primarily 20-hydroxyecdysone (20E), in an effort to characterize potential interactions with herbivorous insects. Rapid phytochemical induction was investigated using damage treatments and applications of defense-related plant-signal analogs, specifically methyl jasmonate (MJ) and methyl salicylate (MSA). Within two days, mechanically damaged roots exhibited two to three fold increases in phytoecdysteroid concentrations. Four days after root damage, small increases in shoot levels were also detectable. Unlike roots, foliar 20E concentrations were unaltered over a range of shoot treatments including insect herbivory (*Spodoptera exigua*), mechanical damage, and MJ applications. Additions of MJ (12.5–50 μ g/liter) to the root systems of hydroponically grown plants stimulated accumulations of root phytoecdysteroids in a dose-dependent manner, similar in magnitude to the response induced by root damage. Under identical conditions, MSA did not affect the accumulation of 20E when added to the hydroponic solutions of undamaged plants. Moreover, MSA inhibited the induction of 20E in wounded roots, but did not interfere with the action of applied MJ. In contrast to mechanical damage, roots did not induce 20E levels when challenged with two different fungal pathogens (*Pythium aphanidermatum* and *Phytophthora capsici*). We propose that wound-induced accumulations of 20E are generated in the roots, signaled via endogenous jasmonates, and may confer enhanced resistance against subterranean herbivorous insects.

Schmelz E.A., Grebenok R.J., Galbraith D.W. and Bowers W.S. (1999) Insect-induced synthesis of phytoecdysteroids in spinach, *Spinacia oleracea*. *Journal of Chemical Ecology* **25** (8), 1739-1757.

Abstract: Spinach (*Spinacia oleracea*) foliage is known to synthesize and accumulate insect molting hormones, predominantly in the form of 20-hydroxyecdysone (20E). We previously demonstrated that root 20E accumulation is increased following root damage. We designed two further experiments to address root responses to both mechanical and insect damage. In plants grown hydroponically, removal of 35% or less of the root mass did not result in changes in root 20E levels. However, removal of 70% of the root mass stimulated 6.0- and 1.5-fold increases in the root and shoot 20E concentrations, respectively. The effects of insect damage on soil-grown plants were investigated by infesting plant roots with black vine weevil (BVW: *Otiiorhynchus sulcatus*) larvae and allowing them to feed for seven days. Decreases in root mass occurred in young plants; however, no changes were detected in mature plants. In all cases, root herbivory resulted in at least a 3.0-fold increase in root 20E concentrations. Our previous experiments implicated jasmonic acid and the analog methyl jasmonate (MJ) in signaling the damage-induced accumulation of root 20E levels. We investigated the activity of other phytohormones and growth regulators (GRs) on the 20E accumulation patterns of young plants as a means of examining the significance of jasmonates in the induction response. Hydroponic additions of MJ (0.5 μ M) and the synthetic auxin, 1-naphthaleneacetic acid (NAA; 0.5 μ M),

resulted in significant increases in root 20E levels. At the concentrations tested, indole-3-acetic acid (IAA), gibberellic acid (GA₃), abscisic acid (ABA), and *trans*-zeatin (Z) had no effects on root 20E concentrations. However, both NAA (0.5–5.0 μM) and Z (5.0 μM) treatments caused increases in the root/shoot dry mass ratios, indicating shifts in resource allocation to the roots. Treatments involving ABA (5.0 μM) and Z (0.5–5.0 μM) caused significant increases in shoot 20E concentrations. No other hormone treatments altered shoot accumulation patterns. The mechanisms underlying the root 20E induction phenomena were investigated through the incorporation of [2-¹⁴C]mevalonic acid ([¹⁴C]MVA). Within one day, excised roots readily incorporated radioactivity into 20E from [¹⁴C]MVA. In intact plants, [¹⁴C]MVA absorbed by the roots was rapidly incorporated into root 20E pools following damage and MJ treatments. This implies that the wound-induced root 20E accumulation is the result of increased *de novo* 20E synthesis in the root.

Schmelz E.A., Grebenok R.J., Ohnmeiss T.E. and Bowers W.S. (2000) Phytoecdysteroid turnover in spinach: long-term stability supports a plant defense hypothesis. *Journal of Chemical Ecology* **26**(12), 2883-2896.

Abstract: Using short (8-day) and long-term (28-day) experiments, we examined the stability of 20-hydroxyecdysone (20E) and the dominant phytosterols synthesized from a pulse of [2-¹⁴C]mevalonic acid ([¹⁴C]MVA) in hydroponically grown spinach (*Spinacia oleracea*). In the short-term experiment, plant dry mass and shoot 20E pools steadily increased. Root uptake of [¹⁴C]MVA resulted in the stable incorporation of ¹⁴C radiolabel into whole plant 20E pools, with no significant changes over time. Levels of free and saponifiable phytosterols increased in the shoots while ¹⁴C-labeled shoot phytosterols remained constant. Unexpectedly, both ¹⁴C-labeled and unlabeled pools of root phytosterols decreased over time. In the long-term experiment, plant dry mass and shoot 20E levels increased over time, while total ¹⁴C-labeled 20E pools remained constant. Both root and shoot phytosterol pools increased over time while the ¹⁴C incorporation in these pools remained constant. Together these experiments indicate that 20E in spinach is metabolically stable and thus shares this characteristic with plant terpenoids of known defensive function. While little is known about phytosterol turnover in plants, our results suggest that phytosterols can indeed exist in a very dynamic state but may also be stable over time.

Schmelz E.A., Grebenok R.J., Ohnmeiss T.E. and Bowers W.S. (2002) Interactions between *Spinacia oleracea* and *Bradysia impatiens*: a role for phytoecdysteroids. *Archives of Insect Biochemistry and Physiology* **51**, 204-221.

Abstract: Plant produced insect molting hormones, termed phytoecdysteroids (PEs), are thought to function as plant defenses against insects by acting as either feeding deterrents or through developmental disruption. In spinach (*Spinacia oleracea*), 20-hydroxyecdysone (20E) concentrations in the roots rapidly increase following root damage, root herbivory, or methyl jasmonate (MJ) applications. In this inducible system, we investigated the plant defense hypothesis by examining interactions of roots, 20E concentrations, and larvae of the dark-winged fungus gnat (*Bradysia impatiens*). Root herbivory by *B. impatiens* larvae resulted in a 4.0- to 6.6-fold increase in root 20E concentrations. In paired-choice tests, increases in dietary 20E stimulated *B. impatiens* feeding deterrence. *B. impatiens* larvae preferred control diets, low in 20E, to those constructed from induced roots and those amended with 20E (25 to 50 micro g/g wet mass). When confined to 20E-treated diets, concentrations as low as 5 micro g/g (wet mass) resulted in significantly reduced *B. impatiens* survivorship compared to controls. The induction of root 20E levels with MJ resulted in a 2.1-fold increase in 20E levels and a 50% reduction in *B. impatiens* larval establishment. In a paired-choice arena, untreated control roots were damaged significantly more by *B. impatiens* larvae than MJ-induced roots that contained 3-fold greater 20E levels. Based on dietary preference tests, the 20E concentrations present in the MJ-induced roots (28 micro g/g wet mass) were sufficient to explain this reduction in herbivory. Interactions between spinach roots and *B. impatiens* larvae demonstrate that PEs can act as inducible defenses and provide protection against insect herbivory.

Schooley D.A., Weiss G. and Nakanishi K. (1972) A simple and general extraction procedure for phytoecdysones based on reversed-phase adsorption chromatography. *Steroids* **19**(3), 377-383.

Seliverstova A.A., Zibareva L.N. and Eremina V.I. (2014) Patterns of phytoecdysteroid distribution in the plants of the section *Otites* of the genus *Silene* L.: chemotaxonomic approach. *Tomsk State University Journal of Biology* **27**(3), 101-114 [in Russian, with an abstract in English].

Abstract: Species of the genus *Silene* L. (Caryophyllaceae) contain a big quantity of biologically active substances such as phenylpropanoids, triterpene saponins and phytoecdysteroids. Many *Silene* species have successfully adapted to different climatic conditions while maintaining the ability to biosynthesis of biologically active substances. But classification of the genus is an enough challenging task for today. To resolve problems of systematization, authors recommend applying not only ecology-geographical and anatomy-morphological criteria, but also a complex biochemical approach. The purpose of this work is chemotaxonomic studying of some species of sections *Otites* of the genus *Silene* and their composition of ecdysteroids.

We conducted a chemotaxonomic study of some species of the genus *Silene* and compared different systematics of *Otites* section described in national and international literature. It is shown that there is currently no single view of classification of species, both in individual sections and in genus. We considered the tendency of section *Otites* separation in the genus *Otites* Adans and isolated and studied the phytoecdysteroids *Silene colpophylla* Wrigley, *Silene sendtneri* Boiss. and *Silene otites* Wibel., *S. pseudotites* Besser. ex Reichenb., *S. colpophylla*, *S. sendtneri* и *S. roemerii*. These species are introduced into Siberian Botanical Garden of Tomsk State University. Plants of all specified species go through the whole vegetative cycle and reach the generative stage on the second year of life. At the end of the second year, viable seeds are formed. Flowering begins in the second half of June - the beginning of July and comes to an end in the beginning of August. We analyzed the ecdysteroid profile of *S. colpophylla* by HPLC and HPLC/MS methods. It is revealed that the given species synthesizes 14 ecdysteroids. Six ecdysteroids of the species were identified: 20-hydroxyecdysone, polygodine B, ecdysone, 2-deoxy-20-hydroxyecdysone, 2-deoxyecdysone and integristerone A. We detected the basic patterns of ecdysteroids distribution characteristic of some species (*S. sendtneri*, *S. roemerii*, *S. otites*, *S. pseudotites*, *S. colpophylla*) of the section and analyzed the occurrence of different 26-oxyderivative ecdysteroids in some species of the genus for section *Otites*.

Selvaraj P., de Britto A.J. and Sahayaraj K. (2005) Phytoecdysone of *Pteridium aquilinum* (L) Kuhn (Dennstaedtiaceae) and its pesticidal property on two major pests. *Archives of Phytopathology and Plant Protection* 38(2), 99-105.

Abstract: Preliminary phytochemical and HPLC analyses of *Pteridium aquilinum* (L) Kuhn (Pteridophyceae) was carried out in the Chloroform (CE) and ethanol extracts (EE) and also the hexane (HF) and aqueous fractions (AF) obtained during the process of phytoecdysteroid separation and crude phytoecdysteroids fraction (CEF). Except in aqueous fraction, all other fraction consists of steroids. Alkaloids are absent in all the extracts. Saponins and xanthoproteins were present in CE, EE and AF but were absent in HF and CEF. Tannins and flavonoids are present in EE and AF and were absent in CE, EE and CEF. HPLC analysis of *P. aquilinum* has both α and β ecdysones. CE recorded β -ecdysone having a Rt value 2.46 (81.3%) and 2.49 (18.6%). In addition to the β ecdysone the EE and CEF contains α ecdysone [RT=1.52 (30.96%) and 0.85 (63.0%)]. Among the three extracts tested, EE was the most effective, followed by CEF. Of the two experimental animals, *Helicoverpa armigera* showed more susceptibility to the fern extracts than *Spodoptera litura*. The different extracts of the same plant showed differences in their toxic as well as growth disrupting responses. The order of concentration required for 50% mortality (LD50) of *H. armigera* was 0.198, 0.112 and 0.198 percentages for CE, CEF and EE. As observed in *H. armigera*, *S. litura* also recorded the highest lethal activity in EE (0.141%) followed by 0.128% of CEF and 0.160% of CE.

Sena Filho J.G., Durringer J., Maia G.L.A., Tavares J.F., Xavier H.S., Sobral da Silva M., da Cunha E.V.L. and Barbosa-Filho J.M. (2008) Ecdysteroids from *Vitex* species: distribution and compilation of their ¹³C-NMR spectral data. *Chemistry & Biodiversity* 5, 707-713.

Abstract: Iridoids and ecdysteroids are found in some genera of the family Verbenaceae. In such cases, they are used as chemotaxonomic markers for the difficult task of taxonomic identification by using morphological characteristics of plants belonging to this family. The present work describes the distribution of ecdysteroids in plants from the genus *Vitex* from a review of previous work on seventeen *Vitex* species. In addition, (¹³)C-NMR data of the main ecdysteroids found in this genus are described. This study attempted to summarize previous research on ecdysteroids distribution in *Vitex* species with the addition of (¹³)C-NMR analysis to further refine the characterization of these compounds in the Verbenaceae family.

Serra L.Z., Felipe D.F. and Cortez D.A.G. (2012) Quantification of β -ecdysone in different parts of *Pfaffia glomerata* by HPLC. *Revista Brasileira de Farmacognosia/Brasilian Journal of Pharmacognosy* 22(6), 1349-1354.

Abstract: *Pfaffia glomerata* (Spreng.) Pedersen, Amaranthaceae, is widely distributed in Brazil. Roots are considered as the world's greatest supplier and β -ecdysone is the most important compound extracted from roots of *Pfaffia glomerata*. So, the aim this study was analyze the presence of β -ecdysone in the inflorescences and stems and compared with the content from roots of *Pfaffia glomerata* and determine the best extractive method of β -ecdysone this plant. The crude extracts were obtained by Soxhlet method, reflux, maceration, percolation and turbolyse. Compound extracts were quantified by High Performance Liquid Chromatography (HPLC). The analysis were carried out a Phenomenex Column C18, 5 μ m, 250x4,6mm, maintained at 30 °C, gradient system using as mobile phase a mixture of methanol and water, flow rate 1,0 mL and detection at 245 nm. Results showed Soxhlet method with ethanol:water (90:10 v/v) presented the higher concentration of β -ecdysone in *P. glomerata* and inflorescences

showed higher amount of this active substance (3,06%), compared with stems (2,37%) and roots (1,63%), showing that the inflorescences and plant stems may also be used as a rich source of β -ecdysone.

Shah V.C. and de Souza N.J. (1971) Amaranthaceae: Ecdysterone from *Cyathula prostrata*. *Phytochemistry* **10**, 1398-1399.

Abstract: Ecdysterone has been found to be the major phytoecdysone *Cyathula prostrata* and is present to an extent of 005% of the dry plant.

Sharma B., Yadav A. and Dabur R. (2019) Interactions of a medicinal climber *Tinospora cordifolia* with supportive interspecific plants trigger the modulation in its secondary metabolic profiles. *Scientific Reports* **9**, 14327 (doi.org/10.1038/s41598-019-50801-0).

Abstract: *Tinospora cordifolia* (TC) is scientifically proven immunomodulatory drug being used for centuries. Ancient literature reported that inter-specific interactions change medicinal properties of TC. Thus, the current study is aimed to understand the influence of interspecific biotic interactions on chemo-profiles of TC. To explore it, TC samples collected from six co-occurring plants, i.e. *Azarditchita indica*, *Acacia nilotica*, *Albezia lebbeck*, *Ficus benghalensis*, *Tamarandus indica* and *Acacia leucophloea* were analyzed by HPLC-ESI-QTOF-MS. Mass data were subjected to multivariate analysis. Support vector machines (SVMs) was found to be best classifier (r2

Shi Q., Yan S., Liang M., Yang Y., Wang Y. and Zhang W. (2007) Simultaneous determination of eight components in *Radix Tinosporae* by high-performance liquid chromatography coupled with diode array detector and electrospray tandem mass spectrometry. *Journal of Pharmaceutical and Biomedical Analysis* **43**(3), 994-999. [*Tinospora sagittata* and/or *T. capillipes*].

Abstract: High-performance liquid chromatography (HPLC) coupled with electrospray tandem mass spectrometry (ESI-MS-MS) and diode array detection (DAD) was used to identify and simultaneously determine eight major ingredients in *Radix Tinosporae*. The assay was performed on a Diamonsil C(18) analytical column with a gradient solvent system of A (water containing 0.2% formic acid, 20mM ammonium acetate) and B (methanol/acetonitrile=1/1, v/v). The 217, 248, 270 and 347 nm, respectively, were chosen as the monitoring wavelengths to determine four structural types of components, say columbin, phytoecdysteroids (including 20-hydroxyecdysone, 2-deoxy-20-hydroxyecdysone 3-O-beta-d-glucopyranoside and 2-deoxy-20-hydroxyecdysone), menisperine and protoberberine alkaloids (including columbamine, jatrorrhizine and palmatine). This method was validated in respect to precision, repeatability and accuracy, and was successfully applied to quantify the eight components in 39 batches of *R. Tinosporae* for quality control purpose. The results indicated that the proposed method could be readily utilized as a quality control method for traditional Chinese medicine (TCM).

Shi Q-W., Dong M., Huo C-H., Su X-H., Li X., Yamada T. and Kiyota H. (2007) 7,8 β -Dihydroponasterone A, a new phytosteroid from the needles of the Japanese yew, *Taxus cuspidata*. *Journal of the Brazilian Chemical Society* **18**(5), 1081-1084 (+ Supplementary Information; S1-S2).

Abstract: A new plant ecdysteroid 7,8beta-dihydroponasterone A, together with ponasterone A, were isolated from the methanol extract of the needles of the Japanese yew, *Taxus cuspidata*. Their structures were elucidated on the basis of spectroscopic analysis including ¹H NMR, ¹³C NMR, ¹H-¹H COSY, NOESY, HMQC and HMBC and confirmed by high-resolution FABMS data.

Shin S-L. and Lee C-H. (2011) Effective extraction of phytoecdysteroids from fronds of *Matteuccia struthiopteris* and *Osmunda japonica*. *Korean Journal of Plant Resources* **24**(4), 351-357 [in Korean].

Abstract: This study was carried out to investigate the effective extraction condition for increase of phytoecdysteroids from fronds of *Matteuccia struthiopteris* (FMS) and *Osmunda japonica* (FOJ). Lyophilized fronds were mixed with three different solvents (MeOH, 80% EtOH or water) and then extraction was carried out by using six different methods, such as, immersion (room temp.), heating (60), stirring (200 rpm) for 6 h, or sonication in 42 kHz ultrasonic bath for 15, 30 and 45 minutes. Contents of 20-hydroxyecdysone (20E) and ponasterone A (PonA) were measured by using HPLC after purification of the extracts by cartridge. Altogether, our results indicate that the extraction using sonication with MeOH as a solvent (for 30 minutes) was the most effective condition for 20E and PonA from both MFS and FOJ. Resulting contents of 20E from FMS and FOJ were 66.76 and 104.48 and PonA were 53.43 and 43.82, respectively.

Shiobara Y., Inoue S-S., Nishiguchi Y., Kato K., Takemoto T., Nishimoto N., de Oliveira F., Akisue G., Akisue M.K. and Hashimoto G. (1992) Pfaffane-type nortriterpenoids from *Pfaffia pulverulenta*. *Phytochemistry* **31**(5), 1737-1740.

Abstract: 7-Oxopurveric acid, 7-hydroxypulveric acid and pulverulactone, three new hexacyclic nortriterpenoids, have been isolated from *Pfaffia pulverulenta*. The structures of the new compounds were elucidated as 3,7,11-

trioxopfaffan-12-en-28-oic acid, 7 α -hydroxy-3,11-dioxopfaffan-12-en-28-oic acid and 3 β ,12 α ,13 β -trihydroxypfaffan-28,13-olide, respectively, by chemical transformations and extensive NMR spectral experiments.

Shiobara Y., Inoue S.-S., Kato K., Nishiguchi Y., Oishi Y., Nishimoto N., de Oliveira F, Akisue G., Akisue M.K. and Hashimoto G. (1993) A nortriterpenoid, triterpenoids and ecdysteroids from *Pfaffia glomerata*. *Phytochemistry* **32**(6), 1527-1530.

Abstract: Glomeric acid, a new triterpenoid and pfameric acid, a new nortriterpenoid, have been isolated together with ecdysterone, rubrosterone, oleanolic acid

Shivakumar G.R., Raman K.V.A., Reddy K.V.R., Magadum S.B., Datta R.K., Hussain S.S., Banerji A. and Chowdhary S.K. (1995) Effect of phytoecdysteroids on the larval maturity and economic parameters of the silkworm, *Bombyx mori* L. *Indian Journal of Sericulture* **34**(1), 46-49.

Shoeb M., MacManus S.M., Kumarasamy Y., Jaspars M., Nahar L., Thoo-Lin P.K., Nazemiyeh H. and Sarker S.D. (2006) Americanin, a bioactive dibenzylbutyrolactone lignan, from the seeds of *Centaurea americana*. *Phytochemistry* **67**(21), 2370-2375.

Abstract: The reversed-phase preparative HPLC analysis of the methanol (MeOH) extract of the seeds of *Centaurea americana* afforded a dibenzylbutyrolactone lignan, 3''-O-caffeoyl arctiin (named americanin), together with five known lignans, arctiin, arctigenin, matairesinol, matairesinoside and lappaol A, and two known phytoecdysteroids, 20-hydroxyecdysone and makisterone A. While the structures of the known compounds were determined by direct comparison of the spectral data with published data, the structure of americanin was elucidated by UV, MS and a combination of 1D and 2D NMR spectral analyses. The antioxidant properties and toxicity of the extracts and the isolated compounds were determined by the DPPH and the brine shrimp lethality assays, respectively.

Sidana J., Devi R., Kumar P., Singh B. and Sharma O.P. (2017) Phytoecdysteroid profiling of *Silene vulgaris* by UPLC-ESI-MS. *Current Science* **113**(10), 1986-1992.

Abstract: *Silene vulgaris* is a wild edible plant consumed in both raw as well as cooked forms in several parts of Europe. The phytoconstituents of *Silene* species include phytoecdysteroids, triterpenoidal saponins, terpenoids, flavonoids and phenolics. *Silene vulgaris* is a relatively unexplored species and the chemical profiling of this plant has not been attempted so far. Hence the UPLC-ESI-MS approach was applied to the extracts of flowers, leaves and roots of *S. vulgaris* for the profiling of phytoecdysteroids. The relative distribution of these compounds varied between flowers and leaves; however, the qualitative composition was similar. Only traces of phytoecdysteroids were present in the roots. The aglycones, sugars and other moieties were determined on the basis of ESI-MS. A total of eight previously known phytoecdysteroids were identified. Partial characterization of eight other phytoecdysteroids was also attempted.

Sihra J. (1974) A study of the ecdysone content of some selected East African plants. M.Sc. Thesis, College of Biological and Physical Sciences, University of Nairobi, Kenya.

Silva M. and Bittner M. (1986) Terpenes of *Podocarpus* species from Chile. *Bol. Soc. Chil. Quím.* **31**(1), 19-35.

Silva T.D., Chagas K., Batista D.S., Felipe S.H.S., Louback E., Machado L.T., Fernandes A.M., Buttros V.H.T., Koehler A.D., Farias L.M., Santos A.F., Silva P.O. and Otoni W.C. (2019) Morphophysiological *in vitro* performance of Brazilian ginseng (*Pfaffia glomerata* (Spreng.) Pedersen) based on culture medium formulations. *In Vitro Cellular & Developmental Biology* pp14 (doi: org/10.1007/s11627-019-10003-9).

Abstract: *Pfaffia glomerata* has potential pharmacological and medicinal properties due to the production of a secondary metabolite known as the phytoecdysteroid 20-hydroxyecdysone (20E). There have been increasing efforts for massive *in vitro* propagation of *Pfaffia* plants due to high extractivism and overharvesting of this species. Research on the species has shown that photoautotrophic cultivation can improve the production of 20E. In addition, other abiotic factors such as the formulations of culture media can influence the morphophysiological behavior of the plants *in vitro*. Therefore, the objective of this study was to analyze the morphological and physiological performances of *P. glomerata* plants in different formulations of culture media, under photoautotrophic and photomixotrophic propagation conditions. Six medium formulations, the Driver and Kuniyuki medium (DKW), Correia *et al.* medium (JADS), Murashige and Skoog medium (MS), Quoirin and Lepoivre medium (QL), Rugini medium (OM), and Woody Plant medium (WPM), all supplemented with DKW vitamins, 100 mg L⁻¹ myo-inositol, 6.5 g L⁻¹ agar, and with or without 3% (w/v) sucrose, were evaluated. Cultures were maintained at 25 ± 2°C, with a 16 h-photoperiod under 60 μmol m⁻² s⁻¹ of irradiance under a fluorescent lamp for 50 d. Results showed that the presence or absence of sucrose, and the different nutritional formulations influenced growth, photosynthetic pigment content, endogenous levels of sugars, leaf morphology, levels of 20E, and transport of water and minerals in *P.*

glomerata. Notably, OM, DKW, QL, and WPM media promoted higher production of 20E under photomixotrophic growth conditions.

Silva T.D., Batista D.S., Fortini E.A., Motta de Castro K., Felipe S.H.S., Fernandes A.M., Mayara de Jesus Sousa R., Chagas K., da Silva S.V.S., Nyara de Freitas Correia L., Farias L.M., Leite J.P.V., Rocha D.I. and Otoni W.C. (2020a) Blue and red light affects morphogenesis and 20-hydroxyecdysone [sic] content of *in vitro* *Pfaffia glomerata* accessions. *Journal of Photochemistry & Photobiology, B: Biology* **203**, 111761.

Abstract: The combination of different colors from light-emitting diodes (LEDs) may influence growth and production of secondary metabolites in plants. In the present study, the effect of light quality on morphophysiology and content of 20-hydroxyecdysone (20E), a phytoecdysteroid, was evaluated in accessions of an endangered medicinal species, *Pfaffia glomerata*, grown *in vitro*. Two accessions (Ac22 and Ac43) were cultured *in vitro* under three different ratios of red (R) and blue (B) LEDs: (i) 1R:1B, (ii) 1R:3B, and (iii) 3R:1B. An equal ratio of red and blue light (1R:1B) increased biomass accumulation, anthocyanin content, and 20E production (by 30–40%). Moreover, 1R:1B treatment increased the size of vascular bundles and vessel elements, as well as strengthened xylem lignification and thickening of the cell wall of shoots. The 1R:3B treatment induced the highest photosynthetic and electron transport rates and enhanced the activity of oxidative stress-related enzymes. Total Chl content, Chl/Car ratio, and NPQ varied more by accession type than by light source. Spectral quality affected primary metabolism differently in each accession. Specifically, in Ac22 plants, fructose content was higher under 1R:1B and 1R:3B treatments, whereas starch accumulation was higher under 1R:3B, and sucrose under 3R:1B. In Ac43 plants, sugars were not influenced by light spectral quality, but starch content was higher under 3R:1B conditions. In conclusion, red and blue LEDs enhance biomass and 20E production in *P. glomerata* grown *in vitro*.

Silva T.D., Batista D.S., Castro K.M., Fortini E.A., Felipe S.H.S., Fernandes A.M., Sousa R.M.J., Chagas K., da Silva J.V.S., Correia L.N.F., Torres-Silva G., Farias L.M. and Otoni W.C. (2020b) Irradiance-driven 20-hydroxyecdysone production and morphophysical changes in *Pfaffia glomerata* plants grown *in vitro*. *Protoplasma* (doi.org/10.1007/s00709-020-01558-1).

Abstract: *Pfaffia glomerata* possesses potential pharmacological and medicinal properties, mainly owing to the secondary metabolite 20-hydroxyecdysone (20E). Increasing production of biomass and 20E is important for industrial purposes. This study aimed to evaluate the influence of irradiance on plant morphology and production of 20E in *P. glomerata* grown *in vitro*. Nodal segments of accessions 22 and 43 (Ac22 and Ac43) were inoculated in culture medium containing MS salts and vitamins. Cultures were maintained at 25 ± 2 °C under a 16-h photoperiod and subjected to irradiance treatments of 65, 130, and 200 $\mu\text{mol m}^{-2} \text{s}^{-1}$ by fluorescent lamps. After 30 days, growth parameters, pigment content, stomatal density, *in vitro* photosynthesis, metabolites content, and morphoanatomy were assessed. Notably, Ac22 plants exhibited 10-fold higher 20E production when cultivated at 200 $\mu\text{mol m}^{-2} \text{s}^{-1}$ than at 65 $\mu\text{mol m}^{-2} \text{s}^{-1}$, evidencing the importance of light quantity for the accumulation of this metabolite. 20E production was twice as high in Ac22 as in Ac43 plants although both accessions responded positively to higher irradiance. Growth under 200 $\mu\text{mol m}^{-2} \text{s}^{-1}$ stimulated photosynthesis and consequent biomass accumulation, but lowered carotenoids and anthocyanins. Furthermore, increasing irradiance enhanced the number of palisade and spongy parenchyma cells, enhancing the overall growth of *P. glomerata*.

Silva da Rosa H., Brum de Camargo V., Camargo G., Garcia C.V., Fuentesfria A.M. and Mendez A.S.L. (2015) Ecdysteroids in *Sida tuberculata* R.E. Fries (Malvaceae): chemical composition by LC–ESI-MS and selective anti-*Candida krusei* activity. *Food Chemistry* **182** 193–199.

Abstract: *Sida tuberculata* is found in a region of South America and has traditionally been consumed as an infusion or tea. The chemical composition and antifungal activity of aqueous infusions from leaves and roots were investigated. LC-ESI-MS mass spectra were successfully obtained and used to identify four ecdysteroids: 20-hydroxyecdysone-3-O- β -D-glycopyranoside, 20-hydroxyecdysone, 20-hydroxyecdysone-3-O- β -D-xylose and a hydroxyecdysterone derivative. The *in vitro* antifungal activity was studied, and the minimal inhibitory concentration (MIC) and minimal fungicidal concentration (MFC) were established against *Candida krusei* isolates. The antibiofilm activity was evaluated by the determination of the biofilm removal efficiency in contaminated central venous catheter (CVC) coupons. The preparations exhibited antifungal activity against the species tested, with MICs ranging from 3.90 to 62.50 $\mu\text{g/ml}$. The infusion removed the *C. krusei* biofilm after 90 min of exposure. The observed bioactivity and composition of ecdysteroids will contribute to the future development of antifungal substances for clinical use or as food additives.

Simon A., Pongrácz Z., Tóth G., Mák M., Máthé I. and Báthori M. (2004) A new ecdysteroid with unique 9 β -OH and four other ecdysteroids from *Silene italica* ssp. *nemoralis*. *Steroids* **69**(6), 389–394.

Abstract: A new natural ecdysteroid, 9 β ,20-dihydroxyecdysone (1) and four related compounds 5 α -20-hydroxyecdysone (2), 5 α -2-deoxy-integristerone A (3), integristerone A (4) and 22-deoxy-integristerone A (5) were isolated from the herb of *Silene italica* ssp. *nemoralis*. Compound 1 is the C-9 epimer of the known 9 α ,20-

dihydroxyecdysone (6) and represents a peculiar steroid skeleton. The structures of the compounds were elucidated by 1D and 2D NMR, IR and MS spectroscopy.

Simon A., Tóth G., Liktör-Busa E., Kele Z., Takács M., Gergely A. and Báthori M. (2007) Three new steroids from the roots of *Serratula wolffii*. *Steroids* **72** 751-755 (on-line: DOI: 10.1016/j.steroids.2007.06.004).

Abstract: Investigation of the methanol extract of the roots of *Serratula wolffii* resulted in an ecdysone-related compound, 2 β ,3 β ,20R,22R,25-pentahydroxy-5 β -cholest-6,8(14)-dien (1), a new ecdysteroid, 24-methylene-shidasterone (2), the known compound stachysterone B (3) and its 14,15- α -epoxide (4), a novel natural product. The structures of compounds 1–4 were established by spectral analysis (¹H NMR, ¹³C NMR, COSY, NOESY, HMQC, HMQC-TOCSY and HMBC).

Simon A., Liktör-Busa E., Tóth G., Kele Z., Groska J. and Báthori M. (2008) Additional minor phytoecdysteroids of *Serratula wolffii*. *Helvetica Chimica Acta* **91** 1640-1645.

Abstract: Three new and one known ecdysteroids were identified in the MeOH extract of the roots of *Serratula wolffii*. The new compounds isolated were (11 α)-11-hydroxyshidasterone (1), (2 β ,3 α ,5 β ,14 β ,22R)-2,3,20,22,25-pentahydroxycholest-7-en-6-one (2), and (2 β ,3 α ,5 β ,22R)-2,3,20,22,25-pentahydroxycholest-7-en-6-one (3), together with the known ponasterone A (4). This latter compound was now better characterized than earlier. The structures of compounds 1–4 were established by extensive spectroscopic techniques, including one- and two-dimensional NMR methods.

Simon A., Tóth N., Tóth G., Kele Z., Groska J. and Báthori M. (2009) Ecdysteroids from *Silene viridiflora*. *Helvetica Chimica Acta* **92** 753-761

Abstract: Two new ecdysteroid acetone derivatives, 5 α -2-deoxy-20-hydroxyecdysone 20,22-acetonide (6) and makisterone C 2,3;20,22-diacetonide (8), were isolated from the dried herb of *Silene viridiflora*. The already known 5 β -2-deoxy-20-hydroxyecdysone 20,22-acetonide (7) is additionally reported here as a new constituent of *S. viridiflora*. Five earlier described *S. viridiflora* ecdysteroids, integristerone A (1), 5,20,26-trihydroxyecdysone (26-hydroxypolipodine B; 2), 20,26-dihydroxyecdysone (3), 2-deoxy-20-hydroxyecdysone (4), 2-deoxyintegristerone A (5), are also included because of their improved characterization. The structures were established via spectroscopic analyses, including one- and two-dimensional NMR and mass spectrometry.

Simon A., Ványolós A., Béni Z., Dékány M., Tóth G. and Báthori M. (2011) Ecdysteroids from *Polypodium vulgare* L. *Steroids* **76** 1419-1424.

Abstract: Three new compounds (3, 7, and 11) together with eight known phytoecdysteroids (1, 2, 4-6, and 8-10) were isolated from the rhizomes of common polypody, *Polypodium vulgare* L. The structures of compounds were elucidated by spectroscopic methods including 1D and 2D NMR measurements. The (1)H and (13)C NMR assignments of compounds 1, 6, 9 and 10 are included.

Singh S.B. and Thakur R.S. (1982) Structure and stereochemistry of paristerone, a novel phytoecdysone from the tubers of *Paris polyphylla*. *Tetrahedron* **38**(14), 2189-2194.

Abstract: Paristerone, a novel phytoecdysone has been isolated from the tubers of *P. polyphylla* and its structure and stereochemistry has been established as 2 α , 3 β , 14 α , 20(R), 22(r), 25-hexahydroxy-5 β -cholest-7-en-6-one (2-epiecdysterone) on the basis of chemical and physical evidence.

Singh S.S., Pandey S.C., Srivastava S., Gupta V.S., Patro B. and Ghosh A.C. (2003) Chemistry and medicinal properties of *Tinospora cordifolia* (Guduchi). *Indian Journal of Pharmacology* **35**, 83-91.

Abstract: *Tinospora cordifolia* (Guduchi) is a widely used shrub in folk and ayurvedic systems of medicine. This review presents a detailed survey of the literature on chemistry and medicinal properties of *Tinospora cordifolia*. The chemical constituents reported from this shrub belong to different classes such as alkaloids, diterpenoid lactones, glycosides, steroids, sesquiterpenoid, phenolics, aliphatic compounds and polysaccharides. The notable medicinal properties reported are anti-diabetic, anti-periodic, anti-spasmodic, anti-inflammatory, anti-arthritic, anti-oxidant, anti-allergic, anti-stress, anti-leprotic, anti-malarial, hepatoprotective, immunomodulatory and anti-neoplastic activities.

Sinlaparaya D., Duanghaklang P. and Panichajakul S. (2007a) Enhancement of 20-hydroxyecdysone production in cell suspension cultures of *Vitex glabrata* R.Br. by precursors feeding. *African Journal of Biotechnology* **6**(14) 1639-1642.

Abstract: The effect of ecdysteroid precursors feeding on cell growth and 20-hydroxyecdysone production of *Vitex glabrata* suspension cultures were studied. On the addition of cholesterol, there was no apparent increase of 20-hydroxyecdysone while growth was partially inhibited at higher levels. Feeding of 7-dehydrocholesterol and ergosterol did not affect the cell growth. Both precursors effectively increased production of 20-hydroxyecdysone.

Feeding of 7-dehydrocholesterol as a precursor was most effective. The maximum 20-hydroxyecdysone productivity of about 1.31 mg/L/day was observed in culture with 10 mg/L 7-dehydrocholesterol. This data is the first indication that 7-dehydrocholesterol and ergosterol feeding are effective precursors for 20-hydroxyecdysone formation in plant cell suspension culture.

Sinlaparaya D., Duanghaklang P., Panichajakul S. and Zhang Y. (2007b) Optimization of cell growth and 20-hydroxyecdysone production in cell suspension cultures of *Vitex glabrata* R.Br. Chinese Journal of Biotechnology (6), 1033-1036 [in Chinese].

Abstract: The effects of the cultivation media, plant growth regulators and inoculum size on the cell growth and 20-hydroxyecdysone production in suspension cultures of *Vitex glabrata* R.Br. were investigated. The cell growth and 20-hydroxyecdysone formation reach the highest when cells are cultured in the Gamborg's B5 medium supplemented with 2.0mg/L BAP (6-benzylaminopurine) and 1.0mg/L 2,4-D. The maximum 20-hydroxyecdysone productivity, of about 1.1mg/L/day, was observed in the culture with 20% PCV (packed cell volume) of inoculum size. These data also show that the increment of the inoculum size to 20% PCV could increase the productivity in 7-folds.

Sipahimalani A.T., Banerji A. and Chadha M.S. (1972) Biosynthesis and interconversion of phytoecdysones in *Sesuvium portulacastrum* L. Journal of the Chemical Society, Chemical Communications 692-693.

Abstract: Incorporation of mevalonic acid and cholesterol into ecdysone and ecdysterone and conversion of ecdysone into ecdysterone in the plant, *Sesuvium portulacastrum* L., have been demonstrated.

Snogan E., Lechat I., Ho R., Bertho G., Girault J.-P., Ortiga S., Maria A. and Lafont R. (2007) Ecdysteroids from the medicinal fern *Microsorium scolopendria* (Burm. f.). Phytochemical Analysis 18, 441-450.

Abstract: Fronds of the fern *Microsorium scolopendria* are widely used in traditional medicine in the Society Islands. They were investigated for the presence of ecdysteroids, which might be responsible for at least some of their medicinal properties. *M. scolopendria* represents an excellent source of ecdysone (0.16% of dry weight) and 20-hydroxyecdysone (0.20%), and also contains significant amounts (0.01-0.02%) of makisterones A and C, inokosterone and amarasterone A, together with lower amounts of poststerone and of a compound tentatively identified as 24,28-diepi-cyasterone. During this study, three new minor phytoecdysteroids, namely 20-deoxymakisterone A, a 25(?)-epimer of amarasterone A and 25-deoxyecdysone 22-glucoside were also isolated by a combination of normal- and reversed-phase HPLC and subsequently identified by NMR.

Song C.Q. and Xu R.S. (1991) Phytoecdysones from the roots of *Tinospora capillipes*. Chinese Chemical Letters 2(1), 13-14.

Abstract: In addition to 2-deoxycrustecdysone(1), we isolated and identified two new natural moulting hormones-2-deoxy-3-epicrustecdysone(2) and 2-deoxycrustecdysone-3-O- β -D-glucopyranoside(3) from the roots of *Tinospora capillipes*, it is the first time to get ecdyst

Sovová H., Sajfřtová M. and Pavlík M. (2007) Extraction of phytoecdysones with ethanol-modified supercritical CO₂. Proceedings of European Congress of Chemical Engineering (ECCE-6), Copenhagen, 16-20 September 2007, 1-6.

Abstract: Biologically active polar substance 20-hydroxyecdysone was extracted from roots and leaves of *Leuzea carthamoides* with ethanol-modified dense carbon dioxide. The effect of operating conditions (pressure 20-27 MPa, temperature 40-60 C, ethanol concentration in the solvent 4.3-7.1 wt.%, and solvent-to-feed ratio up to 100 g/g) on the extraction rate and the concentration in extract was examined. The maximum extraction yields were achieved with 7.1 wt. % ethanol in the solvent. Despite the high solvent-to-feed ratios the extraction was not complete; the maximum yield of 20-hydroxyecdysone from the leaves containing 1.50 mg/g dry mass was only 0.19 mg/g and the maximum yield from the roots containing 0.49 mg/g dry mass was 0.30 mg/g. The maximum concentration of 20-hydroxyecdysone in CO₂ extract from the roots, 2.9 wt.%, was higher than in the extracts obtained with 95% EtOH (1.74 wt.%).

Sovová H., Opletal L., Sajfřtová M. and Bártlová M. (2008) Supercritical fluid extraction of cynaropicrin and 20-hydroxyecdysone from *Leuzea carthamoides* DC. Journal of Separation Science 31, DOI 10.1002/jssc.200700496.

Abstract: *Leuzea carthamoides* is an adaptogenic plant containing biologically active compounds as ecdysteroids and guaianolide-type sesquiterpene lactones, conventionally extracted from the plant with ethanol. It may be a potential source of the mentioned natural compounds. Ethanol-modified near-critical CO₂ was used as selective solvent with the aim to increase the level of 20-hydroxyecdysone in the extract from *L. carthamoides* roots and to remove selectively cynaropicrin, a sesquiterpene lactone of bitter taste, from the leaves. The extraction conditions were varied (pressure 20-28 MPa, temperature 40-60 degrees C, ethanol concentration in the solvent 0-7.1%) and the extraction yield and extract composition were compared with the results of ethanolic extraction. The supercritical fluid extraction (SFE) from finely powdered plant was controlled by phase equilibrium. Cynaropicrin was

quantitatively removed from the leaves where 89% of 20-hydroxyecdysone was retained. The extraction yield of 20-hydroxyecdysone from roots with ethanol-modified CO₂ was lower by 30% than with ethanol but its concentration in the extract was higher by 67%.

Sreejit C.M. (2014) Quantitative ethnobotany and phytochemistry of selected plants used in traditional therapeutics by ethnic tribes of Wayanad District, Kerala. Ph.D. Thesis, Mahatma Gandhi University, Kottayan, Kerala, India.

Sreejit C.M., Bose C., Banerji A. and Thomas P.M. (2018) Bioprospection of Kerala flora for the multipurpose drug – phytoecdysoids. *J. Biol. Chem. Researach* 35(1), 115-123.

Abstract: Phytoecdysoids are analogues of the arthropod steroid hormone ecdysterone, found in plants which are apparently non-toxic to mammals with a wide range of pharmacological - adaptogenic, anabolic, anti-diabetic, hepatoprotective, immunoprotective, wound healing, and perhaps even anti-tumor- activities. Though they have been reported from more than 100 terrestrial plant families, till date, only less than 2 % of the world's flora has been investigated for their presence. Considering its pharmacological activities and extremely non toxic nature, it can very well be the darling of pharmaceutical companies in future. Kerala, being a part of the mighty Western Ghats range has a huge potential in exploiting its rich, unique and highly endemic biodiversity. A bio prospection study for ecdysteroids has been done with regard to the Angiosperm flora of the state for the first time. Fifty plant species were screened from the study area. A simple protocol for screening ecdysteroids using very less amount of the plant source was developed using ultra sonication and Thin Layer Chromatography. Ten species were found positive for phytoecdysoids. Ecdysterone was reported for the first time ever from *Cosciniun fenestratum*

Sreejit C.M., Bose C., Banerji A. and Thomas M.P. (2019) Isolation, quantification and chemical characterisation of ecdysterone from medicinal plants of Kerala, Western Ghats. *Journal of Pharmacognosy and Phytochemistry* 8(2), 254-257.

Abstract: Ecdysteroids are a group of compounds responsible for molting in insects and is variously expressed in plant kingdom, believed to be a means of deterring insects by influencing the metabolism and metamorphosis in these vectors. Kerala flora has not been screened for the presence of ecdysteroids before. This work is a follow up study based on a preliminary Bio prospection study on fifty medicinally important plants used by indigenous tribes of Kerala for the presence of ecdysterone. Four potential plant species which were found to have adequate amount of compound-*Diploclisia*, *Cyathula*, *Sesuvium* and *Cosciniun*-were put to detailed extraction, isolation, quantification and chemical chacterisation using HPLC, UV and IR spectroscopy. Literature survey suggested that soil and geographical regime has direct influence on the expression levels of ecdysterone. Some variations were observed in the expression levels of ecdysterone in our study too, in comparison with published literature but potential sources from indigenous plants were identified during this study. Availability in adequate quantity of this wonder molecule will increase its multi-faceted activity related studies in future.

Sreejit C.M. and Nelshi P.L. (2019) Bioprospection of some medicinal plants used in the traditional system Ayurveda, for the wonder drug – ecdysterone. *International Journal for Research in Applied Science & Engineering Technology* 7(IV), 1163-1165.

Abstract: This work was a bioprospection study to assess the presence of the multipurpose drug Ecdysterone in some plants used in the traditional medicinal system Ayurveda. Fourteen plants namely, *Vernonia cinerea*, *Azadirachta indica*, *Plectranthus amboinicus*, *Cardiospermum helicacabum*, *Ayyappana triplinervis*, *Phyllanthus amarus*, *Terminalia chebula*, *Andrographis paniculata*, *Cosciniun fenestratum*, *Samadera indica*, *Justicia adhatoda*, *Centella asiatica*, *Mimusops elanji*, and *Rauvolfia serpentina* were selected for the study and .screened for the presence of ecdysterone in their active parts. Presence of ecdysterone was confirmed in the seeds of *Cosciniun fenestratum* for the first time ever.

Sripinyowanich S., Kil E-J., Petchsri S., Jo Y., Choi H., Cho W.K. and Lee S. (2021) De novo transcriptome assembly of two *Microsorium* fern species identifies enzymes required for two upstream pathways of phytoecdysteroids. *International Journal of Molecular Sciences* 22: article 2085 (doi: 10.3390/ijms22042085).

Abstract: *Microsorium* species produce a high amount of phytoecdysteroids (PEs), which are widely used in traditional medicine in the Pacific islands. The PEs in two different *Microsorium* species, *M. punctatum* (MP) and *M. scolopendria* (MS), were examined using high-performance liquid chromatography (HPLC). In particular, MS produces a high amount of 20-hydroxyecdysone, which is the main active compound in PEs. To identify genes for PE biosynthesis, we generated reference transcriptomes from sterile frond tissues using the NovaSeq 6000 system. De novo transcriptome assembly after deleting contaminants resulted in 57,252 and 54,618 clean transcripts for MP and MS, respectively. The clean *Microsorium* transcripts for each species were annotated according to gene ontology terms, UniProt pathways, and the clusters of the orthologous group protein database using the MEGAN6 and Sma3s programs. In total, 1852 and 1980 transcription factors were identified for MP and MS, respectively. We obtained transcripts encoding for 38 and 32 enzymes for MP and MS, respectively, potentially involved in mevalonate and

sterol biosynthetic pathways, which produce precursors for PE biosynthesis. Phylogenetic analyses revealed many redundant and unique enzymes between the two species. Overall, this study provides two *Microsorium* reference transcriptomes that might be useful for further studies regarding PE biosynthesis in *Microsorium* species

Staal G.B. (1967) Plants as a source of insect hormones. Proc. Nedl. Akad. Wetenschappen C 70, 409-418.

Stark T.D., Rami J., Froehlich S., Weiss P., Vierling W., Dongmo A.B. and Hofmann T. (2020) A new phytoecdysteroid from the stem bark of *Vitex cienkowskii*. European Food Research and Technology (doi.org/10.1007/s00217-020-03591-2).

Abstract: To investigate differences in the metabolome of the stem bark of *Vitex cienkowskii* harvested at three different locations and three different seasons, their extracts were analyzed by means of UPLC–ESI–IMS–TOF MS[®]. One marker metabolite was isolated and chemically characterized, which was identified as the new compound 20,24-dihydroxy,24-hydroxymethylecdysone (**1**). IMS showed two drift time species for **1** which could be used as new and additional characteristic compound parameters in compound identification to reduce dereplication and false positives.

Stevens J.F., Reed R.L. and Morr  J.T. (2008) Characterization of phytoecdysteroid glycosides in meadowfoam (*Limnanthes alba*) seed meal by positive and negative ion LC-MS/MS. Journal of Agricultural and food Chemistry (published on-line: 10/5/2008).

Abstract: Meadowfoam (*Limnanthes alba*) is an oilseed crop grown in western Oregon. The seed meal has potential value as a biopesticide due to glucosinolate degradation products and phytoecdysteroids, a group of polyhydroxylated triterpenoids with potent activities as arthropod molting hormones. Liquid chromatography in combination with tandem mass spectrometry operated in the precursor ion mode revealed the presence of four ecdysteroid glycosides in meadowfoam seed meal. The carbohydrate sequence and the identity of the ecdysteroid aglycones, ponasterone A and 20-hydroxyecdysone, were determined by product ion scanning. Ecdysteroids were detected in the negative ion mode as [M + formate][−] ions, which yielded [M − H][−] and α-cleavage fragments with retention of hydroxyl groups in MS/MS experiments (not seen in the positive ion mode), allowing the determination of the number of hydroxyl groups in the side chain and in the steroid ring system. MS/MS of glycoside ions ([MH]⁺ or [M + formate][−]) provided carbohydrate sequence information.

Su C-R., Ueng Y~F., Dung N.X, Reddy V.B. and Wu T-S. (2007) Cytochrome P3A4 inhibitors and other constituents of *Fibraurea tinctoria*. Journal of Natural Products (published on-line; 10/11/2007).

Abstract: Four new furanoditerpenoids, fibrauretin A (**1**), fibrauretinolide A (**2**), *epi*-fibrauretinolide A (**3**), and *epi*-12-palmatoside G (**4**), and a new ecdysteroid glucoside, fibraurecdyside A (**5**), together with seven known compounds including two furanoditerpenoids (**6** and **7**), an ecdysteroid (**8**), and four quaternary protoberberine alkaloids (**9–12**) were isolated from the stems of *Fibraurea tinctoria*. The structures of **1–5** were established on the basis of spectroscopic evidence. Among these compounds, palmatine (**9**) and jatrorrhizine (**10**) showed inhibitory effects against cytochrome P450 3A4 (CYP3A4) with IC₅₀ values of 0.9 and 2.1 μM, respectively.

Subeki, Matsuura H., Takahashi K., Yamasaki M., Yamamoto O., Maede Y., Katakura K., Suzuki M., Trimurningishi, Chairul and Yoshihara T. (2005) Antibabesial activity of protoberberine alkaloids and 20-hydroxyecdysone from *Arcangelisia flava* against *Babesia gibsoni* in culture. Journal of Veterinary Medical Science 67(2), 223-227.

Abstract: Bioassay-guided fractionation of the boiled extract from the stems of *Arcangelisia flava* led to the isolation of palmatine (**1**), berberine (**2**), jatrorrhizine (**3**), dihydroberberine (**4**) and 20-hydroxyecdysone (**5**). The chemical structures of these compounds were elucidated on the basis of their chemical and spectral evidence. The isolated compounds were evaluated for their growth inhibiting effects on *Babesia gibsoni* in culture for a week. Compounds (**1–4**) showed significant inhibitions at concentrations from 100 to 1.0 microg/ml, while compound **5** at a concentration of 100 microg/ml, only.

Suksamrarn A. and Sommechai C. (1993) Ecdysteroids from *Vitex pinnata*. Phytochemistry 32(2), 303-306.

Abstract: A new ecdysteroid, pinnatasterone, together with 20-hydroxyecdysone and turkesterone were isolated from the bark of *Vitex pinnata*.

Suksamrarn A., Wilkie J.S. and Horn D.H.S. (1986) Blechnosides A and B: ecdysteroid glycosides from *Blechnum minus*. Phytochemistry 25(6), 1301-1304.

Abstract: The structures of two new ecdysteroid glycosides from *Blechnum minus* have been shown, on the basis of chemical, mass spectral, ¹H NMR and ¹³C NMR spectral evidence, to be 2-deoxyecdysone 3-β- d -glycopyranoside (blechnoside A) and 2-deoxyecdysone 25-β- d -glucopyranoside (blechnoside B).

Suksamrarn A., Sommechai C., Charlupong P and Chitkul B. (1995) Ecdysteroids from *Vitex canescens*. *Phytochemistry* **38**(2), 473-476.

Abstract: In addition to 20-hydroxyecdysone and turkesterone, a new ecdysteroid, canescensterone, was isolated from the bark of *Vitex canescens*. The new compound was shown to be pinnatasterone 24-O-(pyrrole 2-carboxylate).

Suksamrarn A., Promrangsan N., Chitkul B., Homvisasevongsa S. and Sirikate A. (1997) Ecdysteroids of the root bark of *Vitex canescens*. *Phytochemistry* **45**(6), 1149-1152.

Abstract: A new ecdysteroid, 24-epi-abutasterone, was isolated from the root bark of *Vitex canescens*. 20-Hydroxyecdysone, 24-epi-makisterone A, shidasterone, calonysterone and turkesterone were also isolated from this plant species.

Suksamrarn A., Yingyongnarongkul B.-e. and Promrangsan N. (1998) Naturally occurring 20,26-dihydroxyecdysone exists as two C-25 epimers which exhibit different degrees of moulting hormone activity. *Tetrahedron* **54**, 14565-14572.

Abstract: 20,26-Dihydroxyecdysone (20,26-ECD) isolated from *Vitex canescens*, *V. glabrata* and *V. pinnata* has been shown to exist as two C-25 epimers. Synthesis and separation of the two epimers were accomplished and they exhibited different degree of moulting hormone activity in the *Musca* assay.

Suksamrarn A., Yingyongnarongkul B. and Charoensuk S. (1999) Regioselective synthesis of 24-epi-pterosterone. *Tetrahedron* **55**(1), 255-260.

Abstract: In order to have compounds available for structure-activity relationship studies, the ecdysteroid 24-epi-pterosterone was synthesized from 20-hydroxyecdysone and pterosterone was obtained from *Vitex glabrata* stem bark. The former was approximately 7-fold less active than the latter in the *Musca* bioassay for moulting hormone activity.

Suksamrarn A., Promrangsan N. and Jintasirikul A. (2000) Highly oxygenated ecdysteroids from *Vitex canescens* root bark. *Phytochemistry* **53**(8), 921-924.

Abstract: Highly oxygenated ecdysteroids, (24R)-11 alpha, 20,24-trihydroxyecdysone and 11 alpha,20,26-trihydroxyecdysone, have been isolated from the polar fraction of *Vitex canescens* root bark. The latter exists as two C-25 epimers which could be separated by reversed-phase HPLC.

Suksamrarn A., Kumpun S. and Yingyongnarongkul B.-e. (2002) Ecdysteroids of *Vitex scabra* stem bark. *Journal of Natural Products* **65**(11), 1690-1692.

Abstract: Two new ecdysteroids, 24-epi-pinnatasterone (1) and scabrasterone (2), together with 11 known ecdysteroids, calonysterone, pterosterone, 24-epi-makisterone A, 20-hydroxyecdysone (3), polypodine B, ajugasterone C, pinnatasterone (4), 11alpha-hydroxyecdysone, 24-epi-abutasterone, 20,26-dihydroxyecdysone, and turkesterone (5), were isolated from the stem bark of *Vitex scabra*. This plant species contained a very high concentration (1.8%) of 3 and thus provided a good source of this parent ecdysteroid and related rare ecdysteroids. Compounds 1 and 2 exhibited very low moulting activity in the *Musca* bioassay. The low biological activity of these two ecdysteroids was in agreement with those of other 22-deoxyecdysteroids.

Sumayya S. (2005) Studies in the chemical constituents of *Murraya paniculata* and *Ipomoea hederacea*. Ph.D. Thesis, University of Karachi, Pakistan.

Sun F.-f., Li C.-x. and Fan G.-z. (2015) Effects of exogenous CO on the accumulation of β -ecdysterone in *Ajuga* sp. *Pratacultural Science* (9), 1438-1443 [in Chinese, with an English abstract].

Abstract: In order to clarify the regulatory effects of carbon monoxide (CO) on plant secondary metabolites, this study analysed the content of β -morphic steroids in the root and leaf-induced healing tissues of *Ajuga multiflora* and *A. lobata* and suspended the culture of healing tissues with high levels of β -moulting oxytosterone. The addition of different concentrations of CO-supply chloro-high-speed haemolybin to the rib grass suspension culture system found that CO promoted the synthesis of β moulting steroids in suspended cells. At the same time, it was found that when treating 8d, low concentrations of CO treatment (8t betamol. L⁻¹) increased β accumulation of moulting ketones by 80.82%, but high concentration treatment (12, 16, 20 mol. The β -moulting ketone content in suspended cells of L⁻¹ was significantly lower than in the control group (P<0.05). The above results initially confirmed the low concentration of CO (8 μ mol. L⁻¹) Treatment has a promoting effect on the accumulation of β -moulting steroids in the suspended cells.

Sun L-L., Wu J., Zhang Y., Kang H. and Song J., (2017) Optimization of ultrasonic extraction technique in *Achyranthes bidentata* (Bl.) with response surface analysis. *Research and Practice on Chinese Medicines* (6), 48-51 [in Chinese, with an English abstract].

Abstract: Objective To optimize of ultrasonic extracting technique for β -hydroxyecdysone in *Achyranthes bidentata* (Bl.) . Methods Based on the single factor and the response value of β -hydroxyecdysone of *Achyranthes bidentata* (Bl.) , a 3-factor and 3-level response surface method is used to optimize the extracting value of β -hydroxyecdysone in *Achyranthes bidentata* (Bl.) . Results The optimum extraction process of β -hydroxyecdysone in *Achyranthes bidentata* (Bl.) is normal butanol concentration (45%) , the ratio of solid to liquid (15 : 1) and the extraction time (1.7 h) . The predictive and actual values of the extraction quantity of *Achyranthes bidentata* (Bl.) are 5.17 mg/g and 5.29 mg/g under the condition of optimum method, respectively. The most important influencing factor for the extraction quantity of β -hydroxyecdysone in *Achyranthes bidentata* (Bl.) is normal butanol concentration, the ratio of solid to liquid, and the extraction time last. Conclusion The preferred process of optimizing the extraction process of β -hydroxyecdysone in *Achyranthes bidentata* (Bl.) with response surface analysis is stable and feasible.

Sun Y., Qing D-g., Zhang J., Ni H. And Jia X-g. (2012) Determination of β -ecdysone in root of *Stemmacantha carthamoides* (Willd.) Dittrich. *Chinese Journal of Pharmaceutical Analysis* 32 (9), 1614-1616 [in Chinese, with English Abstract and Legends].

Abstract: Objective: To establish a method for determination of β -ecdysone in root of *Stemmacantha carthamoides*(Willd.)Dittrich. Methods: β -Ecdysone was determined by RP-HPLC on Waters XTerra RP C18(250 mm \times 4.6 mm,5 μ m)column with methanol-1% acetic acid(27: 73)as mobile phase at a flow rate of 1.0 mL \cdot min⁻¹,the detection wavelength was 250 nm and the column temperature was 30 $^{\circ}$ C.Results:The calibration curve of β -ecdysone was linear in the range of 1.45-14.5 μ g \cdot mL⁻¹($r=0.9998$),and the average recovery was 100.4%.Conclusions:This method is simple,accurate,sensitive and reproducible.It can be used for determination of β -ecdysone in root of *Stemmacantha carthamoides*(Willd.)Dittrich.

Suri, O. P., R. Kant, R. S. Jamwal, K. A. Suri and C. K. Atal (1982) *Boerhaavia diffusa*, a new source of phytoecdysones. *Planta Medica* 44 (3), 180-181.

No Abstract.

Svatoš A and Macek T (1994) The rate of ecdysteroid production in suspension cultured cells of the fern *Pteridium aquilinum*. *Phytochemistry* 35(3), 651-654.

Abstract: The rate of production of three ecdysteroids, ponasterone, ecdysone and ecdysterone, by a suspension culture of cells of bracken fern, *Pteridium aquilinum*, was investigated. The highest rate of production was between the fourth and 13th day of cultivation, and the concentration of ecdysteroids in the cells (0.02% of fresh wt) was higher in comparison to the original plant. Ponasterone was the major compound identified. A potential value of this culture for a study of the biosynthesis of ecdysteroids is discussed.

Sviridova T.P., Revina T.A. and Yakoleva I.A. (1993) Biological and chemical peculiarities of species of the genus *Rhaponticum* Ludw. grown in the south of the Tomsk district. *Rastitelny Resursy* 29(3), 50-57 [in Russian].

Sviridova T.P., Zibareva L.N. and Kritskaja S.V. (1995) Biological and chemical specialities of species of the genus *Silene* L. grown in the south of the Tomsk Region. Abstract of the International Conference "Specialities of acclimatization of perennial plants accumulating biologically active substances", Krasnodar, Russia, pp. 209-212 [in Russian].

Szendrei K., Varga E., Hajdu Z., Herke I., Lafont R. and Girault J-P. (1988) Ajugasterone C and 5-deoxykaladasterone, and ecdysteroid artifact, from *Leuzea carthamoides*. *Journal of Natural Products* 51(5), 993-995.

No Abstract.

Tabekoueng G.B., Akak C.M., Langat M.K., Azebaze G.B., Waffo A.F.K., Mulholland D.A. and Vardamides J.C. (2019) The chemical constituents of *Penianthus longifolius* Miers. *Phytochemistry Letters* 30, 103-106.

Abstract: Fourteen compounds were isolated from the roots, leaves and twigs of *Penianthus longifolius* Miers (Menispermaceae), including two previously unreported neo-clerodane diterpenoids, penianthin (1), its C-8 epimer (2). In addition, the previously reported O-methylmoschatoline (3), taraxerol (4), taraxerone (5), rubrosterone, (6), panuosterone (7), 22-epi-20-hydroxyecdysone (8), ergosterol peroxide (9), stigmast-5-ene-3,7-dione (10),

stigmasterol (11), β -sitosterol (12), stigmasterol glucoside (13) and β -sitosterol glucoside (14) were isolated. The structures of the compounds were determined by means of NMR spectroscopic and mass spectrometric analysis. The absolute configurations of 1 and 2 were determined by circular dichroism analysis. Compounds 1 and 2 were screened against the NCI60 cancer cell panel but showed no significant activity at 10 μ M.

Tabekoueng G.B., Akak C.M., Happi G.M., Langat M.K., Frese M., Stammer H-G., Neumann B., Azebaze A.G.B., Waffo A.F.K., Wansi J.D., Lenta B.N., Sewald N., Vardamides J.C. and Nkengfack A.E. (2020) The chemistry of the West and Entral African *Penianthus zenkeri* Diels (Menispermaceae). *Phytochemistry Letters* **38**, 12-16.

Abstract: A phytochemical investigation of the roots and twigs of *Penianthus zenkeri* Diels led to the identification of twenty four compounds including one previously undescribed C-28 ecdysteroid (1) and previously described phytosteroids (2–5, 20, 22, 23), phenolic compounds (6–8, 12), diterpenoids (9–11), alkaloids (13,14, 18, 19), a dipeptide (15), ursane type triterpenoids (16–17), a carbohydrate (21) and a fatty alcohol (24). Three semi-synthetic derivatives (5b, 5a, 17a) obtained from acetylation and acetonidation of rubrosterone (5), and allylation of ursolic acid (17), respectively, are also reported. The chemical structures of the compounds were determined using 1D and 2D NMR spectral data, mass spectrometry and by comparison with the data reported in the literature. The absolute configuration of compound 9 was established using X-Ray and ECD analysis. Different extracts of the twigs and roots, compounds 2–4, 6–11, 15, and 17–21 were evaluated for their antibacterial, and cytotoxic activities. The twigs extract showed an antibacterial activity with an MIC of 62.5 μ g/mol against *Staphylococcus aureus* ATCC 4300, while ursolic acid exhibited a moderate cytotoxicity with an IC₅₀ of 50.9 μ M. The chemotaxonomic relevance of the isolated compounds is discussed.

Taha-Salaime L., Davidovich-Rikanati R., Sadeh A., Abu-Nassar J., Marzouk-Kheredin S., Yahyaa Y., Ibdah M., Ghanim M., Lewinsohn E., Inbar M. and Aly R. (2019) Phytoecdysteroids and clerodane content in three wild *Ajuga* species in Israel. *ACS Omega* **4**, 2369-2376.

Abstract: Many species of the genus *Ajuga* (family Lamiaceae) contain phytoecdysteroids and clerodane diterpenes. Phytoecdysteroids are triterpene-derived analogues of steroid hormones that control molting and metamorphosis in arthropods, whereas clerodanes deter phytophagous insects. We identified and quantified phytoecdysteroid and clerodane contents in three *Ajuga* plant species in Israel. Leaves and roots of *Ajuga iva*, *Ajuga chamaepitys* (*Ajuga chia*), and *Ajuga orientalis* were collected from three different populations. Using liquid chromatography–time of flight–mass spectrometry analysis, we identified three phytoecdysteroids: 20-hydroxyecdysone (ecdysterone), makisterone A, and cyasterone and two clerodanes: dihydroajugapitin and columbin. Their contents varied significantly among plant species, organs, and populations. The highest concentrations of 20-hydroxyecdysone, makisterone A, and cyasterone were recorded in leaves and roots of *A. iva*. Cyasterone content tended to be higher in leaves of *A. chamaepitys*. Clerodane concentrations were generally negligible or nonexistent. Dihydroajugapitin concentrations were highest in *A. iva* leaves but were lower or undetectable in the roots and in the other two species. Columbin concentration was similar in all species and organs. Phytoecdysteroid contents also varied among populations within species. Because phytoecdysteroids have disruptive effects on phytophagous insect growth, the potential role of extracts of *A. iva* in pest-management programs is of interest.

Takács M., Simon A., Liktör-Busa E., Báthori M., Zsila F., Bikádi Z., Horváth P., Veress G, Gergely A. and Tóth G. (2010) Structure and stereochemistry of novel ecdysteroids from the roots of *Serratula wolffii*. *Magnetic Resonance in Chemistry* **48** 386–391.

Abstract: Three new natural ecdysteroids viz. 22-dehydro-20-deoxy-ajugasterone C (1), 1-hydroxy-22-deoxy-20,21-didehydro-ecdysone (2) and 22-deoxy-20,21-didehydro-ecdysone (3) were isolated from the methanol extract of the roots of *Serratula wolffii*. The structures of compounds 1–3 were established by various spectroscopic techniques, including one- and two-dimensional NMR, circular dichroism and mass spectroscopic methods.

Takasaki M., Tokuda H, Nishino H. and Konoshima T. (1999) Cancer chemopreventative agents (antitumor-promoters) from *Ajuga decumbens*. *Journal of Natural Products* **62**, 972-975.

Abstract: Sixteen compounds (1-16) isolated from the flowering whole plant of *Ajuga decumbens* have been tested for their inhibitory effects on Epstein-Barr virus early antigen (EBV-EA) induction by the tumor promoter, 12-O-tetradecanoylphorbol-13-acetate (TPA), as a primary screening test for antitumor-promoters (potential cancer chemopreventive agents). Five compounds (6, 9, and 12-14) showed strong inhibitory effects on EBV-EA induction. Of these active compounds, two major constituents of this plant, cyasterone (6) and 8-acetylharpagide (13), showed potent antitumor-promoting activities on a mouse-skin in vivo two-stage carcinogenesis procedure, using 7, 12-dimethylbenz[a]anthracene as initiator and TPA as promoter. Further, compound 13 also exhibited potent chemopreventive activity in a mouse pulmonary tumor model.

Takemoto T., Hikino Y., Nomoto K. and Hikino H. (1967b) Structure of cyasterone, a novel C₂₉ insect-moulting substance from *Cyathula capitata*. *Tetrahedron Letters* (33), 3191-3194.

No Abstract.

Takemoto T., Ogawa S., Nishimoto N., Arihara S. and Bue K. (1967c) Insect moulting activity of crude drugs and plants (1). *Yakugaku Zasshi* 87, 1414-1418 [in Japanese, with an English abstract].

Takemoto T., Ogawa S., Nishimoto N. and Hoffmeister H. (1967d) Steroide mit Häutungshormon-Aktivität aus Tieren und Pflanzen [Steroids with moulting hormone activity from animals and plants]. *Zeitschrift für Naturforschung* 22b, 681-682 [in German].

No Abstract.

Takemoto T., Ogawa S., Nishimoto N., Hirayama H. and Taniguchi S. (1967e) Isolation of insect-moulting hormones from mulberry leaves. *Yakugaku Zasshi* 87, 748 [in Japanese].

No Abstract.

Takemoto T., Ogawa S. and Nishimoto N. (1967f) Studies on the constituents of *Achyranthis radix*. II. Isolation of insect moulting hormones. *Yakugaku Zasshi* 87, 1469-1473 [in Japanese, with an English abstract].

Takemoto T., Ogawa S. and Nishimoto N. (1967g) Studies on the constituents of *Achyranthis radix*. III. Structure of inokosterone. *Yakugaku Zasshi* 87, 1474-1477 [in Japanese, with an English abstract].

Takemoto T., Ogawa S. and Nishimoto N. (1967h) Isolation of moulting hormones of insects from *Achyranthis radix*. *Yakugaku Zasshi* 87, 325-327 [in Japanese].

Takemoto T., Ogawa S., Nishimoto N., Yen K-Y., Abe K., Sato T., Osawa K., and Takahashi M. (1967i) The isolation of ecdysterone from the radix of *Achyranthes obtusifolia* Lam. *Yakugaku Zasshi* 87, 1521-1523 [in Japanese, with an English abstract].

Takemoto T., Ogawa S., Nishimoto N. and Taniguchi S. (1967j) Studies on the constituents of *Achyranthis radix*. IV. Isolation of the insect moulting hormones from Formosan *Achyranthes* spp. *Yakugaku Zasshi* 87, 1478-1480 [in Japanese, with an English abstract].

Abstract: Examinations were made on the insect metamorphosis activity and ecdysterone content of Formosan *Achyranthes obtusifolia* LAM. (Japanese name "Shima-inokozuchi"), *A. rubrofusca* WIGHT (Japanese name "Murasaki-inokozuchi"), and *A. longifolia* MAKIMO (Japanese name "Yanagi-inokozuchi"). Ecdysterone was isolated from the root of *A. rubrofusca* and from the total herb of *A. longifolia*, and identified. A crystalline component of m.p. 225~230° (decomp.) was isolated from the root of *A. rubrofusca*.

Takemoto T., Ogawa S., Nishimoto N. and Mue K. (1967k) Studies on the constituents of *Achyranthis radix*. V. Insect hormone activity of ecdysterone and inokosterone on the flies. *Yakugaku Zasshi* 87, 1481-1485 [in Japanese, with an English abstract].

Takemoto T., Hikino Y., Arai T. and Hikino H. (1968a) Structure of lemmasterone, a novel C₂₉ insect-moulting substance from *Lemmaphyllum microphyllum*. *Tetrahedron Letters* (37), 4061-4064.

No Abstract.

Takemoto T., Arihara S., Hikino Y. and Hikino H. (1968b) Structure of pterosterone, a novel insect-moulting substance from *Lastrea threlypteris* and *Onoclea sensibilis*. *Tetrahedron Letters* (3), 375-378.

Takemoto T., Hikino Y., Jin H., Arai T. and Hikino H. (1968c) Isolation of insect moulting substances from *Osmunda japonica* and *Osmunda asiatica*. *Chemical and Pharmaceutical Bulletin* 16, 1636.

No Abstract.

Takemoto T., Nomoto K and Hikino H. (1968d) Structure of amarasterone A and B, novel C₂₉ insect-moulting substances from *Cyathula capitata*. *Tetrahedron Letters* (48), 4953-4956.

Takemoto T., Arihara S., Hikino Y. and Hikino H. (1968e) Isolation of insect moulting substances from *Pteridium aquilinum* var. *latiusculum*. *Chemical and Pharmaceutical Bulletin* 16, 762.

No Abstract.

Takemoto T., Hikino Y., Jin H. and Hikino H. (1968f) Isolation of ponasterone A from *Taxus cuspidata* var. *nana*. *Yakugaku Zasshi* 88, 359 [in Japanese].

Takemoto T., Hikino Y., Arai T., Konno C., Nabetani S. and Hikino H. (1968g) Isolation of insect moulting substances from *Pleopeltis thunbergiana*, *Neocheiropteris ensata*, and *Lemmaphyllum microphyllum*. *Chemical and Pharmaceutical Bulletin* 16, 759-760.

No Abstract.

Takemoto T., Hikino Y., Okuyama T., Arihara S. and Hikino H. (1968h) Structure of shidasterone, a novel insect-moulting substance from *Blechnum niponicum*. *Tetrahedron Letters* (58), 6095-6098.

No Abstract.

Takemoto T., Nomoto K., Hikino Y. and Hikino H. (1968i) Structure of capitasterone, a novel C₂₉ insect-moulting substance from *Cyathula capitata*. *Tetrahedron Letters* (47), 4929-4932.

Takemoto T., Arihara S. and Hikino H. (1968j) Structure of ponasteroside A, a novel glycoside of insect-moulting substance from *Pteridium aquilinum* var *latiusculum*. *Tetrahedron Letters* (39), 4199-4202.

No Abstract.

Takemoto T., Okuyama T., Arihara S., Hikino Y. and Hikino H. (1969) Isolation of insect-moulting substances from *Blechnum amabile* and *Blechnum niponicum*. *Chemical and Pharmaceutical Bulletin* 17(9), 1973-1974.

No Abstract.

Takemoto T., Hikino Y., Hikino H., Ogawa S. and Nishimoto N. (1969b) Rubrosterone, a metabolite of insect metamorphosing substance from *Achyranthes rubrofusca*: structure and absolute configuration. *Tetrahedron* 25, 1241-1248.

No Abstract.

Takemoto T., Okuyama T., Jin H., Arai T., Kawahara M., Konno C., Nabetani S., Arihara S., Hikino Y. and Hikino H. (1973) Isolation of phytoecdysones from Japanese ferns. I. *Chemical and Pharmaceutical Bulletin* 21, 2336-2338.

No Abstract.

Takenada T., Hikino Y., Arai T., Kawahara M., Konno C., Arihara S. and Hikino H. (1967a) Isolation of insect moulting substances from *Matteuccia struthiopteris*, *Lastrea thelypteris*, and *Onoclea sensibilis*. *Chemical Pharmaceutical Bulletin* 15, 1816.

Tan C.Y. (2002) Research on the chemical constituents and pharmaceutical activities of *Cyanotis arachnoidea* C.B. Clarke. PhD. Thesis, Shenyang Pharmaceutical University, Shenyang, China, pp. 151.

Tan C.Y., Wang J.H., Li X., Meng D. and Li X. (2001) Constituents of phytosterone in *Cyanotis arachnoidea* C.B. Clarke. *Journal of Shenyang Pharmaceutical University* 18(4), 263-265 [in Chinese].

Tan C.Y., Wang J.H., Zhang H. and Li X. (2002a) A new phytosterone from *Cyanotis arachnoidea*. *Journal of Asian Natural Products Research* 4(1), 7-10.

Abstract: From *Cyanotis arachnoidea* C.B. Clarke, a new phytosterone named cyanosterone A (1), along with three known compounds--beta-ecdysone (2), ajugasterone C (3) and beta-sitosterol (4) were isolated. The structure of the new compound was determined as 3beta,14alpha,20R,22R-tetrahydroxy-5alpha-cholest-7-en-6-one on the basis of physicochemical properties and spectral analysis.

Tan C.Y., Wang J.H., Xiao W. and Li X. (2002b) A new phytosterone from *Cyanotis arachnoidea*. *Chinese Chemical Letters* 13(3), 245-246.

Tan C.-Y., Wang J.-H. and Li X. (2003a) Phytoecdysteroid constituents from *Cyanotis arachnoidea*. *Journal of Asian Natural Products Research* 5(4), 237-240.

Abstract: Four phytoecdysteroids that have only 19 or 21 carbons, named 11alpha-hydroxyrubrosterone (1), dihydroxyrubrosterone (2), rubrosterone (3) and poststerone (4), were isolated from the whole plant of *Cyanotis arachnoidea* C.B. Clarke. Among them, 1 was a new compound. Their structures were elucidated by spectroscopic methods.

Tan C.Y., Wang J.H., Li X., Du Y.G. and Bai X.F. (2003b) Chemical constituents of *Cyanotis arachnoidea*. *Yao Xue Xue Bao* 38(10), 760-762 [in Chinese].

Abstract:

Aim: To investigate the chemical constituents of *Cyanotis arachnoidea*.

Methods: By using chromatographic methods for separation and combination with spectral analysis, their chemical structures were determined.

Results: Six compounds were identified as ajugasterone C-20, 22-acetonide (1), 20-hydroxyecdysone-20, 22-acetonide (2), 22-oxo-ajugasterone C (3), 22-oxo-20-hydroxyecdysone (4), beta-sitosterol (5), daucosterol (6).

Conclusion: Compound 3 is a new compound, 4 was a new natural compound.

Tan C-y., Wang J-h., Li X., Du Y-g. and Bai X-f. (2005) Study on chemical constituents of *Cyanotis arachnoidea*. Chinese Pharmaceutical Journal (20), 1537-1538 [in Chinese].

Abstract:

OBJECTIVE: To study the chemical constituents of *Cyanotis arachnoidea* C. B. Clarke. METHODS: The compounds were isolated by column chromatography. The structures were identified by their physico-chemical properties and spectral data respectively. RESULTS: Four compounds were elucidated as 20-hydroxyecdysone-2, 3, 20, 22-diacetonide (I), 20-hydroxyecdysone-2, 3-acetonide (II) and isovitexirone (III) and stachysterone D(IV).

CONCLUSION: All compounds are isolated from *Cyanotis* genus for the first time.

Tan C., Kong L., Li X., Li W. and Li N. (2011) Isolation and analysis of a new phytoecdysteroid from *Cyanotis arachnoidea* C.B. Clarke. Chinese Journal of Chromatography 29(9) 937-941 [in Chinese, with an English abstract].

Abstract: *Cyanotis arachnoidea* is a plant with plenty of phytoecdysteroid. To study the active compound in it, a new phytoecdysteroid with 5 α -cholesta skeleton, was isolated from the whole plant of *Cyanotis arachnoidea* C. B. Clarke by using various chromatographic methods (alumina column chromatography, silica gel column chromatography, octadecyl silane (ODS) column chromatography, thin layer chromatography (TLC) and high performance liquid chromatography (HPLC)). Its structure was analyzed on the basis of 1D and 2D nuclear magnetic resonances (NMR), electrospray ionization mass spectrometry (ESI-MS) methods. It is a compound with structure of 3 β ,14 α , 14 α ,20R,22R,25-hexahydroxy-5 α -cholest-7-en-6-one, which is a rare phytoecdysteroid with 5 α -H.

Tanaka N. and Matsumoto T. (1993) Characterization of *Ajuga* plants regenerated from hairy roots. Shokubutsu Soshiki Baiyo 10(1) 78-83 [in Japanese]/ Plant Tissue Culture Letters 10(1) 78-83 [in English].

20-Hydroxyecdysone (20-HE), one of the phytoecdysteroids in practical possibilities for pest control¹), chemotherapy²), and silk production³). In particular, al.³) have demonstrated the use of 20-HE as a facilitator for the maturation of silkworms just before spinning cocoons. We have reported the establishment of a rapidly growing root clone producing a high amount of steroid in *Ajuga reptans* var. *atropurpurea*, one of the phytoecdysone-producing plants, by transformation with *Agrobacterium rhizogenes*⁴). Hairy roots have the ability to regenerate plants⁵). The plants regenerated from hairy roots also have some special characteristics, so-called 'hairy-root syndrome', such as active formation of adventitious root, high growth rate of roots in culture, reduced apical dominance in both stems and roots, altered leaf and flower morphology, plagiotropic root growth (i. e. with altered geotropism) and reduced pollen and seed production⁵). The dwarfism by shortened internodes is also recognized in regenerated plants. In this report, we demonstrate *Ajuga* plants regenerated from hairy roots, and their various properties such as growth, morphology of leaf, root mass, opine biosynthesis, and 20-HE productivity.

Tanaka N., Uozumi N., Kobayashi T. (1999) Genetic transformation of *Ajuga reptans*. Biotechnology in Agriculture and Forestry 45 (Transgenic Medicinal Plants, Ed. Bajaj Y.P.S.), Springer-Verlag, pp. 30-46.

Abstract: *Ajuga reptans*, a member of the Labiatae, is a horticultural plant that produces blue flowers in spring and is used as ground cover. The plant originated in Europe and has been utilized as a medicinal herb for treatment of jaundice and rheumatism in European countries for centuries. The roots of *A. reptans* contain some steroids, such as 20-hydroxyecdysone (20-HE), ajugasterone, cyasterone, and norcyasterone, which are phytoecdysteroids (Fig. 1B,D; Tomas et al. 1992; Camps and Coll 1993). 20-HE, is identical to β -ecdysone, is a principal physiological inducer of molting and metamorphosis in arthropoda. The physiological properties of 20-HE in insects and in mammals have been investigated in many laboratories, and the possibility of their practical use in pest control (Kubo et al. 1983), in chemotherapy (Yoshida et al. 1971), and in silk production (Kozakai et al. 1990) has been demonstrated. Indeed, a Japanese company has recently succeeded in developing and selling 20-HE as a control chemical for the spinning of cocoons by silkworms.

Tang H-j., Fan C-j., Wang G-y., Wei W., Wang Y. and ye W-c. (2014) Chemical constituents from root tubers of *Serratula chinensis*. Zhongcaoyao (Chinese Traditional and Herbal Drugs) (7), 906-912 [in Chinese, with an English abstract].

Abstract: The chem. constituents from the root tubers of *Serratula chinensis* were studied. The chem. constituents were separated and purified by silica gel, Sephadex LH-20, ODS column chromatog., and preparative HPLC. Their structures were determined by physicochem. properties and spectral data. Sixteen compounds (ecdysteroids) were

isolated from the n-butanol fraction of 95% ethanol extract in the root tubers of *S. chinensis*, and their structures were identified as 20-hydroxyecdysone (1), polypodine B (2), carthamosterone (3), 20-hydroxyecdysone-20, 22-monoacetone (4), 24-epi-abutasterone (5), polypodine C (6), coronasterone (7), 20-hydroxyecdysone 2-O- β -D-glucopyranoside (8), 20-hydroxyecdysone 25-O- β -D-glucopyranoside (9), polypodine B 20,22-acetone (10), shidasterone (11), 2-O-acetyl-20-hydroxyecdysone (12), 3-O-acetyl-20-hydroxyecdysone (13), 20-hydroxyecdysone-20,22-butylidene acetal (14), 24-methylene-shidasterone (15), and ajugasterone D (16). Compounds 2, 4, 5, 7-10, 12, 15, and 16 are isolated from this plant for the first time, and compounds 5, 7-10, and 16 are found in the plants of genus *Serratula* L. for the first time.

Tang W-w., Liang X-k., Ma C-h., Lei J-w., Gong H-y., Ji L., Xie C-x. (2019) Study on fingerprint of *Achyranthes bidentata* in different harvesting seasons. *Journal of Chinese Medicinal Materials* (9), 2079-2085 [in Chinese].
Abstract: Objective: To compare the HPLC fingerprint of *Achyranthes bidentata* in different harvesting seasons in order to explore the influence of harvesting seasons on internal quality of *Achyranthes bidentata*. Methods: Samples were prepared by ultrasonic extraction and HPLC fingerprint was established; The quality of *Achyranthes bidentata* in different harvesting seasons was studied by similarity evaluation, grey correlation analysis and principal component analysis and the differences were compared. Results: Twenty-one and twenty-two common peaks were demarcated by fingerprint pattern of *Achyranthes bidentata* harvested in winter and spring, respectively. The similarity ranged from 0.994 to 1.000 and 0.997 to 1.000, respectively. There were β -ecdysterone, 25R-inokosterone and 25S-inokosterone in *Achyranthes bidentata* collected in two seasons. The results of differential analysis showed that the chromatographic peak 1 was the characteristic peak of *Achyranthes bidentata* in winter and peak 4 was the characteristic peak in spring. The results of grey correlation analysis and principal component analysis showed that the quality of *Achyranthes bidentata* in spring was better than that in winter. Conclusion: The contents and types of chemical constituents of *Achyranthes bidentata* are different in different harvesting seasons. According to the results of this study, the harvest season is suggested to be spring.

Tang X., Pei G., Zhou Z-y. and Tan J-w. (2013) Chemical Constituents from Roots of *Achyranthes bidentata*. *Journal of Tropical and Subtropical Botany* 21(1), 57-62

Tao H-m., Wang L-s., Zhao D-q., Zhu Q-h., Yin Y-g. and Liu Y-h. (2012) Steroids from tubers of *Asparagus filicinus*. *Zhongcaoyao (Chinese Traditional and Herbal Drugs)* 43(9) 1716-1720.

Abstract: Steroids from tubers of *Asparagus filicinus* were investigated. *A. filicinus* was extracted by 90% ethanol and isolated and purified by Silica, Sephadex LH-20 and PR-18 column, and the constituents were identified by ¹H-NMR and ¹³C-NMR. Sixteen steroid compounds were obtained and elucidated as aspafileoside A (1), asparoside A (2), filiasparoside C (3), aspafileoside B (4), asparagusin A (5), Asp-IV (6), aspafileoside C (7), aspafileoside D (8), calonysterone (9), 5-deoxykaladasterone (10), 25-hydroxydacryhainansterone (11), stachysterone (12), stachysterone B (13), ecdysterone (14), β -ecdysterone (15), 20-hydroxyecdysine-20-22-monocetone (16). Compounds 2, 5 and 12 were isolated from *A. filicinus* for the first time, and compound 12 was isolated from *Asparagus* for the first time.

Tao Yi, Du Y-s., Huang S-r., Li W-d. and Cai B-c. (2017) Analysis of chemical constituents in different processed products of *Achyranthes bidentata* Radix by UPLC-Q-TOF/MS. *Chinese Journal of Experimental Traditional Medical Formulae* (12), 1-5 [in Chinese].

Abstract: Objective: To compare the quality control indexes and chemical constituents of crude and processed products of *Achyranthes bidentata* Radix. Method: According to standard in the 2015 edition of Chinese Pharmacopoeia, the quality control indexes and chemical constituents of crude and processed products of *Achyranthes bidentata* Radix were investigated, such as water content, total ash content and alcohol-soluble extract content. UPLC-Q-TOF/MS was established to compare the contents of major constituents in crude and processed products of *A. bidentata* Radix. Moreover, the linearity, precision, stability, repeatability and recoveries of the approach were well validated. Result: The water content, total ash content and alcohol-soluble extract content of crude and processed products of *A. bidentata* Radix all met the requirements of the 2015 edition of Chinese Pharmacopoeia. Ten major constituents were tentatively deduced. Compared with these of the crude products, the contents of benzyl glucoside, benzyl glucoside isomer, polypodine B, β -ecdysterone and ginsenoside Ro in the wine processed products were significantly increased. The contents of zingibroside R₁, bidentatoside I and chikusetsusaponin IV were significantly decreased. For the salt processed products, the contents of benzyl glucoside, benzyl glucoside isomer, achyranthesterone A, polypodine B, β -ecdysterone and inokosterone were significantly increased. The contents of zingibroside R₁, ginsenoside Ro, bidentatoside I and chikusetsusaponin IV were significantly decreased. Conclusion: The contents of phenolic glycosides and steroidal saponins are significantly increased, while the content of triterpenoid saponins is significantly decreased, which may be degraded under heating condition.

Tao Y., Yan J. and Cai B. (2019a) A liquid chromatography-tandem mass spectrometry approach for study the tissue distributions of five components of crude and salt-processed *Radix Achyranthes* in rats. *Biomedical Chromatography* **33**, e4483 (doi: 10.1002/bmc.4483).

Abstract: This study developed a robust and reliable approach using liquid chromatography- tandem mass spectrometry for the simultaneous determination of five saponins in rat tissues: β -ecdysterone, chikusetsusaponin IV, ginsenoside Ro, 25S-inokosterone and chikusetsusaponin IVa. This is the first report on a comparative tissue distribution study of crude and salt-processed *Radix Achyranthes* in rats. After one-step protein precipitation by acetonitrile, the tissue samples were sent to LC-MS/MS for multiple reaction monitoring. The retention times of the five saponins and internal standard were 1.77, 3.14, 3.01, 1.83, 3.26 and 4.77 min. The standard curves showed good linear regression ($r^2 > 0.9991$) in the range of 10.3-1562.5 ng/mL. The intra- and inter-day accuracy and precision were within 15% of the nominal concentration. The recoveries of the five saponins were 92.0-99.9%. Finally, this approach was successfully applied to tissue distribution analysis of the five saponins after oral administration of crude and salt-processed *Radix Achyranthes* in rats. The largest concentration of the five saponins was observed in kidney after salt-processing, which indicated that processing could enhance the bioavailability.

Tao Y., Huang S., Yan J. and Cai B. (2019b) Determination of major components from *Radix Achyranthes bidentata* using ultra high performance liquid chromatography with triple quadrupole tandem mass spectrometry and an evaluation of their anti-osteoporosis effect in vitro. *Journal of Separation Science* **42**: 2214-2221 (doi:10.1002/jssc.20190014b).

Abstract: Ecdysterone and saponins are the most characteristic components of *Radix Achyranthes bidentata*, which acts on the human body to promote collagen synthesis and stimulates cell growth. However, the relationship between these components and the differentiation of MC3T3-E1 osteoblastic cells is unknown. We developed a rapid ultra high performance liquid chromatography with triple quadrupole tandem mass spectrometry method for direct determination of one ecdysterone and four saponins in crude and salt-processed *Radix Achyranthes bidentata*. The method was interrogated in terms of linearity, intra- and inter-day precision, repeatability, stability and recovery. The method was linear within the concentration ranges of 0.003-336 $\mu\text{g/mL}$ for β -ecdysterone, 0.0035-130 $\mu\text{g/mL}$ for 25S-inokosterone, 0.004-423 $\mu\text{g/mL}$ for ginsenoside Ro, 0.0036-66 $\mu\text{g/mL}$ for chikusetsusaponin IV and 0.0044-111 $\mu\text{g/mL}$ for chikusetsusaponin IVa. The intra- and inter-day precisions were all within 2.7%. The standard addition method determined recovery rates for each component (98.7-102.5%). The method was successfully applied to simultaneously quantify five components in ten batches of crude and salt-processed *Radix Achyranthes bidentata*. Subsequently, the examination of these extracts on the differentiation of MC3T3-E1 osteoblastic cells were carried out. Finally, the relationships between the contents of five components and their anti-osteoporosis effect were investigated by using canonical correlation analysis.

Taralkar S.V. and Chattopadhyay S. (2018) Study of engineering principles for extraction of ecdysterone from leaves of *Achyranthes aspera* linn (aghada). *KAHV International Journal of Science, Engineering & Technology* **5**(4), 36-42.

Tarkowská D., Krampolová E. and Strnad M. (2020) Plant triterpenoid crosstalk: the interaction of brassinosteroids and phytoecdysteroids in *Lepidium sativum*. *Plants* **9**, 1325.

Abstract: Plant steroid alcohols, plant sterols, are essential components of cell membranes that perform many functions. Their most prominent function is maintaining membrane semipermeability and regulating its fluidity through their specific interaction with phospholipids and membrane proteins. This work is focused on the study of the interaction of two groups of plant sterols, brassinosteroids (BRs) and phytoecdysteroids (PE). Steroid substances belonging to both groups are important signaling molecules essential for plant growth and development, but while the first group has all the known attributes of plant hormones, the second lacks hormonal function in plants. The aim of this preliminary study was to determine at what concentration level and to what extent substances of this type are able to interact with each other, and thus influence the early growth and development of a plant. It was found that exogenously applied PE 20-hydroxyecdysone (20E) significantly reduced the level of endogenous BRs in four-day-old garden cress (*Lepidium sativum*) seedlings. On the other hand, exogenously applied BRs, 24-*epi*brassinolide (*epi*BL), caused the opposite effect. Endogenous 20E was further detected at the picogram level in garden cress seedlings. Thus, this is the first report indicating that this plant species is PE-positive. The level of endogenous 20E in garden cress seedlings can be decreased by exogenous *epi*BL, but only at a relatively high concentration of $1 \cdot 10^{-6}$ M in a culture medium. The image analysis of garden cress seedlings revealed that the length of shoot is affected neither by exogenous BRs nor PE, whereas the root length varies depending on the type and concentration of steroid applied.

Temirgaziev B.S., Salkeyeva L.K., Agitayeva G.S., Kozhanova A.M., Tuluev B.I. and Adekenov S.M. (2016) Regioselective synthesis of new phosphorus- and nitrogen-2-deoxyecdysone-based derivatives. Abstract of a poster

presentation at the 23rd Conference on Isoprenoids. Proceedings of the National Academy of Sciences of Belarus: Chemical Series (3), 114.

Temirgaziev B.S., Tuleuov U.B., Baizhigit E.A., Minayeva Y.V., Salkeyeva L.K., Tuleuov B.I. and Adekenov S.M. (2018) Optimization of the technology for obtaining ecdysterone from *Serratula coronata* by varying the extraction methods and growth phases. Vestnik – Chemistry Series 90(2), 45-50.

Abstract: A complex study of the aerial part of *Serratula coronata* L. cultivated in the collection area of medicinal plants of the IRPH «Phytochemistry» (Karaganda) in different phases of growth and using the most optimal extraction methods has been carried out. The content of the main active component of ecdysterone (20E) has been studied. Investigation of the seasonal dynamics of ecdysterone distribution under conditions of varying extraction methods shows that its maximum accumulation is observed during the vegetative phase, and the optimal method in this phase is extraction with isobutyl alcohol, the extract of which contains 13.86 % of ecdysterone and the maceration method with 96.2 % ethyl alcohol with a 20E content of 12.03 %, respectively. It is shown that the maceration with 96.2 % ethyl alcohol is technologically optimal, which fully complies with the international standards of good manufacturing practice (GMP) under pharmaceutical production conditions and excludes the use of toxic and expensive isobutyl alcohol solvent. It has been found that the content of ecdysterone from the beginning of vegetation to the final phase goes down, which is confirmed by the data of high-performance liquid chromatography (HPLC). It is assumed that there is an outflow of ecdysterone to the root system, and then its redistribution occurs as the plant develops further with a partial discharge into the soil. Based on the data on the quantitative content of the target component by the HPLC method, it is recommended that for the preparation of the ecdysterone substance of many actoprotective phytopreparations and valuable WS, the preparation of the above-ground biomass of *Serratula coronata* L. should be carried out during the vegetation phase of this taxon.

Temirgaziev B.S., Kucakova K., Baizhigit Y.A., Jurasek M., Dzubak P., Hajduch M., Dolensky B., Drasar P.B., Tuleuov B.I. and Adekenov S.M. (2018a) Bioavailability and structural study of 20-hydroxyecdysone complexes with cyclodextrins. Steroids (doi.org/10.1016/j.steroids.2018.11.007).

Abstract: 20-Hydroxyecdysone – (2 β ,3 β ,5 β ,22R)-2,3,14,20,22,25-hexahydroxycholest-7-en-6-one was isolated in satisfactory yield using ethanol extraction from the aerial part of *Silene wolgensis* (Hornem.) Otth; sometimes *Silene wolgensis* (Willd.) Bess. ex Spreng. The complexation of the phytoecdysteroid with β -cyclodextrin was studied by NMR spectroscopy. By studying the changes in chemical shifts of protons of substrates and receptors it was found that ecdysterone interacts with cyclodextrins to form supramolecular inclusion complexes of stoichiometric composition of 1:1 or 1:2. Ecdysterone- β -cyclodextrin complexes exhibit 100 times higher solubility in water than the parent compound.

Temirgaziev B.S., Tuleuov B.I., Romanova M.A., Seidakhmetova R.B., Seilkhanov T.M., Seilkhanov O.T., Salkeyeva L.K. and Adekenov S.M. (2019) Supramolecular complexes of 3-epi-2-deoxyecdysone with cyclodextrins and their anti-inflammatory activity. Russian Journal of General Chemistry 89(3), 424-428.

Abstract: The ecdysteroid 3-epi-2-deoxyecdysone has been isolated from the aerial part of *Acanthophyllum gypsophiloides* Regel. The complexes formation of the ecdysteroid with α -, β -, γ -, and 2-hydroxypropyl- β -cyclodextrins has been studied by means of NMR spectroscopy. Anti-inflammatory activity of the obtained 3-epi-2-deoxyecdysone complexes with cyclodextrins has been investigated.

Temirgaziev B.S., Seilkhanov T.M., Tyanakh S., Kozhanova A.M., Seilkhanov O.T., Minayeva Y.V., Sal'keeva L.K., Tuleuov B.I. and Adekenov S.M. (2018b) Obtaining and investigation of supramolecular inclusion complex of 2-deoxy-20-hydroxyecdysone with γ -cyclodextrin by NMR spectroscopy method. Kazakh Chemical Journal (2), 36-44 [in English, with abstracts in Kazakh and Russian].

Abstract: For the first time, 2-deoxy-20-hydroxyecdysone (2-deoxyecdysone) has been isolated from the above-ground part of *Silene fruticulosa* (Pall.) Schischk (Caryophyllaceae Juss. family). The formation of complex of phytoecdysteroids with γ -cyclodextrin has been studied with the help of NMR spectroscopy. Due to changing the chemical shifts of the substrate and receptor protons, it has been revealed that 2-deoxy-20-hydroxyecdysone interacts with γ -cyclodextrin to form a supramolecular inclusion complex of the stoichiometric composition of 1:1 with the entry of the fragment A of the substrate molecule into the inner cavity of the receptor.

Termentzi A., Croizat C., Collin K., Morvan P.Y., Vallée R., Halabalaki M. and Skaltsounis L. (2014) HPLC-HRMS/MS analysis of saponins and phytoecdysteroids from a *Silene colorata* hydroglyceric extract. *Planta Medica* 80, p207.

Thanonkeo S., Chamnipa N. And Thanonkeo P. (2011) Induced accumulation of 20-hydroxyecdysone in cell suspension cultures of *Vitex glabrata* R.Br. *African Journal of Biotechnology* 10(52), 10612-10617.

Abstract: This study describes the effects of culture medium, culture temperature, sucrose concentration and cholesterol feeding on cell growth and 20-hydroxyecdysone production in suspension cultures of *Vitex glabrata*, an important medicinal plant in Thailand. Cell growth and 20-hydroxyecdysone production were not significantly different when cells were cultivated on B5 or half-strength MS medium. However, cultivation of *V. glabrata* cell cultures at 25°C yielded 1.06- and 1.09-fold higher values of cell growth and 20-hydroxyecdysone content, respectively than those at 30°C. Sucrose at 30 and 40 g/L favors the production of 20-hydroxyecdysone in suspension cultures of *V. glabrata*. Feeding of cholesterol at 5 mg/L, as precursor for biosynthesis of 20-hydroxyecdysone, yielded 1.11-fold higher accumulation of 20-hydroxyecdysone than the control cells. Increasing of cholesterol to 10 mg/L resulted in decreased production of 20-hydroxyecdysone. Key words : *Vitex glabrata*, 20-hydroxyecdysone, suspension culture, cholesterol.

Thoa N.T.K., Ban N.K., Trang D.T., Linh T.M., Giang V.H., Nhiem N.X. and Kiem P.V. (2018) Ecdysteroids from leaves of *Vitex trifolia*. *Vietnam Journal of Chemistry* 56(2), 162-166.

Abstract: Four known ecdysteroids, ecdysone (**1**), 20-hydroxyecdysone (**2**), 20-hydroxyecdysone 2,3-monoacetone (**3**), and turkesterone (**4**) were isolated from leaves of *Vitex trifolia*. The structure of these compounds was elucidated by means of 1D- and 2D-NMR spectra and was compared with those reported in literature. Compound **3** was reported from *Vitex* genus for the first time; compounds **1**, **2**, and **4** from *V. trifolia* for the first time.

Thuy T.T., Porzel A., Ripperger H., Sung T.V. and Adam G. (1998) Chalcones and ecdysteroids from *Vitex leptobotrys*. *Phytochemistry* 49(8), 2603-2605.

Abstract: In addition to some known chalcones and ecdysteroids three new chalcones have been isolated from aerial parts of *Vitex leptobotrys*, the structures of which have been identified as 2',4'-dihydroxy-4,6'-dimethoxychalcone, 4'-hydroxy-4,2',6'-trimethoxychalcone and 4,2',4', β -tetrahydroxy-6'-methoxy- α,β -dihydrochalcone, respectively.

Thuy T.T., Tam N.T., Anh N.T.H., Hau D.V., Phong D.T., Thang L.Q., Adorisio S., Sung T.V. and Delfino D.V. (2017) 20-Hydroxyecdysone from *Dacrycarpus imbricatus* bark inhibits the proliferation of acute myeloid leukemia cells. *Asian Pacific Journal of Tropical Medicine* 10(2), 157-159 (or 151-153?).

Abstract:

Objective: To investigate the anti-proliferative effects of 20-hydroxyecdysone isolated from the bark of *Dacrycarpus imbricatus* (Blume) de Laub.

Methods: Column chromatography was used for isolation of compounds from plant material. The structure of the isolated compound was identified by mass spectrometry and nuclear magnetic resonance techniques, including HSQC, HMBC, NOE-difference experiments. The isolated compound was tested for its anti-proliferative activity in acute myeloid leukemia (AML) and OCI-AML cells.

Results: Compound 1 was isolated from the ethyl acetate fraction of *Dacrycarpus imbricatus* barks by column chromatography. Its chemical structure was identified as 20-hydroxyecdysone (20HE), a cholestane-type ecdysteroid, by a combination of mass spectrometry and nuclear magnetic resonance spectrometric analyses. Our goal was to test the anti-proliferative activity of 20HE using the OCI-AML cell line. 20HE significantly decreased OCI cell number at a concentration of 1 mg/mL, whereas lower concentrations were ineffective. Moreover, this decrease was due to partial blockage of the G₁/S phase of the cell cycle, with a reduction of cells in the G₂M phase, not due to increased apoptosis.

Conclusions: This indicates that 20HE significantly decreases the number of cells in the G₁/S phase of the cell cycle in human AML cells. This is the first time that the anti-proliferative activity of 20HE against a human tumor cell line has been reported.

Keywords: 20-Hydroxyecdysone; Acute myeloid leukemia; *Dacrycarpus imbricatus*; Ecdysteroid; Proliferation.

Tijjani A., Shettima Y.A., Abdulraman F.I., Khan I.Z., Tom G.M. and Cong L.I. (2017) Isolation and structural elucidation of 20-hydroxyecdysone from *Vitex doniana* Sweet stem bark (black plum) Mustapha. *Medicinal Chemistry (Los Angeles)* 7(3), 828-831.

Abstract: *Vitex doniana* Sweet, a plant commonly known black plum, in English, Prunier noir in French, dinya in Hausa, ucha koro in Igbo, oori-nla in Yoruba and ngarmi in Kanuri is a medium-sized deciduous tree, 8-18 m high, with a heavy rounded crown and a clear bole up to 5 m. *V. doniana* is from Verbenaceae family and abundantly occurring in savannah regions. It can be found throughout tropical Africa. The ethanolic extract of *Vitex doniana* stem bark (11.9 g) was subjected to a silica gel accelerated column chromatography and eluent fractions (150 ml aliquots) obtained were collected and monitored with thin layer chromatography (TLC). Fractions with similar R_f values from same solvents system were pooled together. Phytochemical test of all the fractions were performed. Complete elution yielded 48 fractions (150 ml/fraction) which were pooled to 24 fractions and finally to eight (8) fractions and coded. Fraction Vd8-a (56 mg) has given a single spot a white crystal compound coded V1 on checking with TLC and observed under Ultraviolet lamp. The R_f value was calculated to be 0.433 and melting point

was found to be 241-243°C uncorrected. The infrared spectrum of compound V1 shows prominent peaks that corresponds to OHstr (3365 cm⁻¹) and C=O (1652 cm⁻¹). The ¹H NMR (400 MHz) spectrum of compound V1 in DMSO-d₆ displayed five singlet signals. It further showed a broad singlet at δ 5.58 integrated for 1 H is due to an olefinic H-atom adjacent to the carbonyl carbon atom. Three signals at δ 3.10' (d, J = 9.0 Hz, H-22), 3.59 (m, 1H, 2H-a) and 3.72 (m, 1H, 3H-e) each integrating for one proton is due to an oxymethine protons indicating that three oxymethine H-atoms were present in the compound. The ¹³C-NMR spectrum showed the presence of 27 Carbon atoms, suggesting that may be steroid skeleton and the DEPT-135 spectra showed the presence of five CH₃, eight CH₂, and seven CH groups, and seven quaternary C-atoms. The Molecular formula was established as C₂₇H₄₄O₇ by HRES-MS positive ion mode m/z 481.3179. Based on the spectral analysis, the compound V1 is thus concluded to have ecdysteroid skeleton and conclusively conforms with 2β, 3β 14α, 20R, 22R, 25- hexahydroxy-5 β cholest- 7-ene-6- one, commonly known as 20-hydroxyecdysone. This is the first time this compound was isolated from Vitex doniana sweet.

Timofeev N.P. (2017b) Composition of 65 ecdysterone analogues from *Leuzea*: their activity and yield fro roots, seeds and leaves. M. Vniissok (3), 75-78 [in Russian].

Timofeev N.P. and Linerov V.V. (2017a) Contents of ecdysterone and analogues in liquid extract of *Leuzea carthamoides* from root with rhizomes. M. Vniissok (3), 68-71 [in Russian].

Timofeev NP, Volodin VV and Frolov JM (1998) Distribution of 20-hydroxyecdysone in the structures of the above-ground biomass of *Rhaponticum carthamoides* (Willd.) Ilijn. under conditions of agrocoenosis in the Komi Republic. Rastelny Resursy 34(3), 63-68 [in Russian, with an English abstract].

Tomás J., Camps F., Claveria E., Coll J., Melé E. and Messeguer J. (1992) Composition and location of phytoecdysteroids in *Ajuga reptans* *in vivo* and *in vitro* cultures. Phytochemistry 31(5), 1585-1591.

Abstract: The location and concentration of phytoecdysteroids in *Ajuga reptans* have been studied in different normally grown or *in vitro* micropropagated plants. Some callus cultures were also studied. The relationship of phytoecdysteroid relative concentration with growing conditions and source of tissue are discussed. The ratio of C₂₈/C₂₉ phytoecdysteroids was established amongst the four major compounds (29-norsengosterone and 29-norcyasterone as C₂₈, and cyasterone and ajugalactone as C₂₉) which between them account, on average, for 92% of the total phytoecdysteroid content. This ratio was found to be less than one in all types of leaves from wild material, slightly higher than one in the roots of wild plants, and in the range from three to five in greenhouse and *in vitro* plants. Micropropagated plants had an extremely low phytoecdysteroid content in leaves, whereas that in roots was the highest detected in our experiments. Callus cultures obtained from leaves or roots completely lost their capacity to produce ecdysteroids.

Tomás J., Camps F., Coll J., Melé E. and Messeguer J. (1993) Phytoecdysteroid production by *Ajuga reptans* tissue cultures. Phytochemistry 32(2), 317-324.

Tomita Y and Sakurai E. (1974) Biosynthesis of phytoecdysone: incorporation of 2β,3β,14α-trihydroxy-5β-cholest-7-en-6-one into β-ecdysone and inokosterone in *Achyranthes fauriei*. Journal of the Chemical Society, Chemical Communications 434-435.

Abstract: 2β,3β,14α-Trihydroxy-5β-[3α-³H]cholest-7-en-6-one was incorporated into both β-ecdysone and inokosterone, equally; thus hydroxylation of the side chain of phytoecdysone occurs after formation of the A–B ring system.

Tóth G., Herke I., Gati T., Vagvolgyi M., Berkecz R., Parfenova L.V., Ueno M., Yukoi T., Nakagawa Y. and Hunyadi A. (2021) A commercial extract of *Cyanotis arachnoidea* roots as a source of unusual ecdysteroid derivatives with insect hormone receptor binding activity. Journal of Natural Products (<https://doi.org/10.1021/acs/jnatprod.0c01274>).

Abstract: Ecdysteroids act as molting hormones in insects and as nonhormonal anabolic agents and adaptogens in mammals. A wide range of ecdysteroid-containing herbal extracts are available worldwide as food supplements. The aim of this work was to study such an extract as a possible industrial source of new bioactive ecdysteroids. A large-scale chromatographic isolation was performed from an extract of *Cyanotis arachnoidea* roots. Ten ecdysteroids (1–10) including eight new compounds were isolated and characterized by extensive nuclear magnetic resonance studies. Highly unusual structures were identified, including a H-14β (1, 2, 4, and 10) moiety, among which a 14β(H)17β(H) phytosteroid (1) is reported for the first time. Compounds with an intact side chain (4–10) and 11 other natural or semisynthetic ecdysteroids (11–21) were tested for insect ecdysteroid receptor (EcR) binding activity. Two new compounds, i.e., 14-deoxydacryhainansterone (5) and 22-oxodacryhainansterone (6), showed strong EcR binding activity (IC₅₀ = 41.7 and 380 nM, respectively). Six compounds were identified as EcR agonists

and another two as antagonists using a transgenic ecdysteroid reporter gene assay. The present results demonstrate that commercial *C. arachnoidea* extracts are rich in new, unusual bioactive ecdysteroids. Because of the lack of an authentic plant material, the truly biosynthetic or artifactual nature of these compounds cannot be confirmed.

Tóth I., Báthory M., Szendrei K., Minker E and Blazsó G. (1981) Ecdysteroids in Chenopodiaceae: *Chenopodium album*. *Fitoterapia* **52**, 77- 80.

Tóth N. (2010) Ecdysteroid profile of *Silene viridiflora* and the effect of 20-hydroxyecdysone on rat muscle fibres *in vivo*. Ph.D. Thesis, University of Szeged, Hungary, pp. 66

Tóth N. and Báthori M. (2008) Preparative-scale chromatography of ecdysteroids: a class of biologically active steroids. *Journal of Chromatographic Science* **46**, 111-116.

Abstract: A simple separation procedure is developed for the isolation of the main phytoecdysteroid 20-hydroxyecdysone from the herb *Silene viridiflora*. The purification in four steps uses only a simple preparative-scale separation technique (i.e., liquid-liquid extraction, precipitation, solid-phase extraction on octadecyl silica, and crystallization). This procedure is extended using classical normal-phase liquid column chromatography, rotation planar chromatography, and preparative high-performance liquid chromatography for the isolation of the minor ecdysteroids: integristerone A, 26-hydroxypolypodine B, 2-deoxy-20,26-dihydroxyecdysone, and polypodine B. 2-Deoxy-20,26-dihydroxyecdysone is isolated from this species for the first time. The isolation of these ecdysteroids in adequate amounts makes them readily available for insect physiology experiments and for structure-activity relationship studies. The preparative-scale separation work also results in a minor, as yet unknown ecdysteroid.

Tóth N., Simon A., Tóth G., Kele Z., Hunyadi A. and Báthori M. (2008) 26-Hydroxylated ecdysteroids from *Silene viridiflora*. *Journal of Natural Products* (published on Web: 29/07/2008).

Abstract: Four new 26-hydroxylated phytoecdysteroids, 2-deoxy-5,20,26-trihydroxyecdysone (1), 5,20,26-trihydroxyecdysone 20,22-acetonide (2), 2-deoxy-5,20,26-trihydroxyecdysone 20,22-acetonide (3), and 20,26-dihydroxyecdysone 20,22-acetonide (4), were isolated from the herb *Silene viridiflora*, and their structures were elucidated by means of one- and two-dimensional NMR and mass spectrometry.

Tsiftoglou O.S., Stefanakis M.K. and Lazari D.M. (2018) Chemical constituents isolated from the rhizomes of *Helleborus odoratus* subsp. *cyclophyllus* (Ranunculaceae). *Biochemical Systematics and Ecology* **79**, 8-11.

Abstract: Chemical investigation of butanol extract from the rhizomes of *Helleborus odoratus* subsp. *cyclophyllus* led to the isolation of nine natural products, which are identified, on the basis of MS and NMR spectra as 2-hydroxymethyl- D-ribo- γ -lactone (1), uridine (2), 2-(3,4-dihydroxy)-phenyl-ethyl- β -D-glucopyranoside (3), the bufadienolides deglucohellebrin (4) and hellebrin (5), the furostanols caucasicoside A (6) and helleboroside B (7) and ecdysones 20-hydroxyecdysone (8) and polypodine B (9). This is the first report on the occurrence of compounds (1)–(3) in the genus *Helleborus*.

Tu W., Zhang L., Liu X. and Li X. (2019) Comparative study of sterones and triterpenoid glycosides contents in raw and processed *Achyranthes bidentata* Radix. *Traditional Chinese Drug Research & Clinical Pharmacology* (1), 89-93 [in Chinese].

Abstract: Objective: To establish an HPLC determination method for sterones and triterpenoid glycosides in *Achyranthes bidentata* radix, and compare the contents in raw and processed *Achyranthes bidentata* radix. Methods: The contents of β -ecdysterone, 25R-inokosterone, 25S-inokosterone, ginsenoside Ro and chikusetsusaponin IVa in raw, cubes and wine stir-fried *Achyranthes bidentata* radix were determined by HPLC simultaneously. Results: In *Achyranthes bidentata* radix cubes, the contents of β -ecdysterone, 25R-inokosterone, 25S-inokosterone were slightly lower than those in raw materials, while the contents of ginsenoside Ro and chikusetsusaponin IVa were significantly reduced. In wine stir-fried *Achyranthes bidentata* radix, the contents of β -ecdysterone, 25R-inokosterone, 25S-inokosterone were slightly increased; while the contents of ginsenoside Ro and chikusetsusaponin IVa were higher than those in *Achyranthes bidentata* radix cubes. Conclusion: The method can be used to control the quality of *Achyranthes bidentata* radix. Different processing methods may have certain influence on the contents of sterones and triterpenoid saponins in *Achyranthes bidentata* radix.

Tulevov B.I. (2016) Some results of study of ecdysteroid-containing plants of Kazakhstan. Abstract of a poster presentation at the 23rd Conference on Isoprenoids. Proceedings of the National Academy of Sciences of Belarus: Chemical Series (3), 59-60.

Tulevov B.I., Turdybekov K.M., Khabdolda G., Adekenov S.M., Nurkenov O.A., Tulevova B.K., Kozhanova A.M., and Almagambetov A.M. (2014) Structure and stereochemistry of phytoecdysone from *Silene cretaceae* Fisch. *Russian Journal of General Chemistry* **84**(4) 704–707.

Abstract: 2-Deoxyecdysone (3 β ,14 α ,22R,25-tetraoxy-5 β -cholest-7-en-6-one) has been isolated from *Silene cretacea* Fisch. Caryophyllaceae. The compound structure has been elucidated by X-ray diffraction studies.

Tuluev B.I., Zavarzin I.V., Shashkov A.S., Chernoburova E.I. and Adekenov S.M. (2018) 3 α ,14 α ,22R,25-Tetrahydroxy-5 β (H)-cholest-7-en-one, a phytoecdysteroid from *Acanthophyllum gypsophiloides* possessing anti-inflammatory and analgesic activities. Russian Chemical Bulletin, International Edition 67(4), 663-666 [in English]/Izvestiya Akademii Nauk Seriya Khimicheskaya (4), 663-666 [in Russian].

Abstract: A representative of the ecdysteroid series, 3 α ,14 α ,22R,25-tetrahydroxy-5 β (H)-cholest-7-en-6-one, possessing anti-inflammatory and analgesic activities, was isolated from a water—ethanol extract of the aerial part of *Acanthophyllum gypsophiloides* Regel after chromatographic removal of non-polar components.

Uozumi N., Makino S. and Kobayashi T. (1995) 20-Hydroxyecdysone production in *Ajuga* hairy root controlling intracellular phosphate content based on kinetic model. Journal of Fermentation and Bioengineering 80(4), 362-368.

Abstract: After phosphate depletion in a batch culture medium, the growth rate of *Ajuga* hairy root decreased and the content of a secondary metabolite, 20-hydroxyecdysone (20-HE), increased. The 20-HE content in *Ajuga* hairy root increased when the intracellular phosphate content was kept less than 5 mg/g-dry weight. Indoleacetic acid (IAA) supplementation (0.1 mg/l) increased the growth rate of *Ajuga* hairy root due to an increase in the number of root apical meristems although it led to a decrease in 20-HE content. From the above results, the culture was divided into two phases: a growth phase where all nutrient components in the medium are present above the critical level, and a secondary metabolite production phase where intracellular phosphate content is kept less than 5 mg/g-dry weight. A kinetic model was constructed to simulate the intracellular phosphate content during the culture. The fed-batch culture in which 0.1 mg/l of IAA was supplemented only at the beginning of the culture, the intracellular phosphate content was kept below 5 mg/g-dry weight after 10 d, and then phosphate was eliminated from the medium after 20 d, was carried out in a turbine-blade reactor by controlling the medium condition according to the conductivity of the culture medium and the simulation data. The culture provided approximately 3-fold increase in 20-HE content, compared to the fed-batch culture without phosphate control.

Urbanska M., Nawrot J., Dawid-Pak R., Kaczerowska-Pietrzak K., Morag M., Ratajczak and Nowak G. (2014) Detection of pharmacological active compounds of the Asteraceae family and their chemotaxonomical implications. Journal of Plant Sciences 2(5), 187-191.

Abstract: It can be assumed that sesquiterpene lactones and natural phytosteroids (ecdysones) are the characteristic compounds of the plants from the Asteraceae family. They display certain pharmacological properties and thus are helpful in chemical descriptions of the Asteraceae tribes, subtribes and species. Also some phenolic glycosides found in genus *Klasea* Cass, and in genus *Centaurea* L. may be of medical and chemotaxonomical significance. Our studies on the species of the aforementioned taxons and the isolation of several compounds allowed for interesting conclusions of phytochemical and taxonomical nature.

Usmanov B.Z., Gorovits M.B. and Abubakirov N.K. (1971) Phytoecdysones of *Ajuga turkestanica*. Khimiya Prirodnikh Soedinenii 535-536 [in Russian].

Usmanov B.Z., Gorovits M.B. and Abubakirov N.K. (1973) Phytoecdysones of *Ajuga turkestanica*. II. Khimiya Prirodnikh Soedinenii (9) 125 [in Russian]/Chemistry of Natural Products 125-126 [in English].
No Abstract.

Usmanov B.Z., Gorovits M.B. and Abubakirov N.K. (1973b) Ecdysterone from *Ajuga chia*. Khimiya Prirodnikh Soedinenii (2), 256-257 [in Russian]/ Chemistry of Natural Compounds (2), 270 [in English].
No Abstract.

Usmanov B.Z., Gorovits M.B. and Abubakirov N.K. (1975) Phytoecdysones of *Ajuga turkestanica*. III. Structure of turkesterone. Khimiya Prirodnikh Soedinenii 466-470 [in Russian].

Usmanov B.Z., Saatov Z. and Abubakirov N.K. (1977) Phytoecdysones of *Ajuga turkestanica*. V. Khimiya Prirodnikh Soedinenii 710 [in Russian]/Chemistry of Natural Products 595 [in English].

Abstract: Having continued an investigation of the minor ecdysones of *Ajuga turkestanica* (Regel) Brig. (family Labiatae), in addition to the ecdysterone, turkesterone, and ajugalactone isolated previously [1], we have obtained another compound of low polarity with R_f 0.73 [TLC: SiO₂+ 7% of gypsum; chloroform-ethanol (4 : 1) system]. For its isolation, an ethyl acetate fraction of a methanolic extract of the roots of the plant (3 kg) was subjected to repeated column chromatography on alumina and rechromatography on silica gel by the method described previously [2]. The columns were eluted with mixtures of chloroform and ethanol with gradually increasing concentrations of the latter. C₂H₅OH KBr The substance obtained, C₂₉H₄₆O₇, had mp 238-240°C

(decomp.), X_{max} 246 nm (log ϵ 3.90), \sim -max 3420 (OH), 1660 cm⁻¹ (cyclohexenone). Its mass spectrum (MKh-1303, 180°C, 40 eV) contained the peaks of ions with m/e 506 (M⁺), 488 (M H₂O), 427, 426, 363, 362, 345, 327, 301, 300, 143, 125, 107, 97, and 79.

Usmanov B.Z., Rashkes J.V. and Abubakirov N.K. (1978) Phytoecdysones of *Ajuga turkestanica*. VI. 22-acetylcyasterone. *Khimiya Prirodnykh Soedinenii* 215-219 [in Russian]/*Chemistry of Natural Products* 175-178 [in English].

Abstract: The leaves of *Ajuga turkestanica* (Rgl.) Briq. have yielded a new ecdysone — 22-acetyl-cyasterone. The mass spectrum of this new ecdysone and the features of it due to the presence of the acetoxy group at C-22 have been discussed.

Vaidya V.V., Gadgil J.N., Kekare M.B. and Patil S.H. (2015) Simultaneous densitometric quantitation of bioactive components p-methoxybenzoic acid, 3,4-dimethoxycinnamic acid, ecdysterone and stigmasterol in the extract of *Trianthema portulacastrum* Linn. and its marketed polyherbal formulation using HPTLC. *International Journal of Science, Engineering and Technology* 3(3), 780-784.

Abstract: *Trianthema portulacastrum* Linn. belonging to the family aizoaceae is abundantly available weed which has enormous traditional uses due to different bioactive compounds present and thus used in different herbal formulations. Current research paper describes a developed and validated High Performance Thin Layer Chromatographic Method (HPTLC) for simultaneous quantitation of four bioactive compounds i.e. p-methoxybenzoic acid, 3,4-dimethoxycinnamic acid, Ecdysterone and Stigmasterol from whole plant. The analytes were separated on TLC plates coated with silica gel 60 F254 with Chloroform : Ethyl acetate : toluene : Methanol (4 : 2.5 : 2 : 1.5 v/v) as a mobile phase after chamber saturation of 10 min. The development distance was 80mm. Sterols were derivatized using 1% Anisaldehyde-Sulphuric acid reagent. Detection and quantitation were performed by densitometry using deuterium lamp before derivatization at 254nm and after derivatization at 580nm using tungsten. The validated method was applied for quantitation of the four bioactive components in a marketed formulation containing plant extract. A precise, accurate and robust HPTLC method is developed for simultaneous quantitation of four bioactive components which can be used as a quality control tool in standardization of raw materials and marketed herbal formulations.

Vaisar T. and Piš J. (1993) Cyclic boronates in the mass spectrometry of ecdysteroids. *Rapid Communication in Mass Spectrometry* 7, 46-52.

Abstract: For the structural elucidation of ecdysteroids a reaction with phenylboronic acid has been employed. This reaction takes place exclusively on the C₂₀,C₂₂ diol moiety, thus facilitating fast and easy detection of this moiety in the molecule of ecdysteroid. This derivatization also redirects fragmentation to C₁₇/C₂₀ bond cleavage with charge retention on both fragments enabling assessment of the structure both of the steroid nucleus and of the side-chain. Possible reaction of phenylboronic acid with the diol moiety of the glycosyl group of the glycoside of ecdysteroid, producing di-adduct, is demonstrated; participation of the 5 β -hydroxy group of 5 β ,20-dihydroxyecdysone in the formation of such a di-adduct is discussed; and the role of the enol form of the 6-carbonyl group is proved.

Vaněk T., Macek T., Vaisar T and Breznovits A. (1990) Production of ecdysteroids by plant cell culture of *Pteridium aquilinum*. *Biotechnology Letters* 12(10), 727-730.

Summary: Cells of the fern *P. aquilinum*, both callus and suspension cultures, are able to produce ecdysterone, 5- β -OH-ecdysterone, ecdysone, ponasterone and further five unidentified ecdysteroids. Under the cultivation conditions there appears to be an overproduction of ponasterone and total ecdysteroids, in comparison to the original plant.

Ványolós A. (2012) Investigation of plants containing unusual ecdysteroids with ring in their side-chain. Ph.D. Thesis, Department of Pharmacognosy, University of Szeged, Szeged, Hungary, pp.54.

Ványolós A., Simon A., Tóth G., Polgár L., Kele Z., Ilku A., Mátyus P., and Báthori M. (2009) C-29 Ecdysteroids from *Ajuga reptans* var. *reptans*. *Journal of Natural Products* 72 929–932.

Abstract: Investigation of the ecdysteroid constituents of the herb *Ajuga reptans* var. *reptans* resulted in the isolation of three new ecdysteroids, named reptanslactone A (2), reptanslactone B (3), and sendreisterone (5), and the known 24-dehydroprecyasterone (1) and breviflorasterone (4). The structures of compounds 1–5 were determined by spectroscopic methods including one- and two-dimensional NMR measurements.

Ványolós A., Béni Z., Dékány M., Simon A. and Báthori M. (2012) Novel ecdysteroids from *Serratula wolffii*. *The Scientific World Journal* 2012 Article ID 651275, 5 pages.

Abstract: Two new and one known ecdysteroids were identified in the methanolic extract of the roots of *Serratula wolffii*. The new compounds isolated were ponasterone A-22-apioside (1) and 3-epi-shidasterone (3), together with

the known 3-epi-22-deoxy-20-hydroxyecdysone (2). The structures of compounds 1-3 were determined by extensive spectroscopic techniques, including one- and two-dimensional NMR methods.

Vardanega R., Carvalho P.I.N., Santos D.T. and Meireles M.A.A. (2017) Obtaining prebiotic carbohydrates and beta-ecdysone from Brazilian ginseng by subcritical water extraction. *Inovative Food Science and Emerging Technologies* doi:10.1016/j.ifset.2017.05.007.

Abstract: Subcritical water extraction (SWE) is one of the most promising modern extraction techniques for isolation of bioactive compounds from plant materials. Fructooligosaccharides (FOS) and beta-ecdysone were obtained from Brazilian ginseng roots (BGR) and aerial parts (BGA) by SWE aiming to use the entire plant in a biorefinery approach. The BGR extracts showed a FOS content of up to 8.8 g/100 g of extract and a beta-ecdysone content of up to 0.7 g/100 g of extract. BGA extracts showed a beta-ecdysone content of 0.3 g/100 g of extract. An economic evaluation showed that the manufacturing of BGR for beta-ecdysone production had a good profitability with a short payback time of 1.43 years, while BGA could be used as fuel to supply the energetic requirements of the process and then, enable the utilization of the entire plant.

Vardanega R., Muzio A.F.V., Silva E.K., Prata A.S. and Meireles M.A.A. (2019) Obtaining functional powder tea from Brazilian ginseng roots: effects of freeze and spray drying processes on chemical and nutritional quality, morphological and redispersion properties. *Food Research International* **116**, 932-941.

Abstract: In this work, the aqueous extract obtained from Brazilian ginseng (*Pfaffia glomerata*) roots (BGR), rich in beta-ecdysone and fructooligosaccharides (FOS), was powdered by spray drying and freeze drying techniques aiming to obtain a novel functional food product. The effects of these drying techniques on the chemical and nutritional quality, morphological and redispersion properties of the BGR powders were evaluated. The BGR powders obtained by both spray drying and freeze drying techniques maintained their beta-ecdysone and FOS contents after drying, demonstrating the stability of these functional compounds. It was found that the wettability of the powders obtained by different treatments was affected by the drying technique because freeze-dried particles reached the lower values (66 ± 5 s) while spray-dried particles showed a greater time for dispersion into water (150 ± 25 s). This behavior was mainly associated with differences between powder morphological properties since the freeze-dried particles presented a more porous structure, resulting in a greater water diffusivity into microstructure during the redispersion process. Drying process did not affect the storage stability of powders because the glass transition temperature (T_g) for both samples was approximately 160 °C at a relative humidity of 56%. Thus, both BGR powders presented adequate redispersion properties to constitute a new functional tea or even to be used as a functional ingredient in food products.

Varga E., Szendrei K., Hajdu Z., Hornok L. and Csáki G. (1986) Study of the compounds contained in hungarian-grown *Leuzea carthamoides* D.C. (Asteraceae), with special regard to the ecdysteroids. *Herba Hungarica* **25**(1), 115-133.

Vasconcelos J.M., Saldanha C.W., Dias L.L.C., Maldaner J., Rego M.M., Silva L.C. and W.G. (2014) *In vitro* propagation of Brazilian ginseng [*Pfaffia glomerata* (Spreng.) Pedersen] as affected by carbon sources. *In Vitro Cellular & Developmental Biology – Plant* **50**, 746-751.

Abstract: This study aimed to establish a protocol for *in vitro* propagation of two accessions (Ac) of *Pfaffia glomerata* (Ac 4 and Ac 13) and to evaluate the effect of different carbon sources on the production of 20-hydroxyecdysone (20E) in leaves and roots. For the assessment of axillary shoot proliferation *in vitro*, nodal segments were inoculated onto Murashige and Skoog (MS) medium supplemented with 2.22 μ M 6-benzyladenine and 2.68 μ M alpha-naphthaleneacetic acid and carbon sources (glucose or sucrose) at varying concentrations (0.1, 0.2, or 0.3 M). To assess the *in vitro* production of 20E, nodal segments were inoculated into MagentaA (R) containers containing MS medium with different carbon sources (glucose, sucrose, or glucose + sucrose at 0.1 or 0.2 M) and placed in plastic bags with bacterial filters. Both experiments were composed of five repetitions for each treatment and analyzed after 30 d of culture. Multiple shoot formations were genotype-dependent when segments were cultivated on a medium supplemented with glucose or sucrose at 0.1 M, yielding 35 and 43 shoots per explant for Ac 4 and 4.4 and 2.8 shoots per explant for Ac 13, respectively. For the 20E content, significant effects were also observed among accessions and carbon sources. Ac 13 had the highest average 20E levels for both roots and leaves. Under the experimental conditions, Ac 4 had more favorable characteristics for large-scale multiplication than Ac 13, and glucose at 0.2 M was the best carbon source for the cultivation of *Pfaffia*, both for producing multiple shoots and for *in vitro* 20E production. This is the first report using a combination of auxin and cytokinin to enable effective *Pfaffia* *in vitro* axillary shoot proliferation from nodal explants

Vereskovskii V.V., Chekalinskaya I.I. and Pashina G.V. (1983) The dynamics of ecdysterone content in different species of *Rhaponticum* Ludw. *Rastitelny Resursy* **19**, 60-65 [in Russian].

Vokáč K., Buděšinsky M. and Harmatha J. (1999) Minor ecdysteroids from *Leuzea carthamoides*. *Chemický Listy, Symposia* 93, S60.

Vokáč K., Buděšinsky M. and Harmatha J. (2002) Minor ecdysteroid components of *Leuzea carthamoides*. *Collection of Czechoslovak Chemical Communications* 67, 124-139.

Abstract: Fourteen minor ecdysteroid components were isolated and identified from the roots of *Leuzea carthamoides* (Willd.) DC. Two of them are new phytoecdysteroids: leuzeasterone (1) a six-member side-chain lactone, and (24Z)-29-hydroxy-24(28)-dehydromakisterone C (4) a structurally related sitostane type analogue and assumed biogenetic precursor of 1. The next one, 5 α -20-hydroxyecdysone (6), is a rare A/B-ring trans-annelated epimer of the most common phytoecdysteroid 20-hydroxyecdysone. Further compounds: makisterone C (3), 3-epi-20-hydroxyecdysone (5), integristerone A (7), integristerone B (8), 22-oxo-20-hydroxyecdysone (10), taxisterone (11), rubrosterone (12), dihydrorubrosterone (13) and poststerone (14), are new constituents of *L. carthamoides*, though already reported as compounds isolated from other natural sources. Two earlier reported minor *Leuzea* ecdysteroids: the five-membered side-chain lactone carthamosterone (2) and the 11-hydroxy-substituted analogue isovitexirone (9), are also included because they are now better characterised. Certain previously described *Leuzea* ecdysteroids were not found in our material, which may indicate geographic, seasonal or cultivar variations

Volodin V.V., Mishurov V.P., Kolegova N.A., Tiukavin Y.A., Portiagina N.V. and Postnikov B.A. (1993) Ecdysteroids of plants of the Family Asteraceae. *Scientific Reports of the Komi Science Centre*, Issue 319, 20 pp.

Volodin V.V., Alexeeva L.I., Kolegova N.A., Sarker S.D., Šik V., Lafont R. and Dinan L. (1998a) Further ecdysteroids from *Serratula coronata*. *Biochemical Systematics and Ecology* 26(4), 459-461.

Volodin V.V., Luksha V.G., Dinan L., Punegov V.V., Alekseeva L.I., Kolegova N.A., Tyukavin Y.A. and Rebrov A.I. (1998b) Inokosterone and makisterone A from *Serratula coronata*. *Russian Journal of Plant Physiology* 45(3), 322-325 [in English]/*Fiziologiya Rastenii* 45(3), 378-381 [in Russian].

Abstract: The composition of the ecdysteroids of *Serratula coronata* L. from northeastern Russia was studied. This plant species was shown to contain the well-known ecdysteroids 20-hydroxyecdysone and ecdysone, as well as inokosterone and makisterone A, which were found in this plant species for the first time.

Volodin V., Chadin I., Whiting P., Dinan L. (2002) Screening plants of European North-East Russia for ecdysteroids. *Biochemical Systematics and Ecology* 30, 525-578.

Abstract: A strategy for screening plants for ecdysteroid content based on the 'positive tribe' principle is developed and applied, for the first time, to screen the flora of European North-East Russia to identify species which accumulate ecdysteroids; 700 samples representing 411 species from 380 genera of 82 families were investigated. It is established that species with moderate to high ecdysteroid content (detectable with the *Drosophila melanogaster* B II cell bioassay) are not numerous (4% of all screened species). They are found in 14 families of different kinship level. Within families, ecdysteroid-containing plants form groups of closely cognate species (within certain tribes and/or genera); most ecdysteroid-containing species in this study were present in the tribe Cardueae (within the Asteraceae) and in the tribe Lychnideae (within the Caryophyllaceae). Radioimmunoassay, using an ecdysteroid-specific antiserum, allowed us to detect trace amounts of phytoecdysteroids (0.1–0.5 μ g ecdysone equivalents/g plant matter) below the threshold detectable by the insect ecdysteroid receptor-based bioassay. It was found that such trace amounts of ecdysteroids are typical of many of the other plant species tested. We propose that a positive response in the bioassay is an appropriate criterion for detecting species with ecdysteroid content sufficient for protecting the plant against non-adapted phytophagous insects. Analysis of the geographical distribution of ecdysteroid-containing species (as detected by the bioassay) reveals that many of them belong to the southern or polyzonal latitudinal groups. This supports the impact of ecological–geographical factors upon ecdysteroid distribution in plants.

Volodin V., Dinan L., Volodina S., Tkachenko K., Shi Ley, Kanev V. and Gorovoy P. (2006) Ecdysteroids in world flora. Ferns: Urals, Far East, China. *Vestnik of the Institute of Biology, Komi Science Centre, Ural Division of Russian Academy of Sciences* 99(1), 2-7 [in Russian].

Volodin V.V., Loan V.T., Volodina S.O. and Kuznetsov A.N. (2018) Ecdysteroid-containing plants of the National Park Cuc Phuong (Northern Vietnam) *Reports of the Komi Research Centre* 35(3), 46-53 [in Russian, with an English abstract].

Abstract: On the principles of chemosystematics, screening of plants of the National Park Cuc Phuong (Northern Vietnam) for the content of ecdysteroids was carried out. A total of 23 ecdysteroid-containing species in the families

of Acanthaceae, Amaranthaceae, Anacardiaceae, Aquifoliaceae, Capparaceae, Convolvulaceae, Malvaceae, Verbenaceae were identified, in which the detection of ecdysteroids was most likely. Among them there are species of medicinal and nutritional value in the traditional medicine and cuisine of Vietnam. The most interesting for further research are plants of the genera *Vitex*, *Sida*, plant species *Achyranthes bidentata* and *Cyathula prostrata*, used in folk and scientific medicine in Vietnam and other countries of Southeast Asia, as well as *Dracontomelon duperreanum* and *Averrhoa carambola*, which fruits are edible, and *Ilex purpurea*, the leaves of which are used for tea making. The bark of *Vitex quinata* tree, the leaves and young shoots of *Sida acuta*, *Sida rhombifolia*, *Cyathula prostrata* and *Achyranthes bidentata* are recommended as raw materials for the pharmaceutical industry.

Vorob'eva A.N., Rybin V.G., Zarembo E.V., Boltenev E.V. and Verbitskii G.A. (2004) Phytoecdysteroids from *Serratula komarovii*. *Chemistry of Natural Compounds* 40(5), 492-495 [in English]/*Khimiya Prirodnikh Soedinenii* (5), 404-406 [in Russian].

Abstract: Integristerone A and 2-deoxy-20-hydroxyecdysone were observed for the first time in the aerial and subterranean organs of the eastern Asian plant *Serratula komarovii* Iljin. a-Ecdysone was not found in the plants. The dynamics of phytoecdysteroid content (integristerone A, 20-hydroxyecdysone, and 2-deoxy-20-hydroxyecdysone) in the vegetative and generative organs of this species were investigated.

Vorob'eva A.N., Rybin V.G., Zarembo E.V. and Boltenev E.V. (2005) Phytoecdysteroids from *Serratula centauroides*. *Chemistry of Natural Compounds* 41(1), 105-106 [in English]/*Khimiya Prirodnikh Soedinenii* (1), 85-86 [in Russian].

No Abstract available

Vorob'eva A.N., Rybin V.G., Zarembo E.V. and Boltenev E.V. (2006) Phytoecdysteroids from *Stemmacantha uniflora*. *Chemistry of Natural Compounds* 42(6), 742-744 [in English]/*Khimiya Prirodnikh Soedinenii* (6), 604-605 [in Russian].

No Abstract available

Wang B., Zhong H., Cao J., Fang P. and Li G. (2011) Chemical constituents of *Premna serratifolia*. *Zhongcaoyao* 42(6), 1072-1074 [in Chinese].

Abstract: Chem. constituents of *Premna serratifolia* were researched. A variety of chromatog. was applied to isolation and purification, and phys. and chem. properties and spectral methods were used for structure identification. Nine compounds were isolated from ethanol extract of *Premna serratifolia*. They were picrocin D, β -ecdysterone, genkwanin-5-O- β -D-glucoside, quercetin, 2-hexylidene-3-methylsuccinic acid, kaempferol-3-O- β -D-galactopyranoside, 5,7,3'-trihydroxy-4'-methoxy-flavone, 20,22-acetonides of inokosterone (8), 10-O-(E)-p-coumaroylcatalpol (9). Compounds 1-9 were isolated from *Premna serratifolia* for the first time.

Wang G-l., Hou S-s., Mi L-s., Wang J-f. And Yuan Y-m. (1984) Studies on polygodine B of *Murdannia triquetra* (Wall) Bruckn. *Acta Botanica Sinica* 26(5), 554-557 [Article in Chinese].

Wang H., Tan J., Shang X., Zheng X., Liu X., Wang J., Hou X. and Du Y. (2019) Porous organic cage incorporated monoliths for solid-phase extraction coupled with liquid chromatography-mass spectrometry for identification of ecdysteroids from *Chenopodium quinoa* Willd. *Journal of Chromatography A* 1583, 55-62 (<https://doi.org/10.1016/j.chroma.2018.11.019>).

Abstract: Here, a porous organic cage (POC)-incorporated polymeric monolith was fabricated in a syringe through the introduction of the POC into poly(ethylene glycol dimethacrylate) monolith in a one-step traditional free-radical polymerization process. The resulting monolithic phases were characterized by scanning electron microscopy (SEM), transmission electron microscopy (TEM), Fourier-transform infrared spectroscopy (FT-IR), powder X-ray diffraction (PXRD), nitrogen adsorption/desorption experiments and thermogravimetric analysis (TGA), which confirmed the successful incorporation of the POC in the monolithic matrix. The functionality of the POC-incorporated poly(EDMA) monolith facilitated for the solid phase extraction (SPE) of 20-hydroxyecdysone (an ecdysteroid) from *Chenopodium quinoa* Willd. extract coupled with UPLC-QqQ-MS/MS, exhibiting satisfactory accuracy (93-106%), precision (< 6.5%) and reusability. In addition, UPLC-Q-Exactive-Orbitrap-MS/MS analysis of the quinoa sample after SPE by POC-incorporated monolith provided the identification of 20-hydroxyecdysone and three other ecdysteroids. These results demonstrate the potential of POC-incorporated monoliths for the SPE of ecdysteroids from complex plant systems.

Wang H-m., Chen W., Liu D-j., Hou X-h. and Liang N. (2012a) Determination of ecdysterone in Radix *Achyranthes bidentatae* and Juejin Granula by HPLC. *Chinese Journal of The Inuate Traditional Medical Formulae* (9), 122-125 [in Chinese].

Abstract: Objective: To establish an efficient method of liquid chromatography to determine the use of β -moulting steroids in ox-knee plants and moulting particles. Methods: The β -moulting ketone in ox-knee and dejin particles was extracted by reflux and ultrasonic method respectively, and the extraction method was optimized, the content of β -moulting steroid was determined by HPLC, and Kromasil C18 column (4.6 mm \times 250 mm, 5 μ m) column, acetonitrile-water (17:83) was used as mobile phase. Speed 1.0 mL min⁻¹, detection wavelength 242 nm, column temperature 30 degrees C. Results: The best extraction condition for ox-knee is to add 15 times the amount of 70% ethanol reflux extraction 2 times, each 1h, the best extraction condition for the decisive particles is water as a solvent ultrasound extraction of 30 min; L⁻¹, the regression equation is Y=24 867X-1563 (r=0.9995), the average recovery rate of ox-knee and decisive particles is 98.5% and 98.4%, RSD is 1.6%, 1.4% (n=9).

Wang Q-h., Yang L., Jiang H., Yang B-y. and Kuang H-x. (2012b) Isolation and identification of chemical constituents of phytoecdysteroids from the root of *Achyranthes bidentata* Bl. Acta Chinese Medicine and Pharmacology (1), 69-71 [in Chinese].

Abstract: In order to clarify the steroid chemical composition of ox-knee that promotes the proliferation of osteoblasts, five ketone compounds were isolated from the 50% ethanol elution components of ox-knee residue by means of silica column chromatography, ODS reverse-phase column chromatography, and preparation of high-performance liquid chromatography. The chemical structures by NMR and MS were determined to be β -moulting ketones (β -ecdysterone) (1), 25S-ox-knee ketone (25S-inokosterone) (2) and 25R-ox-knee ketone (25R-inokosterone).

Wang J-L., Ruan D-C., Cheng Z-Y. and Yang C-R. (1996) The dynamic variations of 20-hydroxyecdysone in *Cyanotis arachnoidea*. Acta Botanica Yunnanica 18(4), 459-464.

Wang J-q., Zhao M-j., Lin L., Chai Y-l. and Zeng S. (2005) Accelerated solvent extraction of ecdysterone from *Achyranthes bidentata*. Chinese Traditional and Herbal Drugs (12), 1797-1800 [in Chinese].

Abstract: Objective: To extract moulting steroids from cattle knees by accelerating solvent extraction to explore the feasibility of their application in the quality control of Chinese medicine. Methods: With moulting steroids as the index component and RP-HPLC as the detection method, a single-factor investigation method was used to investigate the factors affecting the accelerated solvent extraction of moulting steroids in cattle knees. Results: The optimal conditions for the extraction of moulting steroid accelerated solvent in ox-knee were selected: waterless methanol as solvent, the particle size of the herbs was 0.3 to 0.45 mm, the extraction temperature was 100 degrees C, the extraction pressure was 10.34 MPa, the extraction time was 6 min, and the extraction was 1 time. Conclusion: Accelerated solvent extraction technology can extract moulting steroids in ox-knee quickly and efficiently.

Wang P., Li S., Ownby S., Zhang Z., Yuan W., Zhang W. and Beasley R.S. (2009) Ecdysteroids and a sucrose phenylpropanoid ester from *Froelichia floridana*. Phytochemistry 70(3) 430-436.

Abstract: Phytoecdysteroid glycosides (1-5) and a phenylpropanoid ester of sucrose (6) were isolated from the whole plant of *Froelichia floridana*, along with eight known compounds including three ecdysteroids (7-9), four flavonoids (10-13), and one phenolic compound (14). Structures were determined using a combination of spectroscopic techniques. Compounds 1, 2 and 6-14 were tested in vitro for their activity against human DNA topoisomerase I. Compound 13 (diosmetin) showed marginal inhibition against topoisomerase I with IC(50) of 130 microM in conjunction with low intercalation ability.

Wang Q., Guo Z. and Zou K. (2012) Studies of chemical constituents from rhizome of *Reineckia carnea*. Sanxia Daxue Xuebao, Ziran Kexueban (Journal of China Three Gorges University, Natural Sciences) 34(1), 82-84 [in Chinese].

Abstract: To study the chem. constituents from rhizome of *Reineckia carnea* and search for bioactive compounds with potential antitumor activity. The compounds were isolated by RP-C₁₈ column chromatog. and preparative HPLC. The structures of the compounds were identified on the basis of the spectroscopic anal. and chem. evidence. Three steroids (1-3) and two flavonoids (4-5) were obtained from the *Reineckia carnea* rhizomes. Their structures were determined as Nicotianoside B(1), Daucosterol(2), β -ecdysone (2 β , 3 β , 14 α , 20R, 22R, 25-hexahydroxy-5 β -cholest-7-en-6-one) (3), quercetin(4) and rutin(5), resp. Compound 3 was isolated from the *Reineckia carnea* for the first time.

Wang Q-H., Yang L., Jiang H., Wang Z-B., Yang B-Y. and Kuang H-X. (2011) Three new phytoecdysteroids containing a furan ring from the roots of *Achyranthes bidentata* Bl. Molecules 16 5989-5997 (doi:10.3390/molecules16075989).

Abstract: Three new phytoecdysteroid compounds, named niuxixinsterone A (1), B (2) and C (3) with acetal functions in the side-chain were isolated from *Achyranthes bidentata* Bl. The structures were established as (20R, 22R, 24S)-20-O, 22-O-(5'-hydroxymethyl)-furfurylidene-2 β , 3 β , 14 α , 25-tetrahydroxy-5 β -ergost-7-en-6-one (1),

(20R,22R)-20-O,22-O-(5'-hydroxymethyl)-furfurylidene-2 β ,3 β ,25-trihydroxy-14 β -methyl-18-nor-5 β -cholesta-7,12-dien-6-one (2) and (20R,22R,25R)-20-O,22-O-(5'-hydroxymethyl)-furfurylidene-2 β , 3 β ,5 β ,14 α ,26-pentahydroxycholest-7-en-6-one (3) by means of spectroscopic evidence.

Wang Q.J., Zheng L.P., Sima Y.H., Yuan H.Y. and Wang J.W. (2013) Methyl jasmonate stimulates 20-hydroxyecdysone production in cell suspension cultures of *Achyranthes bidentata*. *Plant Omics Journal* 6(2), 116-120.

Abstract: 20-Hydroxyecdysone (20E), one of major phytoecdysteroids (i.e. analogues of insect steroid hormones) in plants, has important agrochemical, medicinal and pharmaceutical uses. In order to develop a sustainable source of 20E, cell suspension cultures were established from shoot cultures of *Achyranthes bidentata*. When cultivated in Murashige and Skoog medium supplemented with 1.5 mg/l 1-naphthylacetic acid (NAA) and 1.5 mg/l 6-benzyladenine (6-BA), *A. bidentata* cells in suspension culture grew rapidly, yielding 20E (5.4 mg/l) after 24 days. The increase of 20E was dependent on the growth stage of cell cultures as well as on the dose of methyl jasmonate (MeJA) applied. When cells of 18-day-old cultures were exposed to 0.6 mM MeJA for 6 days, it was found that total (intracellular and extracellular) 20E production reached the maximum yield 75 mg/l, a 2.6-fold increase over the control. This is the first report of 20E production in cell suspension cultures of *A. bidentata*.

Wang Q.J., Zheng L.P., Zhao P.F., Zhao Y.L. and Wang J.W. (2014) Cloning and characterization of an elicitor-responsive gene encoding 3-hydroxy-3-methylglutaryl coenzyme A reductase involved in 20-hydroxyecdysone production in cell cultures of *Cyanotis arachnoidea*. *Plant Physiology and Biochemistry* 84, 1-9.

Abstract: *Cyanotis arachnoidea* contains a rich source of bioactive phytoecdysteroids (i.e. analogues of insect steroid hormones). 3-Hydroxy-3-methylglutaryl coenzyme A reductase (HMGR) supplies mevalonate for the synthesis of many secondary metabolites including 20-hydroxyecdysone (20E), one of metabolism-enhancing phytoecdysteroids. In this study, in order to develop a sustainable source of 20E, cell suspension cultures were established from shoot cultures of *C. arachnoidea*, and a full length cDNA encoding HMGR (designated as CaHMGR) was cloned and characterized. The cDNA contained 2037 nucleotides with a complete open reading frame (ORF) of 1800 nucleotides, which was predicted to encode a peptide of 599 amino acids. Expression analysis by real-time PCR revealed that CaHMGR mRNA was abundant in *C. arachnoidea* stems, roots and leaves. When cultivated in Murashige & Skoog medium supplemented with 0.2 mg L⁻¹ 1-naphthylacetic acid (NAA) and 3.0 mg L⁻¹ 6-benzyladenine (6-BA), *C. arachnoidea* cells in suspension culture grew rapidly, yielding 20E (124.14 μ g L⁻¹) after 12 days. The content of 20E in cell cultures elicited by 0.2 mM methyl jasmonate (MeJA), 100 mg L⁻¹ yeast elicitor (YE) or 25 μ M AgNO₃ was increased 8-, 2-, and 6-fold over the control, respectively. Quantitative real-time PCR analysis showed that CaHMGR was expressed at a higher level under the treatment of MeJA or Ag⁽⁺⁾ elicitor. Our results suggested that 20E accumulation may be the result of the expression up-regulation of CaHMGR involved in the biosynthesis under the treatment of various elicitors.

Wang S., Wu Y., Li L., Zhang J. and Wu J. (2011) A study on chemical constituents of *Tinospora sagittata*. *Guiyang Yixueyuan Xuebao* 36(1), 9-10, 14 [in Chinese].

Abstract: The objective of this study is to investigate the chem. constituents of *Tinospora sagittata*. Various chromatog. techniques including silica gel, Sephadex LH-20, and RP-18 trabeulum were used to isolate the constituents of *T. sagittata*, and their structures were identified by spectral and chem. methods. Five compounds were isolated from *T. sagittata*, and their structures were identified as 20 β -hydroxy-ecdysone (I), tinoside (II), columbamine (III), acutumine (IV), and jatrorrhizine (V). This is the first report that compound V being isolated from *T. sagittata*.

Wang S.F., Dai J.Q., Chen X.G. and Hu Z. (2002) Identification and determination of ecdysones and flavonoids in *Serratula strangulata* by micellar electrokinetic capillary chromatography. *Planta Medica* 68(11), 1029-1033.

Abstract: The simultaneous determination of 20-hydroxyecdysone (1), 25-deoxy-11,20-dihydroxyecdysone (2), kaempferol 4',7-dimethyl ether (3) and biochanin A (4) in the fruit, leaf and root of *Serratula strangulata* Iljin has been investigated by micellar electrokinetic capillary chromatography for the first time. With an electrolyte containing 15 mmol/L borate, 60 mmol/L SDS, 20 % (v/v) methanol, at pH 9.08 and 20 kV applied voltage, the four analytes were completely separated within 22.7 min. The effects of concentration of borate, electrolyte pH, the concentrations of SDS and organic modifier and the applied voltage on electrophoretic behavior and separation were studied. Regression equations revealed linear relationships (correlation coefficients: 0.9986 - 0.9997) between the peak areas of the analytes and the corresponding concentrations. The levels of analytes in the different parts of *S. strangulata* were easily determined with recoveries ranging from 95.3 % to 104.0 %.

Wang T., Cui S.Y., Suo Y.R. and Lu R.H. (2004) Studies on water-soluble chemical constituents in root of *Achyranthes bidentata*. *Zhongguo Zhong Yao Za Zhi* 29(7), 649-652 [in Chinese].

Abstract:

Objective: To study the water-soluble chemical constituents in root of *Achyranthes bidentata*.

Method: The chemical constituents were isolated by silica gel column chromatography and the structures were elucidated by the NMR spectra and physico-chemical properties.

Result: Seven compounds were obtained and identified as n-butyl-beta-D-fructopyranoside (I), oleanoic acid (II), 3-O-[beta-D-glucopyranosyl], oleanoic acid 28-O-beta-D-glucopyranoside (III), allantoin (IV), 20-hydroxy ecdysone (V), glutamic acid (VI), 3-O-[beta-D-glucopyranosyl], oleanoic acid 28-O-beta-D-glucopyranosyl ester (VII).

Conclusion: Compounds III-VII were obtained from this plant for the first time.

Wang X., Ding X., Wu K. and Lan J. (2001) Studies on chemical constituents of *Rhaponticum uniflorum*. *Zhongcaoyao* 32(7), 590 [in Chinese].

Abstract: The chem. constituents of *Rhaponticum uniflorum* were studied. 6 Compounds were isolated by solvent extraction and purified by chromatog. on silica gel column. Their structures were identified by UV, EIMS, ¹HNMR, and physicochem. properties. The 6 compounds were identified as β -sitosterol, hexadecanoic acid, tetracosanoic acid, ecdysterone, rhaponsterone, and daucosterol.

Wang X-J., Zhang Q., Peng Y-R., Li L., Qu J., Liu Y-B., Xu S., Ma S-G., Li Y., Zou Z-M., Wang R-B. and Yu S-S. (2018) Two azafluoranthene alkaloids and a phytoecdysone from the stems of *Cyclea barbata*. *Journal of Asian Natural Products Research* 21(3), 217-226 (doi: 10.1080/10286020.2018.1564137)

Abstract: Two new azafluoranthene alkaloids (**1** and **2**), and a new phytoecdysone (**3**), were isolated from the stems of *Cyclea barbata* Miers, together with six known compounds (**4-9**). Their structures were elucidated by spectroscopic data analysis and comparison with published data. This is the first report of azafluoranthene alkaloids (**1** and **2**) and phytoecdysones (**3**, **8**, and **9**) from *Cyclea* genus. In *in vitro* bioassay, four isolates (**3**, **5**, **6**, and **9**) showed moderate hepatoprotective activity against *N*-acetyl-*p*-aminophenol (APAP)-induced toxicity in HepG2 cells.

Wang X-m., Chi D-f. and Yu J. (2018) The effect of jasmonic acid methylester on cell growth and β -ecdysterone accumulation in *Ajuga lobata*. *Acta Prataculturae Sinica* 27(9), 95-109 [in Chinese].

Abstract: The suspended cells contain higher p-moulting steroids, and in order to further improve their p-moulting steroid content, a series of experimental studies were conducted by adding methyl jasmine (MeJA). With 4 to 10 generations of rib-grass suspended cells as the test material, the effect of MeJA on cell growth, β -moulting ketone accumulation was determined by measuring the amount of cell growth, and the content of p-moulting ketone was detected using high-efficiency liquid phase. The results showed that the growth curve of the suspended cells and the accumulation curve of β -moulting steroids conformed to the Logics model. The addition of different concentrations of MeJA in the initial (day 4) or medium term (day 7) of rapid cell growth has a relatively small effect on cell growth, with small peaks in the growth curve, which occur on the 5th and 3rd days after treatment, with dry matter accumulation reaching 0.60 and 0.62 g, respectively. The peak of cell rapid growth added MeJA, cell growth curve showed a downward trend, cell fresh weight and dry weight significantly lower than CK ($P < 0.05$). After the initial or medium period of rapid cell growth, cell fresh weight and β -moulting ketone accumulation showed a significant correlation. Add 10 to 50 mol in the initial or medium period of rapid cell growth. After L-1 MeJA, the cell weight was significantly increased compared to CK, with the addition of 50 μ mol. L-1 MeJA has the best cell weight, up to 35.90 g, significantly higher than other treatments ($P < 0.05$), while under the same conditions β -ecdysterone shows a small increase in accumulation, with a maximum of 0.5095 mg.g-1. Add 100 to 200 mol. L-1 MeJA inhibits cell growth, adding 200 μ mol. L-1 MeJA cell fresh weight decreased significantly compared to CK, with a maximum inhibition rate of 38.88%. L-1 MeJA post-p moulting ketone accumulation showed a significant increase, up to 3.5315 mg.g-1, 14.44 times ($P < 0.01$) of CK during the same period. After L-1 MeJA, cell weight decreased compared to CK, indicating that adding these concentrations of MeJA at this time inhibits cell growth, with a maximum inhibition rate of 31.01%. After L-1 MeJA, the accumulation of β -ecdysterone can surge in a short period of time, and the accumulation of β -mol ketone can reach a maximum of 1.4136 mg.g-1, which is 5.06 times that of CK ($P < 0.01$). L-1 MeJA accumulates less. Add 100 μ mol in the middle of a cell's rapid growth. Cell stimulation was small under L-1 MeJA conditions, with the highest accumulation of β -moulting ketones reaching 3.5315 mg.g-1

Wang Y., Zhang Z., Wang X., Xu X. and Zhang H. (2003) Study on the fingerprints of *Achyranthes bidentata* Bl. by HPLC/UV/MS. *Zhong Yao Cai* 26(11), 787-789 [in Chinese].

Abstract:

Objective: To establish the fingerprints of *Achyranthes bidentata* Bl. by HPLC/UV/MS.

Method: Separation was performed on Aglient Zorbax SB-C18 column. Gradient elute was performed by the mobile phase consisting of methanol and 0.1% formic acid-water with the flow rate of 1.0 ml/min.

Result: Perfect fingerprints were obtained which can be used for the evaluation of *Achyranthes bidentata* Bl. Four common peaks were confirmed in fingerprints. Three compounds were elucidated as 5-hydroxymethyl furaldehyde, ecdysterone, inokosterone by HPLC/MS.

Conclusion: The method can be applied to the quality control of *Achyranthes bidentata* Bl.

Wang Y., Han Q., Li X., Zuo H., Yang J., Xia M., Luo J., Li H. and Yang Y. (2017) *Paris dulongensis* ethanol extract and its application in pharmacy. Chinese Patent CN107446014 A 2017-12-08 [in Chinese].

Abstract: The invention relates to *Paris dulongensis* ethanol extracts and its application in pharmacy for treatment of cancer. Paris saponin V, Paris saponin I, β -ecdysone, protodioscin, Parisaponin I and ginsenoside Rg1 are isolated from *Paris dulongensis*.

Wang Y., Liu E. and Li P. (2017) Chemotaxonomic studies of nine *Paris* species from China based on ultra-high performance liquid chromatography tandem mass spectrometry and Fourier transform infrared spectroscopy. *Journal of Pharmaceutical and Biomedical Analysis* **140**, 20-30.

Abstract: *Paris* species, which contain steroid saponins, have been used as herb folk medicines in Asia. In the present study, a comprehensive strategy based on liquid chromatography-tandem mass spectrometry (LC-MS/MS) and Fourier transform infrared (FT-IR) spectroscopy was firstly proposed to evaluate the chemotaxonomic relationships of nine *Paris* species sampled from different geographical regions in China. Principle component analysis (PCA) based on FT-IR data revealed chemical similarities in term of the nine species and geographical regions, indicating the accumulation of metabolites affected by the combination of geographical factors and species. The chemotaxonomic relationships of four species supported the morphological taxonomy and implied ancestry from *P. polyphylla*. After high-efficiency chromatographic separation, ions trap/time-of-flight mass spectrometry (IT-TOFMS) and triple quadrupole mass spectrometry (QQQ-MS) were used to identify unknown metabolites and simultaneously determine six key compounds (polyphyllin I, II, V, VI, VII and gracillin) in *Paris* species, respectively. The tentative identification of 22 steroid saponins was indicative of a common biosynthetic pathway in *Paris* species. Phytoecdysones, gracillin and open-chain steroid saponins were considered as key precursors. According to Pearson's correlation analysis, an insignificant correlation was found between diosgenin-type and pennogenin-type saponins belonging to the same biosynthetic pathways in the current stage. Our results could provide a reasonable foundation for chemotaxonomy or further studies of *Paris* species.

Wang Y.C., Zhao J.J. and Chi D.F. (2018) β -Ecdysterone accumulation and regulation in *Ajuga multiflora* Bunge suspension culture. *3Biotech* **8**(2): 87 (doi: 10.1007/s13205-018-1117-2).

Abstract: *Ajuga multiflora* Bunge cells contain β -ecdysterone (β -EC) that regulates the molting process of insect larvae. In this study, different conditions of culture have been studied to optimize the production of β -EC. *A. multiflora* Bunge growth fitted the curve of logistic equation with one growth cycle of 17 days. The electric conductivity of medium had a negative correlation with not only the weight of dry cell but also the β -EC accumulation, and thus, could be used for monitoring the peak of both cell growth and β -EC accumulation. The pH value of the culture medium varied from 4.67 to 5.84 and reached the maximum at the end of the culture (on the 17th day). The relation of cell growth and nutrient consumption in *A. multiflora* Bunge cell suspension culture was distinctly correlated. Continuous subculture caused a reduction in β -EC synthesis; passages 7–15, the β -EC content declined ($p < 0.05$). At passage 11, the β -EC content was only 42.72% of that at passage 5. Additives such as mevalonic acid (MVA), L-phenylalanine (L-Phe), α -pinene, terpineol, and nitric oxide (NO) in the suspension culture medium, could significantly promote the cell growth and stimulate β -EC accumulation. The optimal concentrations of L-Phe, MVA, terpineol, and α -pinene were 0.2 mmol/l, 10 mg/l, 1 mmol/l and 6 mmol/l, respectively, with the β -EC concentrations as 1.914 ± 0.1948 mg/g ($p < 0.01$), 6.012 ± 0.4252 mg/g ($p < 0.01$), 5.147 ± 0.4819 mg/g ($p < 0.01$), 2.801 ± 0.1253 mg/g ($p < 0.01$), respectively. The optimal concentration of sodium nitroprusside, the provider of NO, was 3 mmol/l with the β -EC concentration 2.87 ± 0.2493 mg/g ($p < 0.01$). The results offer a strategy for massive production of β -EC.

Wang Y-H., Avula B., Jadhav A.N., Smillie T.J. and Khan I.A. (2008) Structural characterization and identification of ecdysteroids from *Sida rhombifolia* L. in positive electrospray ionization by tandem mass spectrometry. *Rapid Communications in Mass Spectrometry* **22**, 2413-2422.

Abstract: Seven ecdysteroids isolated from *Sida rhombifolia* L. were studied by electrospray ionization multi-stage tandem mass spectrometry (ESI-MS(n)) in the positive ion mode using an ion trap analyzer and high-performance liquid chromatography coupled with a diode-array detector (HPLC/DAD). The HPLC experiments were performed by means of a reversed-phase C(18) column and a binary mobile phase system consisting of water (containing 0.05% formic acid) and acetonitrile (containing 0.05% formic acid) under gradient elution conditions. According to mass spectral features and the substitution at C-2, C-20, C-24 and C-25, ecdysteroids in *S. rhombifolia* were classified into three sub-groups. Structural identification of these three sub-groups of ecdysteroids was established by LC/multi-stage ion trap mass spectrometry on-line or off-line. The fragmentation patterns of ecdysteroids yielded ions of successive loss of 1-4 water molecules. Furthermore, ions corresponding to the complete loss of the side chain at C-17 will help to identify the sub-groups of ecdysteroids in addition to containing a hydroxyl moiety at one of the above-mentioned positions. Based on the HPLC retention behavior, the diagnostic UV spectra and the molecular

structural information provided by ESI-MS(n) spectra, a total of nine naturally occurring ecdysteroids were identified, of these two are identified for the first time in *S. rhombifolia*.

Wang Y-S., Yang J-H., Luo S-D., Zhang H-B. and Li L. (2006) New cytotoxic steroid from *Stachyurus himalaicus* var *himalaicus*. *Molecules* **11**, 536-542.

Abstract: A phytochemical study of the ethanolic extract of *Stachyurus himalaicus* var. *himalaicus* was undertaken and as a result a new polyoxygenated steroid, named stachsterol ((20*S*)-20, 25-dihydroxy-4-cholesten-3-one, **1**) and three known ecdysteroids, 20-hydroxyecdysone (**2**), 20-hydroxyecdysone-20, 22-monoacetone (**3**) and polypodine B-20,22-monoacetone (**4**), were isolated. Their structures were elucidated by spectroscopic methods, including UV, NMR, MS and HR-MS. The purified product **1** was found to have *in vitro* cytotoxic activity against human HeLa cell lines with an IC₅₀ value of 2.5 µg/mL. This is the first time that phytoecdysteroids have been found in the genus *Stachyurus*.

Watanabe M., Miyashita T. and Devkota H.P. (2021) Phenolic compounds and ecdysteroids of *Diplazium esculentum* (Retz.) Sw. (Athyriaceae) from Japan and their chemotaxonomic significance. *Biochemical Systematics and Ecology* **94**, article 104211, pp4.

Abstract: Four phenolic compounds, (2*R*)-3-(4'-hydroxyphenyl) lactic acid, *trans*-cinnamic acid (**2**), protocatechuic acid (**3**) and rutin (**4**), and three ecdysteroids, amarasterone A1 (**5**), makisterone C (**6**) and ponasterone A (**7**) were isolated and identified from the young fronds of *Diplazium esculentum* (Retz.) Sw. (Athyriaceae). The structures of these compounds were elucidated on the basis of NMR spectral data and comparison with literature values. Compounds **1**, **2** and **5–7** were isolated for the first time from title plant. Their structure elucidation and chemotaxonomic significance are explained in detail.

Wei Z-h., Wang X-m., Hou C-b., Li X-f. and Chen S-l. (2013) Study on difference of the contents of active ingredient and some elements in differently-processed *Achyranthes bidentata* B1. by HPLC and ICP-OES. *Journal of Zhengzhou College of Animal Husbandry Engineering* (3), 1-3 and 7 [in Chinese].

Abstract: The HPLC and ICP-OES methods were applied to determine the contents of active ingredient and some elements in differently-processed *Achyranthes bidentata* B1. The results showed that the Oleanolic Acid contents of all the differently-processed *Achyranthes bidentata* B1. were higher than unprocessed product, and the highest were alcohol steamed product and alcohol-broiled product, while the ecdysterone contents of all the products were similar. The contents of Zn and Mg in differently-processed *Achyranthes bidentata* B1. were higher than that of unprocessed product except for salt-broiled product of Zn content, while the contents of Cu among all the products were not different. The ratios of the Zn/Cu value were about 2.2.

Werawattanametin K., Podimuang V. and Suksamrarn A. (1986) Ecdysteroids from *Vitex glabrata*. *Journal of Natural Products* **49**, 365-366.

No Abstract.

Wessner M., Champion B., Girault J-P., Kaouadji N., Saidi B. and Lafont R. (1992) Ecdysteroids from *Ajuga iva*. *Phytochemistry* **31**, 3785-3788.

Abstract: *Ajuga iva* contains large amounts of three major ecdysteroids (makisterone A, 20-hydroxyecdysone and cyasterone) together with several minor compounds including 24,28-dehydromakisterone A and two new phytoecdysteroids (22-oxocyasterone and 24,25-dehydroprecyasterone) which have been identified by spectrometric procedures.

Whitehead D.L. and Sellheyer K. (1982) The identification of ecdysterone (20-hydroxyecdysone) in 3 species of molluscs (Gastropoda: Pulmonata). *Experientia* **38**, 1249-1251.

Abstract: Ecdysterone has been identified in the schistosomiasis vector *Biomphalaria glabrata* Say and in *Helix aspersa* Müller as well as in *Lymnaea stagnalis* L. by chromatography, bioassay, radioimmunoassay, derivatization and by mass spectroscopy. Analysis of the food, faeces and hepatopancreas suggest that the sterol is derived from the diet. The probable function of ecdysterone in relation to calcification of the shell is discussed in this paper.

Whiting P., Savchenko T., Sarker S.D., Rees H.H. and Dinan L. (1998) Phytoecdysteroids in the genus *Limonium* (Plumbaginaceae). *Biochemical Systematics and Ecology* **26**, 695-698.

No Abstract.

Wilson I.D. (2000) Multiple hyphenation of liquid chromatography with nuclear magnetic resonance spectroscopy, mass spectrometry and beyond. *Journal of Chromatography A* **892**(1/2), 315-327.

Abstract: The advent of sensitive and reliable HPLC-NMR and HPLC-MS systems has revolutionised the identification of compounds eluting from chromatographic systems. More recently systems have been described

wherein both NMR and MS are used together to provide an immensely powerful means of characterising compounds in chromatographic eluents. Here the construction and application of combined HPLC-NMR-MS systems to the analysis of mixtures of pharmaceuticals, drug metabolites in biological fluids and natural products in plant extracts is reviewed. In addition preliminary work with alternative systems such as HPLC-UV-NMR-FTIR-MS is highlighted and the prospects for such complex systems considered.

Wilson I.D., Lafont R., Kingston R.G. and Porter C.J. (1990) Thin layer chromatography-tandem mass spectrometry directly from the adsorbent: application to phytoecdysteroids of *Silene otites*. *Journal of Planar Chromatography* **3**, 359-361.

Wilson I.D., Morgan E.D., Lafont R. and Wright B. (1998) High-performance liquid chromatography coupled to nuclear magnetic resonance spectroscopy - application to the ecdysteroids of *Silene otites*. *Journal of Chromatography A* **799**(1/2), 333-336.

Summary: HPLC coupled in parallel to nuclear magnetic resonance (NMR) and mass spectrometry (MS) has been used to obtain ¹H NMR and mass spectra of a number of ecdysteroids present in an extract of the plant *Silene otites*. Reversed phase gradient chromatography was performed using a D20-acetonitrile-based solvent system. NMR and mass spectra were obtained for integristerone A, 20-hydroxyecdysone, 2-deoxy-20-hydroxyecdysone and 2-deoxyecdysone to provide structural confirmation using continuous and stopped flow HPLC-NMR. The combined HPLC-NMR-MS system described here provided a more comprehensive analysis of the ecdysteroids present in the extract than HPLC-NMR alone.

Wilson I.D. and Morden W. (1999) Practical applications of HPTLC and HPTLC-MS/MS. *LC.GC International* **12**(2), 72-80.

Wilson I.D., Morgan E.D., Lafont R., Shockor J.P., Lindon J.C., Nicholson J.K. and Wright B. (1999) High performance liquid chromatography coupled to nuclear magnetic resonance spectroscopy and mass spectrometry applied to plant products: identification of ecdysteroids from *Silene otites*. *Chromatographia* **49**(7/8), 374-378.

Abstract: HPLC coupled in parallel to nuclear magnetic resonance (NMR) and mass spectrometry (MS) has been used to obtain ¹H NMR and mass spectra of a number of ecdysteroids present in an extract of the plant *Silene otites*. Reversed phase gradient chromatography was performed using a D20-acetonitrile-based solvent system. NMR and mass spectra were obtained for integristerone A, 20-hydroxyecdysone, 2-deoxy-20-hydroxyecdysone and 2-deoxyecdysone to provide structural confirmation using continuous and stopped flow HPLC-NMR. The combined HPLC-NMR-MS system described here provided a more comprehensive analysis of the ecdysteroids present in the extract than HPLC-NMR alone.

Wong L.Z., Li H.Y., Chang Y.Y., Zhu G.Q., Shong S.X, Li X.H. and Ye J.S. (1979) Identification and physiological tests of phytoecdysones from Chinese flora with the silkworm *Bombyx mori* L. *Acta Entomologica Sinica* **22**, 396-403.

Wu G. and Zhang Z. (2017) Effect of different types of ethanol processing methods on the fingerprint of medicinal *Cyathula* root. *World Chinese Medicine* (6), 1443-1446 [in Chinese].

Abstract: Objective: To compare the fingerprints among Medicinal *Cyathula* root raw products, distilled by solid water and different concentrations of ethanol, so as to study the effects of medicinal *Cyathula* root products processed by different concentration of ethanol. Methods: By using HPLC method, taking the SHIMADZU VP-ODS (4.6 × 250 mm, 5 μm) as the chromatographic column, acetonitrile-water as mobile phase gradient elution, flow-rate 0.8 mL/min, column temperature 25°C, detection wavelength 243 nm, sample volume 15 μL, acquisition for 90 min. Results: Most of the peak area of medicinal *Cyathula* root that processed by ethanol were slightly higher than raw products; raw products were lower than the distilled water products, but not very obviously. Conclusion: In this experiment, medicinal *Cyathula* root processed by ethanol is conducive to the dissolution of most active ingredient, high concentration ethanol processed product is better than the low concentration ethanol in the terms of the active ingredient dissolution.

Wu H., Guo J., Chen S., Liu X., Zhou Y., Zhang X. and Xu X. (2013) Recent developments in qualitative and quantitative analysis of phytochemical constituents and their metabolites using liquid chromatography-mass spectrometry. *Journal of Pharmaceutical and Biomedical Analysis* **72** 267-291.

Abstract: Over the past few years, the applications of liquid chromatography coupled with mass spectrometry (LC-MS) in natural product analysis have been dramatically growing because of the increasingly improved separation and detection capabilities of LC-MS instruments. In particular, novel high-resolution hybrid instruments linked to ultra-high-performance LC and the hyphenations of LC-MS with other separation or analytical techniques greatly aid unequivocal identification and highly sensitive quantification of natural products at trace concentrations in complex

matrices. With the aim of providing an up-to-date overview of LC-MS applications on the analysis of plant-derived compounds, papers published within the latest years (2007-2012) involving qualitative and quantitative analysis of phytochemical constituents and their metabolites are summarized in the present review. After briefly describing the general characteristics of natural products analysis, the most remarkable features of LC-MS and sample preparation techniques, the present paper mainly focuses on screening and characterization of phenols (including flavonoids), alkaloids, terpenoids, steroids, coumarins, lignans, and miscellaneous compounds in respective herbs and biological samples, as well as traditional Chinese medicine (TCM) prescriptions using tandem mass spectrometer. Chemical fingerprinting analysis using LC-MS is also described. Meanwhile, instrumental peculiarities and methodological details are accentuated.

Wu J., Gao L., Shang L., Wang G., Wei N., Chu T., Chen S., Zhang Y., Huang J., Wang J. and Lin R. (2017) Ecdysterones from *Rhaponticum carthamoides* (Willd.) Iljin reduce hippocampal excitotoxic cell loss and upregulate mTor signalling in rats. *Fitoterapia* (doi: 10.1016/j.fitote.2017.03.015).

Abstract: Glutamate-induced excitotoxicity is a key pathological mechanism in many neurological disease states. Ecdysterones derived from *Rhaponticum carthamoides* (Willd.) Iljin (RCI) have been shown to alleviate glutamate-induced neuronal damage; although their mechanism of action is unclear, some data suggest that they enhance signaling in the mechanistic target of rapamycin (mTOR) signaling pathway. This study sought to elucidate the mechanisms underlying ecdysterone-mediated neuroprotection. We used *in silico* target prediction and simulation methods to identify putative ecdysterone binding targets, and to specifically identify those that represent nodes where several neurodegenerative diseases converge. We then used histological analyses in a rat hippocampal excitotoxicity model to test the effectiveness of ecdysterones *in vivo*. We found that RCI-derived ecdysterones should bind to glutamatergic NMDA-type receptors (NMDARs); specifically, *in vivo* modeling showed binding to the GRIN2B subunit of NMDARs, which was found also to be a node of convergence in several neurodegenerative disease pathways. Computerized network construction by using pathway information from the Kyoto Encyclopedia of Genes and Genomes (KEGG) database showed putative links between GRIN2B and mTOR pathway elements including phosphoinositide-3kinase (PI3K), mTOR, and protein kinase C (PKC); these elements are associated with neuronal survival. Brain tissue western blots of ecdysterone-treated rats showed upregulated PI3K, Akt, mTOR, and phosphorylated Akt and mTOR, and down regulated GRIN2B and the apoptotic enzyme cleaved caspase-3. Ecdysterone treatment also prevented glutamate-induced rat hippocampal cell loss. In summary, RCI-derived ecdysterones appear to prevent glutamatergic excitotoxicity by increasing mTOR/Akt/PI3K signaling activity.

Wu J., Kuang Y. and Liu X. (2019) Study on dynamic change of yield and ecdysterone of *Serratulae chinensis* radix at different harvest times. *Chinese Journal of Information on Traditional Chinese Medicine* (10), 81-84 [in Chinese].

Abstract: Objective: To study the yield of wide-lift hemp and its moulting ketone content in different harvesting periods, and to provide the basis for determining the harvesting period during the standardized cultivation of wide-raising hemp. Methods: The root quality was determined by weighing the wide-lifting hemp plants collected in different harvesting periods. HPLC measures the content of moulting steroids in roots, with chromatography columns ZORBAX Eclipse XDB-C18 (250 mm×4.6 mm, 5 μm) and flow phase methanol-water (45.55, V/V), flow rate of 1.0 mL/min, detection wavelength 248 nm, column temperature of 30 degrees C, sample volume of 10 μL. Results The root quality and root of the broad-lifting hemp plant in different harvest periods showed dynamic changes in the content of moulting steroids. Root quality reached its maximum in late October, after which it was basically stable; The moulting ketone content peaked in early November and then decreased slightly. Conclusion: Comprehensive root yield, moulting steroid content and the waiting period of broad -litre hemp plants determined that the optimal harvesting period of broadly raised hemp root was in early and middle November.

Wu J-j., Wang H., Ye W-c. Zuo X-f. and Zhao S-x. (2006) Chemical constituents from *Asparagus filicinus*. *Journal of China Pharmaceutical University* 37(6) 487-490.

Abstract: Aim: To investigate the chemical constituents from *Asparagus filicinus* Ham. ex D. Don. Methods: Column chromatographic techniques were used for the isolation and purification of chemical constituents of the plant and their structures were elucidated by spectroscopic analysis. Results: Analyses showed that five ecdysteroids, β-ecdysterone (1), 25-hydroxydacryhainansterone (2), stachysterone B (3), 5-deoxykaladasterone (4), calonysterone (5), two lignans, syringaresinol-4'-O-β-D-glucopyranoside (6), (+)-nyasol (7), one phenylpropanoid 1-O-feruloylglycerol (8), β-sitosterol (9), and β-daucosterol (10) were isolated from the CHCl₃ elution fraction of this plant, respectively. Conclusion: Compounds 2-10 are isolated from this plant for the first time and compound 2 is obtained from natural source for the first time.

Wu J-J., Cheng K-W., Wang H., Ye W-C., Li E.T.S. and Wang M. (2009) Simultaneous determination of three phytoecdysteroids in the roots of four medicinal plants from the genus *Asparagus* by HPLC. *Phytochemical Analysis* 20 58–63.

Abstract:

Introduction: The genus *Asparagus* is known to contain phytoecdysteroids that have been shown to exhibit many beneficial pharmacological properties such as improving lipid metabolism, modulating immunological responses, etc. Currently, knowledge about the contents of phytoecdysteroids in the roots of *Asparagus* species is limited and HPLC methods for their analyses are unsatisfactory.

Objective: To develop an HPLC method for the simultaneous determination of three phytoecdysteroids, 20-hydroxyecdysone, ecdysone and ajugasterone C, in the roots of four *Asparagus* species.

Methodology: Reference standards of phytoecdysteroids were isolated from the roots of *Asparagus filicinus* by open column chromatography. HPLC analysis was performed on an Alltima C(18) column with gradient elution using aqueous 0.2% formic acid solution containing 0.2% isopropanol and acetonitrile.

Results: All calibration curves showed good linear correlation coefficients ($r(2) > 0.9994$) within the tested ranges. Limits of detection (S/N = 3) and quantification (S/N = 10) for the three analytes were less than 2.7 and 9.9 ng, respectively. Intra- and inter-day RSDs of retention times and peak areas were less than 2.61%. The recoveries were between 93.2 and 107.5%, and the RSDs were less than 3.83% for the root samples of *A. filicinus*.

Conclusion: The HPLC method established is appropriate for the efficient quantitative and qualitative analyses of important phytoecdysteroids in *Asparagus* species. This study showed that *A. filicinus* is rich in phytoecdysteroids, especially 20-hydroxyecdysone. However the three studied phytoecdysteroids were not detected in *A. cochinchinensis*, *A. officinalis* and *A. setaceus*.

Wu J-J., Cheng K-W., Zuo X-F., Wang M-F., Li P., Zhang L-Y., Wang H. and Ye W-C. (2010) Steroidal saponins and ecdysterone from *Asparagus filicinus* and their cytotoxic activities. *Steroids* 75 734–739.

Abstract: Two new spirostanoides, filiasparosides E (1) and F (2), one new furostanoside, filiasparoside G (3), and one new ecdysterone, stachysterone A-20, 22-acetonide (4), together with six known steroidal saponins, asparagusin A (5), filiasparoside A (6), filiasparoside B (7), aspafilioside A (8), aspafilioside B (9), and filiasparoside C (10) were isolated from the roots of *Asparagus filicinus* Buch.-Ham. Their structures were elucidated on the basis of spectroscopic and chemical evidence. Compounds 1-10 were investigated for their cytotoxicities against human breast adenocarcinoma MDA-MB-231 cell line and compounds 8-10 exhibited cytotoxic activities with IC(50) values ranging from 3.4 to 6.6 μ M.

Wu M., Zhao S., Ren L., Wang R., Bai X., Han H., Li B. and Chen H. (2011) Research on relationship between tissue quantitative distribution of 3H-*Achyranthes bidentata* ecdysterone and channel-tropism of herbal drugs in mice. *Chinese Journal of Chinese Materia Medica* 36(21), 3018-3022 [in Chinese, with English Abstract].

Abstract:

Objective: To study the relationship between tissue quantitative distribution and pharmacokinetics of 3H-*Achyranthes bidentata* ecdysterone and the channel-tropism of herbal drugs in mice.

Method: 3H-*Achyranthes bidentata* ecdysterone was used as a tracer agent and injected into mice by the caudal vein. In 36 hours, the contents of the tracer agent of samples involving 9 different tracing phases and organ or tissue were determined in order to observe the dynamic quantitative distribution and excretion and pharmacokinetics of 3H-*Achyranthes bidentata* ecdysterone and to understand the channel-tropism of herbal drugs *Achyranthes bidentata*.

Result: 3H-*Achyranthes bidentata* ecdysterone of same organs in different tracing phases and the contents of 3H-*Achyranthes bidentata* ecdysterone in same tracing phases of different organs were significantly different ($P < 0.01$). 3H-*Achyranthes bidentata* ecdysterone was mainly distributed, in the liver, kidney, adrenal gland, small intestine and lung. The concentration-time profiles of *Achyranthes bidentata* ecdysterone in rats injected into mice by the caudal vein were shown to fit a two-compartment open model with half-lives of (778.65 \pm 12.36) min, the elimination of *Achyranthes bidentata* ecdysterone from plasma was found to be in accord with linear kinetics.

Conclusion: The above mentioned selective distribution of 3H-*Achyranthes bidentata* ecdysterone basically coincides with the meridian affinity and zang fu selection of the traditional Chinese medicine drug *Achyranthes bidentata*. This study will provide a scientific basis for the channel-tropism of *A. bidentata*.

Wu P., Xie H., Tao W., Miao S. and Wei X. (2010) Phytoecdysteroids from the rhizomes of *Brainea insignis*. *Phytochemistry* 71 975–981.

Abstract: Phytoecdysteroid glucosides, brainesterosides A-E, were isolated from the rhizomes of *Brainea insignis* along with three known phytoecdysteroids, ponasteroside A, ponasterone A, and 20-hydroxyecdysone. Their structures were elucidated by spectroscopic and chemical means. A possible biogenetic pathway is postulated for these compounds. The chemosystematic significance of ponasterone A is discussed.

Xiao C-m., Huang J., Tan X-y. Tang M. and Zhang H. (2009) *Chemical constituents of Paris polyphylla var. pseudothibetica*. *Huaxi Yaoxue Zazhi* 24(1), 7-9 [in Chinese].

Abstract: The aim of this paper is to study the chem. constituents of *Paris polyphylla var. pseudothibetica*. The chem. constituents were isolated and purified by silica column chromatog. Their structures were identified by chem. and spectral anal. Five compounds were isolated and identified as stigmaterol 3-O- β -D-glucopyranoside (1), 20 β -

hydroxy-ecdysone (II), β -L-thymidine (III), pennogenin 3-O- α -D-rhamnopyranosyl-(1 \rightarrow 2)- β -D-glucopyranoside (IV) and pennogenin 3-O- α -L-rhamnopyranosyl-(1 \rightarrow 4)- α -L-rhamnopyranosyl-(1 \rightarrow 4)-[α -L-rhamnopyranosyl-(1 \rightarrow 2)- β -D-glucopyranoside (V). Compound III from this genus and all compounds from this plant were isolated for the first time.

Xu B-l. and Jiang J-q., (2014) Study on chemical constituents of ethyl acetate fraction of *Humulus scandens*. Jiangsu Pharmaceutical and Clinical Research (2), 124-127 [in Chinese, with an English abstract].

Abstract: Eleven compounds isolated from the ethyl acetate fraction of 80% ethanol extract of *Humulus scandens*(Lour.) Merr.were identified as buddleoside(1),5,4'-dihydroxy-3,7-dimethoxy-6-C-methylfavone(2),meso-dihydroguaiaretic acid(3), β -ecdysone(4),(24S)-5 α -lanost-9(11)-ene-3 β ,24,25-triol(5),asiatic acid(6),p-hydroxybenzoic acid(7),trans-cinnamic acid(8),coniferyl aldehyde(9),sinapic aldehyde(10) and uracil(11).Compounds 1~5 and 7~11 were firstly isolated from the plants in *Humulus* Linn.and compound 6 was isolated from this plant for the first time.

Xu H. (2010) Determination of β -ecdysone in *Cyanotis vaga* (Lour.) Roem. et Schult by HPLC. Journal of Guangxi Academy of Sciences (3), 239-241 [in Chinese].

Xu H., Shi X., Ji X., Du Y., Zhu H. and Zhang L. (2011) Qualitative and quantitative determination of nine main active constituents in *Pulsatilla cernua* by high-performance liquid chromatography coupled to electrospray ionization tandem mass spectrometry. Journal of Separation Science 34 308–316

Abstract: A novel qualitative and quantitative method using high-performance liquid chromatography coupled to electrospray ionization tandem mass spectrometry (HPLC-ESI-MS/MS) was developed for simultaneous determination of the nine major active constituents in *Pulsatilla cernua* (Thunb.) Bercht. et Opiz., namely anemoside A3 (1), anemoside B4 (2), 23-hydroxybetulinic acid (3), cirenshenoside S (4), pulsatilloside B (5), pulsatilloside C (6), oleanolic acid (7), ajugasterone C (8) and β -ecdysterone (9), respectively. A Sapphire C18 column (250 mm \times 4.6 mm, 5 μ m) and gradient elution were used during the analysis. The identification and quantification of the analytes were achieved on a hybrid quadrupole linear ion trap mass spectrometer. Multiple-reaction monitoring (MRM) scanning was employed for quantification with switching electrospray ion source polarity between positive and negative modes in a single run. All calibration curves showed good linearity ($r^2 > 0.9948$) within the test ranges. The intra and interday variations for nine analytes were less than 3.95 and 3.78%, respectively. The developed method was successfully applied to determine the investigated compounds in 15 batches of natural and cultured samples of *P. cernua*. The results indicated that the method was simple, rapid, specific and reliable, which is helpful to comprehensive evaluation of quality of *P. cernua*.

Xu Y., Wang Z., Zhou R., Liu X., Wang Z. And Liu Z. (2016) Content determination of ecdysterone in different parts of *Radix serratulae* by HPLC. China Pharmacy 27(15), 2147-2149.

Abstract: OBJECTIVE: To establish a method for the content determination of ecdysterone in different parts of *Radix serratulae*. METHODS: HPLC was performed on the column of ZORBAX Eclipse XDB-C18 with mobile phase of methanol-water at a flow-rate of 1.0 ml/min (46:54, V/V), detection wavelength was 248 nm, column temperature was 30°C, and the injection volume was 10 μ L. Compare the contents of ecdysterone in different parts of *R. serratulae*. RESULTS: The linear range of ecdysterone was 0.12-1.21 μ g; RSDs of precision, stability and reproducibility tests were lower than 1.5%; recovery was 98.4%-101.9% (RSD=1.64, n=6). The content of ecdysterone in roots was found at the highest level, followed by tubers, and lower in stems and leaves, and it was not detected in seeds. CONCLUSIONS: The method is simple and rapid with good accuracy and reproducibility, and suitable for the content determination of ecdysterone in different parts of *R. serratulae*. It is feasible to develop the medicinal parts of *Radix serratulae* from roots to roots, flowers, tubers and buds on tubers.

Ya J., Zhang X., Ye W., Xu Y. and Shi Y. (2006) Chemical constituents from the seeds of *Morus atropurpurea*. Zhongguo Yaoke Daxue Xuebao 37(4), 301-303 [in Chinese].

Abstract: The chem. constituents of the seeds of *Morus atropurpurea* were studied. The constituents were isolated and purified by column chromatogs. Their structures were identified by physico-chem. properties and spectral data. Ten compounds were isolated from the seeds of *Morus atropurpurea*, and their structures were elucidated as β -ecdysone (1), friedelin (2), ursolic acid (3), naringin (4), naringenin (5), kaempferol (6), quercetin (7), 1-O- β -D-glucopyranosyl benzoate (8), benzoic acid (9), and β -sitosterol. All compounds are isolated from this plant for the first time

Yakubova M.R., Genkinka J.L., Shakirov T.T. and Abubakirov N.K. (1978) Chromatospectrometric method of detecting ecdysterone in plants. Khimiya Prirodnykh Soedinenii 737-740 [in Russian]/Chemistry of Natural Products 627-629 (1979) [in English].

Yakubova M.R. and Sakharova N.A. (1980) The dynamics of ecdysterone content in underground parts of *Rhaponticum carthamoides* (Willd.) Iljin. *Rastitelny Resursy* **16**, 98-100 [in Russian].

Abstract: Specimens of *R. carthamoides* (Willd.) Iljin (a valuable medicinal plant) were collected in different locations of the Kuznetskii Ala Tau area, USSR. Ecdysterone content significantly changes, depending on the developmental phase. Its highest amount (up to 0.81%) was found at the beginning of vegetation, and a lower amount (up to 0.51%), in the autumn vegetation phase. Minimal ecdysterone content was found during blooming phase. Ecdysterone accumulation is influenced by meteorological factors: it is the highest during a year with a moderately warm and moderately moist summer. No significant changes were found in ecdysterone content of the subterranean organs of *R. carthamoides* within the whole area of Kuznetskii Ala Tau.

Yang B., Fan Z., Zhu J-p., Yang T., Peng G-t., Tan Q-l. and Zhao Z-x. (2014) Chemical constituents from *Phymatopteris hastata*. *Chinese Traditional and Herbal Drugs* (21), 3053-3056 [in Chinese, with an English abstract].

Abstract:

Objective: To study the chemical constituents from the whole plants of *Phymatopteris hastata*. Methods: The chemical constituents were isolated by repeated chromatography on silica gel column, ODS column, and Sephadex LH-20. Their structures were elucidated by physicochemical properties and spectral analysis. Results: Fifteen compounds were isolated from methanolic extract of *P. hastata* and identified as kaempferol-3-O- α -L-arabinofuranosyl-7-O- α -L-rhamnopyranoside (1), caffeic acid-4-O- β -D-glucopyranoside (2), kaempferol-7-O- α -L-rhamnopyranoside (3), kaempferol-3, 7-di-O- α -L-rhamnopyranoside (4), (-)-epiafzelechin (5), kaempferol (6), quercetin (7), (24R)-24-(2-hydroxyethyl)-20-hydroxyecdysone (8), hop-22 (29)-ene (9), hapan-29-ol acetate (10), 22-hydroxyhopane (11), dryocrassol (12), β -sitosterol (13), fumaric acid (14), and β -daucosterol (15). Conclusion: Compounds 5-15 are isolated from the plants of *Phymatopteris Pic. Serm.* for the first time

Yang C., Xu J., Dong Y. and Liu X. (1996) Studies on the isolation and identification of β -ecdysone from *Zebrina pendula* Schnizl. and its antiarrhythmic effect. *Tianran Chanwu Yanjiu Yu Kai Fa* (Natural Product Research and Development) **8**(3), 17-19 [in Chinese, with an English abstract].

Abstract: It was shown that β -ecdysone, isolated from *Zebrina pendula* Schizl, in some animals eliminates arrhythmia induced by aconitine.

Yang D-s., Li Z-l., Wang X., Yang Y-p., Peng W-b., Liu K-c. and Li X-l. (2015) Chemical constituents from roots of *Campanumoea javanica* and their antiangiogenic activities. *Chinese Traditional and Herbal Drugs* (4), 470-475 [in Chinese].

Abstract: Objective: To study the chemical constituents from the roots of *Campanumoea javanica* and their antiangiogenesis activities. Methods: The compounds were isolated and purified by various chromatographic techniques and their structures were elucidated by spectral analysis, the antiangiogenic activities of the isolated compounds were evaluated using a zebrafish model. Results: Fourteen compounds were isolated and identified from 90% ethanol extract in ethyl acetate fraction in the roots of *C. javanica*, including campanumoside (1), lobetyol (2), tetradeca-4E,8E,12E-triene-10-yne-1,6,7-triol (3), 9-(tetrahydropyran-2-yl)-non-trans-8-ene-4,6-diyn-3-ol (4), 9-(tetrahydropyran-2-yl)-nona-trans,trans-2,8-diene-4,6-diyn-1-ol (5), lobetyolinin (6), (Z)-3-hexenyl-O- α -L-arabinopyranosyl-(1 \rightarrow 6)- β -D-glucopyranoside (7), 3,4-dihydroxybenzoic acid (8), tangshenoside II (9), zanthocapsin (10), ampelopsin (11), agathisflavone (12), β -ecdysterone (13), and α -tocopherolquinone (14). Conclusion: Compound 1 is a new polyacetylene glucoside named campanumoside. Compounds 2—14 are isolated from the plants of *Campanumoea* Bl. for the first time. Compounds 3 and 4 exhibit the certain antiangiogenic activity in the pharmacological evaluation with a zebrafish model.

Yang F-Y., Su Y-F., Wang Y., Chai X., Han X., Wu Z-H. and Gao X-M. (2010) Bufadienolides and phytoecdysones from the rhizomes of *Helleborus thibetanus* (Ranunculaceae). *Biochemical Systematics and Ecology* **38**(4) 759-763

Abstract: Two new bufadienolide glycosides with an A/B trans ring structure, 14 β ,16 β -dihydroxy-3 β -[β -D-glucopyranosyloxy]-5 α -bufa-20,22-dienolide (1), and 14 β ,16 β -dihydroxy-3 β -[β -D-glucopyranosyl-(1 \rightarrow 4)-(β -D-glucopyranosyloxy)]-5 α -bufa-20,22-dienolide (2), two known ecdysteroids (polypodine B and 20-hydroxyecdysone) (3-4), and six known bufadienolide and its glycosides with 5 β -OH (hellebrigenin, 16 β -hydroxyhellebrigenin-3-O- α -L-rhamnoside, hellebrigenin 3-O- β -D-glucoside, hellebrin, 16 β -hydroxyhellebrigenin-3-O- β -D-glucoside, and deglucohellebrin) (5-10) were isolated from the rhizomes of *Helleborus thibetanus*. The structures of compounds 1 and 2 were elucidated using various spectroscopic methods. All compounds were reported for the first time from the title plant and their chemotaxonomic significance for the genus *Helleborus* was discussed.

Yang L., Jiang H., Yan M-L., Xing X-D. Zhang Y-Y. and Wei N. (2016) A new phytoecdysteroid from the roots of *Achyranthes bidentata* Bl. *Natural Products Research* (DOI/10.1080/14786419.2016.1272114).

Abstract: A new phytoecdysteroid compound, named Niuxixinsterone D (**1**), together with two known phytoecdysteroid compounds (**2** and **3**) were isolated from *Achyranthes bidentata* Bl.. The structure of the new compound was elucidated by extensive spectral analysis, including HR-ESI-MS, 1D and 2D NMR methods. Compounds **1–3** were tested for their inhibitory effects against LPS-induced NO production in RAW 264.7 macrophages, and compound **1** and **3** exhibited anti-neuroinflammatory activity with inhibited 29.7 and 26.0% NO production.

Yang L., Jiang H., Su X., Yan M., Xing X., Guo X., Hou A. and Man W. (2019) Simultaneous determination of β -ecdysterone, ginsenoside R0 and chikusetsusaponin IVa in radix *Achyranthes bidentata* by HPLC-DAD-ELSD. *Information on Traditional Chinese Medicine* (3), 87-91 [in Chinese].

Abstract: Objective: A method of high-performance liquid chromatography-diode array detector-evaporative light scattering detection (HPLC-DAD-ELSD) was developed for simultaneous determination of β -ecdysterone (**1**), ginsenoside R0 (**2**) and chikusetsusaponin IVa (**3**) in Radix *Achyranthes bidentata*. Methods: Optimum separations were obtained with a Agilent 5 TC-C18 column (4.6 mm \times 250 mm, 5 μ m), using a gradient elution with 0.08% aqueous formic acid (containing 2.5% isopropyl alcohol) and acetonitrile (containing 0.08% formic acid) as mobile phase. β -Ecdysterone was detected by DAD at 248 nm, whereas ginsenoside R0 and chikusetsusaponin IVa were monitored by ELSD connected in series with DAD, temperature for the drift tube was 110°C and the nitrogen flow-rate was 3.2 L/min. Results: The calibration curve of β -ecdysterone showed good linear regression ($r^2=0.9999$). The regression equation showed a good liner relationship between logarithm of the peak area and logarithm of the concentration of ginsenoside R0 and chikusetsusaponin IVa ($r^2=0.9995$, $r^2=0.9994$). The average recoveries of the three compounds were 99.29% (RSD: 2.14%), 99.13% (RSD: 2.04%) and 98.30% (RSD: 2.11%) respectively. Conclusion: The method is simple, fast, acceptable stability, precision and repeatability. The results demonstrated that the method is suitable for the simultaneous determination of ketosteroid and saponins in Radix *Achyranthes bidentata*, and the method is also can be used as the quality control of Radix *Achyranthes bidentata*.

Yang N., Wang H., Lin H., Liu J., Zhou B., Chen X., Wang C., Liu J. and Li P. (2020) Comprehensive metabolomics on UPLC-Q/TOF-MS^E and the anti-COPD effect of different parts of *Celastrus orbiculatus* Thunb. *RSC Advances* 10, 8396.

Abstract: The root, stem and leaf of *Celastrus orbiculatus* Thunb. (COT) have all been used as Chinese folk medicine. Aiming at revealing the secondary metabolites and screening the anti-COPD effect of COT, the comprehensive phytochemical and bioassay studies were performed. Based on the ultra-high performance liquid chromatography combined with quadrupole time-of-flight mass spectrometry (UPLC-Q/TOF-MS^E), the screening analysis of components in COT was conducted with the UNIFI platform, the metabolomics of the three parts were analyzed with multivariate statistical analysis. Cigarette smoke extract (CSE)-stimulated inflammatory model in A549 cells was used to investigate the biological effect of the three parts. A total of 120 compounds were identified or tentatively characterized from COT. Metabolomics analysis showed that the three parts of COT were differentiated, and there were 13, 8 and 5 potential chemical markers discovered from root, stem and leaf, respectively. Five robust chemical markers with high responses could be used for further quality control in different parts of COT. The root, stem and leaf of COT could evidently reduce the levels of pro-inflammatory factors in a dose-dependent way within a certain concentration range. The stem part had a stronger anti-COPD effect than root and leaf parts. This study clarified the structural diversity of secondary metabolites and the various patterns in different parts of COT, and provided a theoretical basis for further utilization and development of COT.

Yang S-G., Zhang X., Sun X-S., Ling T-J., Feng Y., Du X-Y., Zhao M., Yang Y., Xue D., Wang L. and Liu R-T. (2010) Diverse ecdysterones show different effects on amyloid- β_{42} aggregation but all uniformly inhibit amyloid- β_{42} -induced cytotoxicity. *Journal of Alzheimer's Disease* 22, 107-117.

Abstract: Amyloid- β (A β) plays a pivotal role in Alzheimer's disease (AD) pathogenesis and in toxic mechanisms such as oxidative stress, mitochondrial dysfunction, calcium turbulence, and apoptosis induction. Therefore, interfering with A β aggregation has long been one of the most promising strategies for AD treatment. Ecdysterones (ECRs) are steroidal hormones in insects and terrestrial plants that have high structural diversity and multiple beneficial pharmacological activities. Here, we studied the effects of six ECRs on A β aggregation and cytotoxicity. Two ECRs with an acetoxyl group at the 2 or 3 position and saturated chains as side groups showed apparent promotion of A β_{42} fibrilization, resulting in less A β_{42} oligomers in the samples. Another three with unsaturated side chains clearly inhibited A β aggregation and disaggregated preformed fibrils, but increased the A β_{42} oligomer levels. Nevertheless, our MTT results showed that all ECRs tested inhibited A β_{42} -induced cytotoxicity. This protective activity may be partly attributable to ECR-mediated amelioration of A β_{42} -induced release of reactive oxygen species. Taken together, our findings suggest that ECRs, a series of natural compounds in many plants and insects, have therapeutic potential in AD and that the deduced structure-activity relationships may be beneficial in drug design for the treatment of AD and other amyloidoses.

Yany Y-y., Chi D-f. and Yu J. (2017) Influence of exogenous ABA on endogenesis hormone and ecdysterone in suspension cultures cell of *Ajuga lobata* D. Don. Journal of Natural Science of Hunan Normal University (4), 26-33 [in Chinese].

Abstract: The optimal concentration of abscisic acid (ABA) was evaluated in this work by adding different concentrations of ABA into the suspension cell cultures system of *Ajuga lobata* D. Don. The growing status, biomass of suspension cultures cell, concentration of ecdysterone (20E) were measured after adding those substances. Upon the selection of optimal concentration, 0.15 mg·L⁻¹ ABA was added into *Ajuga lobata* D. Don suspension cell culture system, then changes of endogenous phytohormones, such as zeatin (ZR), gibberellin 3(GA3), Hetero auxin (IAA) and 20E content were determined with HPLC, and the impact of ABA on plant endogenous hormone content and 20E synthesis in suspension cultures cell of *Ajuga lobata* D. Don were analysed. Adding ABA in the suspension cell culture system, we found that the growing speed of suspension cell was decreased, but without browning, ABA promoted the accumulation of 20E. At 48 h and 72 h after the addition of ABA, the content of 20E was significantly higher than that in the control group, and at 96 h after its addition, those treated with suspension cells became browning, and the dry weight of those cells was significantly lower than that of the control group. The content of endogenous ZR, GA3 and IAA in the treated suspension cell was also decreased. After the addition of Na₂WO₄ into the suspension cell culture system, the change trend of ABA, ZR, GA3 and IAA was opposite to that of adding ABA, and their range of 20E concentration change never reached to the significant difference level as compared with that of the control group.

Yao J-N., Li Z-F., Lou H-Y., Huang L., Liang G-Y., Cao P-X. and Pan W-D. (2014) A new ecdysteroidal glycoside from *Lepidogrammitis drymoglossoides* (Bak.) Ching. Journal of Carbohydrate Chemistry 33 206-211.

Abstract: *Lepidogrammitis drymoglossoides* (Bak.) Ching is a medicinal plant that belongs to the family of Polypodiaceae. Previous phytochemical investigation on this genus had revealed the presence of flavonoids, sterols, and alkaloids. Here, further investigation on this medicinal plant led to the isolation of a new ecdysteroidal glycoside, ponasteroside B (**1**), along with a known steroidal compound, β -ecdysterone (**2**). The structure of **1** was determined on the basis of extensive spectroscopic analysis, including HR-MS and 1D and 2D NMR experiments, as well as a chemical hydrolysis study.

Yao Q.Y. and Hu D.F. (1989) Determination of ecdysones in polyploid and monoploid *Acyranthes bidentata*. Zhongguo Zhongyao Zashi 14(4), 210-211 [in Chinese].

Abstract: Ecdysones, i.e. ecdysterone and inokosterone, were determined in pharmaceutical preparations of roots of polyploid, diploid, and monoploid *A. bidentata* by column chromatog. on an alumina column. The contents of ecdysones in monoploid and diploid *A. bidentata* were similar, but in polyploids the drug content was >14-fold higher.

Yatsyuk Y.K. and Segel G.M. (1970) The separation of ecdysterone. Khimiya Prirodnikh Soedinenii (6), 281 [in Russian].

Yeh P.-H. and Chiang H.-C. (1982) Studies on the constituents of *Paris formosana* Hayata Part II. Journal of the Chinese Chemical Society 29, 39-46.

Abstract: Methyl palmitate (I), methyl stearate (II), stigmasterol (III), β -sitosterol (IV), (O-acyl)- β -D-glucopyranosyl-(13)-stigmasterol (V), (O-acyl)- β -D-glucopyranosyl-(13)- β -sitosterol (VI), β -D-glucopyranosyl-(13)-stigmasterol (VII), β -D-glucopyranosyl-(13)- β -sitosterol (VIII), β -D-ecdysone (IX), diosgenin-3- α -L-rhamnopyranosyl-(12)-[α -L-arabinofuranosyl-(14)]- β -D-glucopyranoside (X), diosgenin-3-O- β -chacotrioxide (dioscin) (XI), and diosgenin-3-O- α -L-rhamnopyranosyl-(14)- α -L-rhamnopyranosyl-(14)-[α -L-rhamnopyranosyl-(12)]- β -D-glucopyranoside (XII) were isolated and characterized from the stems of *Paris formosana* Hayata (Liliaceae).

Yen K-Y., Yang L-L., Okuyama T., Hikino H. and Takemoto T. (1974) Screening of Formosan ferns for phytoecdysones. 1. Chemical and Pharmaceutical Bulletin 22, 805-808.

Yi J.H., Luo Y.G., Li B.G. and Zhang G.L. (2004) Phytoecdysteroids and glyceramide from *Eriophyton wallchii*. Steroids 69(13/14), 809-815.

Abstract: Two new compounds, 28-epi-cyasterone and eriophytonoide, along with 11 known compounds, cyasterone, ajuforrestins A and B, 20-hydroxyecdysone, polypodin B, ajugalactone, 8-O-acetylharpagid, apigenin, N-[2hydroxy-(nonadecanoyl-tricosanoyl)]-4-hydroxy-trans-8-sphingenine, beta-sitosterol, and daucosterol, were isolated from the aqueous ethanolic extract of the whole herb of *Eriophyton wallchii* Benth. The structures of 28-epi-cyasterone and eriophytonoide were elucidated as (22R,24S,25S,28S)-5beta-stigmast-7-en-26-oic acid, 2beta,3beta,14,20,22,28-hexa hydroxy-6-oxo-gamma-lactone, and 1-O-beta-d-glucopyranosyl-(2S,3S,4R,8E)-2-

[(2R)-2-hydroxyhexadecanoylamino]-1,3,4-octadecanetriol-8-ene, respectively, on the basis of spectral and chemical evidence.

Yin W., Song Z.R., Liu J.Q. and Zhang G.S. (2015a) Chemical constituents of *Paris polyphylla* var. *chinensis* aerial parts. *Zhong Yao Cai* 38(9), 1875-1878 [in Chinese].

Abstract:

Objective: To study the chemical constituents of aerial parts of *Paris polyphylla* var. *chinensis*.

Methods: Aerial parts of *Paris polyphylla* var. *chinensis* was extracted with 95% EtOH, and separated and purified by silica gel, RP 18 and Sephadex LH-20 col- umn chromatography. The structures were identified by spectroscopic analysis.

Results: A total of ten compounds were isolated and identified as β -sitosterol (1) ergosta-7, 22-dien-3-one (2), β -ecdysone (3), kaempferol (4), daucosterol (5) luteolin (6) calonysterone (7), luteolin-7-O-glucoside (8), quercetin (9), and 3β , 5α , 9α -trihydroxyergosta-7, 22-dien-6-one (10).

Conclusion: Compounds 2,6 and 10 are isolated from *Paris polyphylla* var. *chinensis* for the first time.

Yin W., Song Z.R., Liu J.Q. and Zhang G.S. (2015b) Chemical constituents of *Citrus medica* fruit. *Zhong Yao Cai* 38(10), 2091-2094.

Abstract:

Objective: To study the chemical constituents of *Citrus medica* fruit.

Methods: The fruit of *Citrus medica* was extracted with 95% EtOH, and the compounds were separated and purified by silica gel, RP-18 and Sephadex LH-20 column chromatography. The structures were identified by spectroscopic analysis.

Results: A total of 16 compounds were isolated and identified, including methyl ferulic acid (1), dihydro-N-caffeoyltyramine (2) acacetin (3), β -ecdysterone (4), (-)-balanophonin (5), p-methoxy cinnamic acid (6), umbelliferone (7), ferulic acid (8), pyrocatechualdehyde (9), diosmetin (10), 4-methoxy salicylic acid (1), β -amyrin acetate (12), epigallocatechin (13), betulinic acid (14), lupeol (15) and nicotinamide (16).

Conclusion: All the compounds are isolated from the fruit of *Citrus medica* for the first time.

Yokosuka, A., Inomata, M., Yoshizawa, Y., Iguchi T. and Mimaki Y. (2021) Bufadienolides and ecdysteroids from the whole plants of *Helleborus niger* and their cytotoxicity. *Journal of Natural Medicines* <https://doi.org/10.1007/s11418-021-01481-6>.

Abstract: A new bufadienolide (**1**), two new bufadienolide glycosides (**2** and **3**), a new ecdysteroid (**4**), and four known compounds (**5–8**), were isolated from the whole plants of *Helleborus niger* L. (Ranunculaceae). The structures of the new compounds (**1–4**) were determined by spectroscopic analysis, including 2D NMR spectral data, and hydrolytic studies. Compounds **1–6** showed cytotoxicity against HL-60 human leukemia cells, A549 human lung adenocarcinoma cells, and SBC-3 human small-cell lung cancer cells, with IC₅₀ values ranging from 0.0055 to 1.9 μ M. HL-60 cells treated with either **3** or **4** showed apoptosis characteristics, such as nuclear chromatin condensation, accumulation of sub-G₁ cells, and activation of caspase-3/7.

Yusupova U.Y., Usmanov D.A. and Ramazonov N.S. (2019) Phytoecdysteroids from the plant *Dianthus helena*. *Chemistry of Natural Compounds* 55(2), 393-394 [in English]/*Khimiya Prirodnykh Soedinenii* (2), 329 [in Russian].

Abstract: Phytoecdysteroids are a large group of polyhydroxylated steroids with wound-healing, adaptogenic, immunomodulating, anabolic, and hypoglycemic activity [1]. A literature analysis showed that the family Caryophyllaceae comprised the greatest number of species containing ecdysteroids. Several ecdysteroids were isolated from plants of the genus *Dianthus*, e.g., *D. angrenicus*, *D. hoeltzeri* [2], and *D. uzbekistanicus*. Therefore, the endemic plant *D. helena* was studied in a search for new raw-material sources containing ecdysteroids. Dried and ground aerial part of the plant (2.0 kg) was extracted (4 \times) with MeOH (7 L). The extract was concentrated and diluted with an equal volume of H₂O. The resulting precipitate was filtered off and evaporated. The aqueous part was extracted sequentially with CHCl₃ and EtOAc. The EtOAc fraction was evaporated in a rotary evaporator to produce a thick resinous residue (55 g) that was separated by column chromatography over silica gel (30:1) with elution by CHCl₃–MeOH (100:1, 50:1, 30:1, 20:1, 15:1, 9:1, 4:1) to isolate pure polygodine B (**1**); 20-hydroxyecdysone 2,3,20,22-diacetonide (**2**), 20-hydroxyecdysone (**3**), and cyasterone (**4**).

Yusupova U.Y., Usmanov D.A. and Ramazonov N.S. (2020a) Phytoecdysteroids from the aerial part of *Silene popovii*. *Chemistry of Natural Compounds* 56(3), 562-563,[in English]/*Khimiya Prirodnykh Soedinenii* (3), 483-484 [in Russian].

Abstract: Phytoecdysteroids are physiologically active compounds and analogs of insect molting and metamorphosis hormones with adaptogenic, immunomodulating, wound-healing, anabolic, and hypoglycemic activity. Members of the family Caryophyllaceae that represent the genera *Dianthus* and *Silene* are promising sources of phytoecdysteroids. The endemic plant *S. popovii* was studied by us for the first time to seek new sources

of phytoecdysteroids. The dried and milled aerial plant part (1.0 kg) was extracted with MeOH (5 × 6 L). The extract was concentrated and diluted with an equal volume of H₂O. The resulting precipitate was filtered off. The MeOH was evaporated. The aqueous residue was extracted sequentially with CHCl₃ and n-BuOH. The BuOH fraction was concentrated under vacuum at 40°C to produce a thick resinous residue (22 g). The BuOH fraction was chromatographed over silica gel with elution by CHCl₃–MeOH (100:1, 50:1, 30:1, 20:1, 15:1, 9:1, 6:1, 4:1) to isolate pure cyasterone (1), polypodine B (2), 20-hydroxyecdysone (3), 2-deoxyecdysone (4), and the 2,3,20,22-diacetonide of 20-hydroxyecdysone (5).

Yusupova U., Usmanov D., Azamatov A., Ramazonov N. and Rejepov J. (2020b) Phytochemical constituents and biological activities of *Dianthus helenae* Vved., growing in Uzbekistan. *Natural Product Research* (DOI: [10.1080/14786419.2020.1862834](https://doi.org/10.1080/14786419.2020.1862834)).

Abstract: In the present study, six known compounds were investigated that were isolated from the aerial parts of plant *Dianthus helenae* Vved. The structures of these compounds were identified as polypodine B (1), 2,3,20,22-diacetonide-20-hydroxyecdysone (2), 20-hydroxyecdysone (3), cyasterone (4), α -ecdysone (5) and 2-deoxy- α -ecdysone (6). Their structures were confirmed by NMR-, ESI-MS, and IR-spectroscopy. The compounds (5) and (6) are reported for the first time from this species. Furthermore, compounds (2) and (4) were isolated for the first time from the *Caryophyllaceae* family. In addition, all these phytoecdysteroids were investigated for a nootropic activity. Thus, the total phytoecdysteroids-containing preparation at a dose of 25 mg/kg increases two times the motor activity, an approximate reaction – four times and exploratory behavior – 1.4 times compared to control animals.

Yusupova U.Y., Sasmakov S.A., Usmanov D.A., Ramazonov N.S., Azimova S.S. and Sagdullaev S.S. (2020c) Phytoecdysteroids from *Silene claviformis* and their antibacterial and antifungal activities. *Polish Journal of Natural Sciences* **35**(3), 313-321.

Abstract: Phytoecdysteroid compounds, such as makisterone A (1), polypodine B (2), 20-hydroxyecdysone (3), 2,3,20,22-diasetanide 20-hydroxyecdysone (4), integristeron A (5), cyasterone (6), 5 α -2-deoxy- α -ecdysone (7), α -ecdysone (8) were isolated from *Silene claviformis* plant and their structures were confirmed by NMR, ¹H and IR spectroscopy. In addition, an antibacterial and antifungal potential of each pure compounds and plant extracts were assessed against different microorganisms using the agar-discs diffusion assay. Results revealed that *S. claviformis* extracts and individual phytoecdysteroids did not exhibit antimicrobial activity against tested strains of microorganisms.

Zakirova R.P. and Malikova M.K. (2000) Effect of N-nitroso-N-methylurea on the biosynthetic activity of *Ajuga turkestanica* callus tissue. *Khimia Prirodnykh Soedinenii* (4), 315-317 [in Russian]/*Chemistry of Natural Compounds* **36**(4), 384-386 [in English].

Zakirova R.P. and Malikova M.K. (2001) Accumulation dynamics of ecdysterone and carbohydrates in callus tissue of *Ajuga turkestanica*. *Khimiya Prirodnykh Soedinenii* (3), 226-227 [in Russian]/*Chemistry of Natural Compounds* **37**(3) 266-268 [in English].

Abstract: The accumulation dynamics of ecdysterone, water-soluble polysaccharides, and pectinic substances during growth of callus tissue of *Ajuga turkestanica* were studied.

Zakirova R.P., Putieva J.M. and Saatov Z. (1998) Influence of the conditions of cultivation on the growth of callus and cell cultures of *Ajuga turkestanica* and the biosynthesis of ecdysterone. *Khimiya Prirodnykh Soedinenii* (4), 505-509 [in Russian].

Abstract: The influence of light on the total productivity of a tissue culture of *Ajuga turkestanica* has been studied. When calluses are grown in the absence of light the yield of biomass and its ecdysterone content increase. In a study of the influence of α -naphthylacetic acid on the productivity of a suspension cell culture it has been shown that with an increase in the concentration of the auxin the yield of crude mass rises but the ecdysterone content falls. The activation of the biosynthesis of the secondary product takes place with a retardation of the processes of growth and cell division.

Zarembo E.V., Rybin V.G., Boltentkov E.V., Verbitskii G.A. and Gorovoi P.G. (2003) 20-Hydroxyecdysone from *Stemmacantha uniflora* subsp. *satzyperovii*. *Chemistry of Natural Compounds* **39**(5), 479-481/*Khimiya Prirodnykh Soedinenii* (5), 392-394 (2003).

Abstract: The dynamics of 20-hydroxyecdysone content in vegetative and generative organs of *Stemmacantha uniflora* subsp. *satzyperovii* (Soskov) Dittrich, which is distributed over Primorskii Krai, were investigated. A high content of 20-hydroxyecdysone during the plant vegetative period is characteristic of growing organs. The amount is maximal for young leaves during development of racemes (7.8 g/mg) and for ripe achenes (11.15 g/mg).

Zatsny I.L., Gorovits M.B. and Abubakirov N.K. (1971) Ecdysterone from *Serratula sogdiana*. *Khimiya Prirodnykh Soedinenii* (6), 840-841 [in Russian].

Abstract: Compound (I) was acetylated with acetic anhydride in pyridine (40°C, 4 h). The reaction product was separated on a column of silica gel. Elution with chloroform and with chloroform-ethanol (19:1) gave a tetraacetate of substance (1), C₃₅H₅-O₁₁, mp 204-206°C (acetone-hexane), [α]_D + 59.6° (c 0.89; CH₂OH), and a triacetate C₃₃H₁₅O₁₀ with mp 196-198°C (ether), [α]_D + 57.7° (c 0.75; CH₂OH) [2]. When (I) was treated with anhydrous acetone in the presence of phosphomolybdic acid (room temperature, 2 h), with subsequent separation in a thin layer of silica gel in the chloroform-ethanol (19:1) system, a diacetonide of (1), C₃₃H₁₅-O₇, was obtained with mp 232-233.5°C (ether-hexane), [α]_D + 40.5 (c 0.88; CH₂OH) [2].

Zatsny I.L., Gorovits M.B. and Abubakirov N.K. (1973) Phytoecdysones of *Serratula* II. Viticosterone E from *Serratula sogdiana* and its partial synthesis. *Khimiya Prirodnykh Soedinenii* 175-178.

Summary: A phytoecdysone identified as viticosterone E has been isolated from a methanolic extract of the leaves of *Serratula sogdiana* Bge. 2. A partial synthesis of viticosterone E from ecdysterone has been effected by two methods — via the 2,3: 20,22-diacetonide and via the 2,3,22,25-tetraacetate.

Zatsny I.L., Gorovits M.B., Rashkes J.V. and Abubakirov N.K. (1975) Phytoecdysones of *Serratula*. III. Mass spectrometric studies of ecdysterone acetates and acetonides and viticosterone E. *Khimiya Prirodnykh Soedinenii* 155-158 [in Russian].

Abstract: Features of the fragmentation of the side chain of the acetates and acetonides of ecdysterone and of viticosterone E have been studied. It has been shown that the presence of an oxygen-containing function at C₂₅ in these compounds imparts to the spectra certain distinguishing features which have analytical value.

Zeng J., Shang X., Zhang P., Wang H., Gu Y. and Tan J-N. (2019) Combined use of deep eutectic solvents, macroporous resins, and preparative liquid chromatography for the isolation and purification of flavonoids and 20-hydroxyecdysone from *Chenopodium quinoa* Willd. *Biomolecules* 9, 776 (doi:10.3390/biom9120776).

Abstract: Deep eutectic solvents (DESs) were used in combination with macroporous resins to isolate and purify flavonoids and 20-hydroxyecdysone from *Chenopodium quinoa* Willd by preparative high-performance liquid chromatography (HPLC). The extraction performances of six DESs and the adsorption/desorption performances of five resins (AB-8, D101, HPD 400, HPD 600, and NKA-9) were investigated using the total flavonoid and 20-hydroxyecdysone extraction yields as the evaluation criteria, and the best-performing DES (choline chloride/urea, DES-6) and macroporous resin (D101) were further employed for phytochemical extraction and DES removal, respectively. The purified extract was subjected to preparative HPLC, and the five collected fractions were purified in a successive round of preparative HPLC to isolate three flavonoids and 20-hydroxyecdysone, which were identified by spectroscopic techniques. The use of a DES in this study significantly facilitated the preparative-scale isolation and purification of polar phytochemicals from complex plant systems.

Zeng X.N., Coll J., Camps F. and Palacin M.J. (2000) Analysis of phytoecdysteroids in cultured plants of *Ajuga nipponensis* Makino. *Journal of Asian Natural Products Research* 2(4), 263-269.

Abstract: Cultured plants of *Ajuga nipponensis* contained cyasterone (1), ajugasterone C (2), cyasterone-22-acetate (3) and 22-dehydrocyasterone (4) based on HPLC and NMR data, whereas 20-hydroxyecdysone was not detectable. The presence of compounds 2-4 is reported for the first time in this species. Compound 1 is the main phytoecdysteroid component found in both preblossom and blossom plants, but the latter contained higher amount than the former. Compared with other parts of the plant, the highest percentage of 1 and 3 occurred in leaves, amounting to 60.1% and 88.0% respectively, whereas the flowers contained mainly 2, which represented 72.8% of the total amount in whole plant. The contents of phytoecdysteroids in stems were very low.

Zeng Y., Lu Y., Chen Z., Tan J., Bai J., Li P., Wang Z. and Du S. (2018) Rapid characterization of compounds in *Bolbostemma paniculatum* by UPLC/LTQ-Orbitrap MSⁿ analysis and multivariate statistical analysis for herb discrimination. *Molecules* 23, 1155 (doi: 10.3390/molecules23051155).

Abstract: *Bolbostemma paniculatum* is a traditional Chinese medicine (TCM) showed various therapeutic effects. Owing to its complex chemical composition, few investigations have acquired a comprehensive cognition for the chemical profiles of this herb and explicated the differences between samples collected from different places. In this study, a strategy based on UPLC tandem LTQ-Orbitrap MSⁿ was established for characterizing chemical components of *B. paniculatum*. Through a systematic identification strategy, a total of 60 components in *B. paniculatum* were rapidly separated in 30 min and identified. Then based on peak intensities of all the characterized components, principle component analysis (PCA) and hierarchical cluster analysis (HCA) were employed to classify 18 batches of *B. paniculatum* into four groups, which were highly consistent with the four climate types of their original places. And five compounds were finally screened out as chemical markers to discriminate the internal quality of *B. paniculatum*. As the first study to systematically characterize the chemical components of *B.*

paniculatum by UPLC-MSn, the above results could offer essential data for its pharmacological research. And the current strategy could provide useful reference for future investigations on discovery of important chemical constituents in TCM, as well as establishment of quality control and evaluation method.

Zhang H., Tan Y. and Dong X. (2020) Two new ecdysteroid glycosides from the rhizomes of *Silene tartarinowii* Regel. *Records of Natural Products* (doi.org/10.25135/mp.195.20.07.1726) pp. 8.

Abstract: Two new ecdysteroid glycosides, sileneosides I and J (**1** and **2**), along with four known ecdysteroids (**3–6**) were isolated from the rhizomes of *Silene tatarinowii* Regel. Their structures were determined based on physicochemical properties and spectroscopic methods. In addition, the cytotoxicity of compounds **1–6** was evaluated *in vitro* in human SGC790, HCT116, A549, and BEL7404 tumor cell lines. The results showed that compounds **1** and **2** exhibited weak cytotoxicity against SGC790 (IC₅₀ 82.58 ± 0.53 μM) and A549 cells (96.62 ± 0.58 μM), respectively.

Zhang L.-j., Sun D.-d., Tu W.-q. and Liu Q.-s. (2013a) Study on determination of β-ecdysterone and fingerprints of *Achyranthes bidentata* Bl. from different areas. *Natural Product Research and Development* (4), 500-505 and 510 [in Chinese].

Abstract: The HPLC content determination and fingerprinting method of β-moulting steroids in ox-knee plants were established to compare the mass differences between different origins of ox-knee. Waters SunFire C18 chromatography column (250mm×4.6mm, 5 μm, methanol and water as the mobile phase, gradient elution, flow-rate of 1.0 mL/min, column temperature of 30 degrees C, detection wavelength 250 nm. β-moulting ketones at 0.26 μg to 2.60 μg linear relationship is good (r=0.9994), the average recovery rate is 100.29% (RSD-2.42%, n=6); The method is simple, accurate and reproducible, which can provide the basis for the quality control and geo-research of ox-knee.

Zhang L.-j., Zhang G.-. and Tu W.-q. (2013b) Simultaneous determination of β-ecdysterone and chikusetsusaponin-1 in *Achyranthes bidentata* Bl. by HPLC. *Chinese Traditional Patent Medicine* (5), 1010-1013 [in Chinese].

Abstract: Objective: to establish an efficient liquid chromatography method for the simultaneous determination of β-moulting steroids and bamboo ginseng-1 in ox-knee plants.

Methods: Waters SunFire C18 column (250 mm×4.6 mm, 5 μm), methanol-water as the mobile phase, gradient elution, volume flow of 1.0 mL/min, column temperature of 30 degrees C, maximum absorption wavelength detection (β-moulting ketone 250 nm, bamboo ginseng-1 is 240 nm). Results: β-moulting steroids showed a good linear relationship in the range of 0.515 to 1.545 μg, and bamboo ginseng-1 in the range of 4.965 to 14.895 μg, with correlation coefficients of 0.9998 and 0.9999, respectively. β-moulting steroids and bamboo ginseng-1 had an average recovery rate of 99.17% (RSD 1.44%) and 98.70% (RSD 1.03%), respectively. Conclusion: The method is simple, accurate and reproducible, and can be used for the quality control of ox-knee.

Zhang L.-j., Liu X.-m., Tu W.-q. and Li X.-y. (2018) Simultaneous determination of five components in *Achyranthes bidentata* Radix by HPLC. *Chinese Journal of Pharmaceutical Analysis* (4), 623-629 [in Chinese].

Abstract: Objective: To establish an HPLC method for simultaneous determination of β-ecdysterone, 25R-inokosterone, 25S-inokosterone, ginsenoside Ro and chikusetsusaponin IVa in *Achyranthes bidentata* Radix. Methods: The chromatographic separation was performed on a ZORBAX SB-C18 column (4.6 mm×250 mm, 5 μm) with the mobile phase consisting of acetonitrile (A) and 0.5% phosphoric acid (B) in gradient elution mode at a flow-rate of 1.0 mL·min⁻¹. The detection wavelength was set at 250 nm for β-ecdysterone, 25R-inokosterone and 25S-inokosterone, and 203 nm for ginsenoside Ro and chikusetsusaponin IVa. Results: The analytes were separated well. The linear ranges of β-ecdysterone, 25R-inokosterone, 25S-inokosterone, ginsenoside Ro and chikusetsusaponin IVa were 0.218-2.725 μg (r=0.999 8), 0.080-0.995 μg (r=0.999 9), 0.155-1.935 μg (r=0.999 9), 0.352-4.400 μg (r=0.999 8) and 0.338-4.225 μg (r=0.999 8), respectively. The average recovery rates were 98.3%, 101.5%, 98.4%, 99.3% and 101.4%, with RSD of 0.89%, 0.93%, 1.4%, 0.69% and 1.3%, respectively. In *Achyranthes bidentata* Radix from different habitats, the contents of β-ecdysterone, 25R-inokosterone, 25S-inokosterone, ginsenoside Ro and chikusetsusaponin IVa were in ranges of 0.032%-0.074%, 0.008%-0.017%, 0.008%-0.017%, 0.044%-0.107% and 0.047%-0.103%, respectively. Conclusion: The method can be used for quality evaluation of *Achyranthes bidentata* Radix.

Zhang L.-y., Ren L.-z., Wang T.-h., Dong X., Wan M.-z., Wu H.-f., Mei Q.-x. and Gao Y.-h. (2014) Chemical constituents from *Lepidogrammitis drymoglossoides*. *Zhongcaoyao* 45(20), 2890-2894 [in Chinese].

Abstract: The chem. constituents from *Lepidogrammitis drymoglossoides* were studied. The constituents were isolated and purified by various column chromatogs., and their structures were identified on the basis of chem. evidence and spectroscopic analysis including MS, ¹H-NMR, and ¹³C-NMR. Fourteen compounds were isolated and identified as β-ecdysterone (1), physcion (2), emodin (3), umbelliferone (4), scoparone (5), aesculetin (6), caffeic acid (7), chlorogenic acid (8), protocatechuic acid (9), pyrocatechualdehyde (10), gallic acid (11), 4-hydroxybenzoic

acid Me ester (12), docosanyl tetracosanoate (13), and hexadecanoic acid (14). Compounds 3-5, 8, and 11-13 are isolated from the plants of genus *Lepidogrammitis* Ching for the first time.

Zhang M., Stout M.J. and Kubo I (1992) Isolation of ecdysteroids from *Vitex strickeri* using RLCC and recycling HPLC. *Phytochemistry* 31(1), 247-250.

Abstract: Six phytoecdysteroids were efficiently isolated from the root bark of *Vitex strickeri* using a combination of rotation locular countercurrent chromatography (RLCC) and recycling high performance liquid chromatography (R-HPLC). They were identified as 20-hydroxyecdysone, ajugasterone C, abutasterone, 1 la-hydroxyecdysone, 20-hydroxyecdysone-20,22-monoacetone and ajugasterone C-20,22-monoacetone by means of spectroscopic data. This is the first report of 1la-hydroxyecdysone and ajugasterone C-20,22-monoacetone isolated from a natural source.

Zhang M., Zhou Z-Y., Wang J., Cao Y., Chen X-Y., Zhang W-M., Lin L-D. and Tan J-W. (2012) Phytoecdysteroids from the roots of *Achyranthes bidentata* Blume. *Molecules* 17 3324-3332 (doi:10.3390/molecules17033324).

Abstract: Two new phytoecdysteroids, (25*S*)-20,22-*O*-(*R*-ethylidene)inokosterone (**1**) and 20,22-*O*-(*R*-3-methoxycarbonyl)propylidene-20-hydroxyecdysone (**2**), together with six known phytoecdysteroids **3-8** were isolated from the roots of *Achyranthes bidentata* Blume. The new structures were established on the basis of spectroscopic studies and chemical evidences. The absolute configuration at C-25 in the structure of known compound **3** was determined by chemical and spectroscopic means.

Zhang M.L., Huo C.H., Dong M., Lian C.H., Gu Y.C. and Shi Q.W. (2007) Non-taxoid chemical constituents from leaves of *Taxus mairei*. *Zhongguo Zhong Yao Za Zhi* 32(14), 1421-1425 [in Chinese].

Abstract: OBJECTIVE: To study the non-taxoids in the leaves of *Taxus mairei*. METHOD: The chemical constituents were isolated by chromatography and identified by spectral data. RESULT: Five compounds, taxamairin A (**1**), taxamairin B (**2**), sciadopitysin (**3**), (-) matairesinol (**4**), ponasterone A (**5**) were isolated and identified. CONCLUSION: Compounds 3-5 were isolated from this plant for the first time, compounds 1 and 2 were isolated from the leaves of *T. mairei* for the first time.

Zhang M-m., Zhao H-q., Zhou S-d., Wang D-j., Wang X., Liu D-c., Geng Y-l. and Mu Y-z. (2015) Content determination of β -ecdysterone and oleanolic acid in *Achyranthes bidentata* Blume by HPLC and their fingerprints. *Shandong Science* 28(5), 1-6 (DOI:10.3976/j.issn.1002-4026.2015.05.001) [in Chinese, with Abstract in English].

Abstract: An HPLC fingerprint map of the Chinese medicine ox-knee was established and the contents of chia fruit acid and β -moulting steroids were also measured. Using the Agilent ZORBAX SB-C18 column, the mobile phase is acetonitrile-0.6% formic acid, the column temperature is 25 degrees C, the detection wavelength is 260 nm, and the analysis of 13 batches of ox-knee herb is carried out. The HPLC fingerprint map of ox-knee herbs has 16 common peaks and 4 characteristic peaks, two of which are quantitatively measured β -moulting steroids and chitosin. Through the similarity analysis, it is concluded that there are obvious differences in quality between ox-knee herb in different regions. This research provides methods and technical support for the establishment of the quality control system of ox-knee herb.

Zhang Q., Ni G. and Yu D. (2009) Study on the chemical constituents of *Vitex canescens*. *Zhongguo Zhongyao Zazhi* 34(10), 1305-1306 [in Chinese].

Abstract: The objective of this paper was to isolate and identify the chem. constituents of *Vitex canescens*. The ethanol extract of *V. canescens* was isolated and purified by Sephadex LH-20, silica gel, ODS, and preparative HPLC. The chem. structures of constituents were identified by spectral anal. such as UV, IR, NMR, and MS. Six compounds were identified as 20-hydroxyecdysone (**1**), turkesterone, apiolin (**3**), (+)-lyoniresinol-3-*O*-*D*-glucopyranoside (**4**), (-)-lyoniresinol-3-*O*-*D*-glucopyranoside (**5**), and isochlorogenic acid. The compound 4, 5, and 6 were isolated from genus *Vitex* for the first time.

Zhang X., Xu H., Wang F., Yan Y-l., Wu J. and Song X-m. (2017) Chemical composition and anticancer activity of *Paris polyphylla*. *Central South Pharmacy* (9), 1237-1240 [in Chinese, with an English abstract].

Zhang X-J., Ma J., Zhu Z-Z. and Zhang H. (2012) Determination of ecdysterone in *Lepisorus marginatus* Ching by RP-HPLC. *Chinese Journal of Spectroscopy Laboratory* (3), 1834-1837 [in Chinese].

Abstract: Using the column Shimadzu C18 (250mm \times 4.6mm, 5 μ m), the mobile phase is methanol-water (49:51, V/V), the flow-rate is 0.8mL/min, the detection wavelength is 324nm, the column temperature is 30C, and the reverse-phase high-efficiency liquid spectrum is established to determine the content of moulting ketones in the sideway [?]. The results showed that moulting steroids had a good linear relationship in the range of 0.11-1.65 μ g (R²-0.9991), with an average recovery rate of 103.4% and RSD of 1.53%.

Zhang Y., Wu X., Li Y. and Wang G. (2014) Chemical constituents of *Paris polyphylla* var. *yunnanensis*. Journal of Jinan University (Natural Science and Medicine Edition) (1), 66-72 [in Chinese, with an English abstract].

Zhang Y., Yang L., Zhang Y-Y., Jiang H., Yang B-Y., Wang Q-H., Xiao H-B. and Kuang H-X. (2015) Determination of contents of split fractions of *Radix Achyranthis bidentatae*. Information on Traditional Chinese Medicine (5), 50-53 [in Chinese, with an English abstract].

Zhang Y-y. and Li J. (2017) Study on 7 chemical constituents in *Cyclea hypoglauca* (Schauer) Diels. by HPLC-ESI-MS/MS. China Journal of Traditional Chinese Medicine and Pharmacy (6), 2762-2764 [in Chinese].

Abstract: Objective: To establish a HPLC-ESI-MS/MS method for the quantitative analysis of 7 components in *Cyclea hypoglauca* (Schauer) Diels. Methods: The determination was performed on the Waters CORTECS C₁₈ column (2.1mm×100mm, 1.6μm) with the mobile phase consisting of acetonitrile-water (containing 0.1%formic acid) in gradient elution. Agilent mass spectrometry system equipped with an electro-spray ionization source was used as the detector and operated in the positive-ion mode. Multiple reaction monitoring (MRM) was performed to analyse *Cyclea hypoglauca* (Schauer) Diels. Results: magnoflorine, β-ecdysterone, cycleanine, columbamine, jateorhizine, palmatrubine and palmatine showed good linearity ($R^2>0.9997$). The RSD of the precision, repeatability and stability tests were less than 5%. The average recoveries were in the range of 96.7%-104.8%. Conclusion: The established method is accurate and sensitive, which can be used for the quality control of *Cyclea hypoglauca* (Schauer) Diels.

Zhang Y.H. and Wang H.Q. (2001) Ecdysteroids from *Rhaponticum uniflorum*. Pharmazie 56(10), 828-829.

Abstract: Five phytoecdysteroids were isolated from the roots of *Rhaponticum uniflorum*. They were identified as ecdysterone, ajugasterone C, ajugasterone C-20,22-monoacetone, ajugasterone C-2,3,20,22-diacetone and 5-deoxykaladasterone-20,22-monoacetone by means of spectroscopic data. This is the first report of ajugasterone C-2,3,20,22-diacetone and 5-deoxykaladasterone-20,22-monoacetone isolated from a natural source.

Zhang Y.H., Xin P., Lu Z.G. and Wang H.Q. (2001) A new ecdysteroid from *Rhaponticum uniflorum*. Chinese Chemical Letters 12(9), 797-798.

Zhang Z., Zhao D. and Deng J. (2011) Chemical constituents of *Paris bashanensis*. Zhongyaocai, 34(3), 389-392 [in Chinese].

Abstract: Chem. constituents of rhizomes of *Paris (P.) bashanensis* to search after alternative resource for Chinese medicinal material *Rhizoma Paridis* were studied. The n-BuOH extracts of *P. bashanensis* were applied to silica gel column and eluted with EtOAc-EtOH, then the gained fractions were further purified by chromatog. on Sephadex LH-20 column and Pre- RP-HPLC to give pure compounds whose structures were elucidated mainly on the basis of analyzing the spectral data of MS, ¹H-NMR, ¹³C-NMR and 2D-NMR. Five compounds were isolated and identified as β-ecdysone (1), pinnatasterone (2), pennogenin-3-O-α-L-rhamnopyranosyl(1→2)-[α-L-arabinofuranosyl(1→4)]-β-D-glycopyranoside (3), diosgenin-3-O-α-L-rhamnopyranosyl(1→2)-[α-L-arabinofuranosyl(1→4)]-β-D-glycopyranoside (4) and pennogenin-3-O-α-L-rhamnopyranosyl(1→4)-α-L-arabinofuranosyl(1→4)-[α-L-arabinofuranosyl(1→2)]-β-D-glycopyranoside (5). Compounds 1-5 are obtained from this plant for the first time.

Zhang Z-l., Zuo Y-m., Cai M-t. and Wang Y-y. (2013) Studies on chemical constituents in roots and rhizomes of *Trillium tschonoskii* (II). Zhongcaoyao (Chinese Traditional and Herbal Drugs) 44(20), 2808-2811 [in Chinese, with an English abstract].

Abstract: The chem. constituents in the roots and rhizomes of *Trillium tschonoskii* were extracted with 70% ethanol and separated by chromatog. on polyamide, silica gel, RP-C₁₈, and Sephadex LH-20 columns. Chem. structures were identified by MS, 1D and 2D NMR experiments Twelve compounds were isolated and identified from the Et acetate extract from the roots and rhizomes of *T. tschonoskii* and n-butanol fractions were identified as β-ecdysone (1), pinnatasterone (2), polypodine B (3), Me ferulorate (4), regalosite A (5), 4-hydroxybenzoic acid (6), vanillin (7), β-D-glucopyranosyl-(1→4)-O-[α-L-rhamnopyranosyl-(1→2)]-O-β-D-glucopyranoside (8), polyphyllin V (9), polyphyllin III (10), trillenoside A (11), and trillenoside C (12). Compounds 1, 2, 6, 7, 9 and 10 were isolated from the plants in this genus for the first time, and compounds 3-5, 11 and 12 were isolated from this plant for the first time. This was the second instalment of a multi-part article.

Zhang Z-l., Cai M-t., Zuo Y-m., and Wang Y-y. (2014) Studies on chemical constituents in the fruits of *Trillium tschonoskii* Maxim. Shizhen Guoyi Guoyao (Lishizhen Medicine and Materia Medica) 25(3), 541-543 [in Chinese, with an English abstract].

Abstract: The chem. constituents in the fruits of *Trillium tschonoskii* Maxim were investigated. Various column chromatog. methods were used in the isolation and purification, physiochem. constant determination and spectral

anal. were adopted to determine the chem. structures. Fourteen compounds had been isolated from the fruit of 70% methanol extract, the structure of isolated compounds were elucidated as follow: quercetin (1), quercetin-3-O-β-D-glucopyranoside (2), rutin (3), quercetin-3-O-α-L-rhamnopyranosyl-(1→2)-[α-L-rhamnopyranosyl(1→6)]-β-D-glucopyranoside (4), isorhamnetin-3-O-[2"-O-acetyl-α-L-arabinopyranosyl]-(1→6)-β-D-galactopyranoside (5), pennogenin-3-O-α-L-rhamnopyranosyl-(1→2)-β-D-glucopyranoside (6), pennogenin-3-O-α-L-rhamnopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→2)]-β-D-glucopyranoside (7), pennogenin-3-O-α-L-rhamnopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→2)]-β-D-glucopyranoside (8), β-ecdysone (9), trillenoside A (10), trillenoside C (11), trillenoside B(12), 3-O-feruloylsucrose (13), heronioside A (14). All these compounds are isolated from the fruits of *Trillium tschonoskii* Maxim firstly, compounds 1-5, 12-14 for the first time isolated from *Trillium tschonoskii* Maxim.

Zhang Z.Y., Yang W.Q., Fan C.L., Zhao H.N., Huang H.J., Wang Y. and Ye W.C. (2017) New ecdysteroid and ecdysteroid glycosides from the roots of *Serratula chinensis*. Journal of Asian Natural Product Research 18, 1-7 (DOI: 10.1080/102860020.2016.1209492). [Preprint].

Zhang Z.Y., Yang W.Q., Fan C.L., Zhao H.N., Huang H.J., Wang Y. and Ye W.C. (2017) New ecdysteroid and ecdysteroid glycosides from the roots of *Serratula chinensis*. Journal of Asian Natural Product Research 19, 208-214 (DOI: 10.1080/102860020.2016.1209492). [Final Version]

Zhao B., Chang Z.-z., Shi B. and Sun X.-d. (2011) HPLC simultaneous determination of contents of 5-hydroxymethylfurfural and ecdysterone in *Achyranthes bidentata* B1. Chinese Journal of Pharmaceutical Analysis (8), 1582-1585 [in Chinese].

Abstract: Objective: To establish an efficient liquid chromatography method of 5-hydroxymethylphenyl and moulting steroids in ox-knee herb. Methods: SHIMADZU shim-pack VP-ODS (4.6 mm×250 mm, 5 m) column, mobile phase: acetonitrile-water gradient wash- flow-rate: 1.0 mL.min⁻¹, column temperature: 30 degrees C, detection wavelength: 245nm and 279 nm. RESULTS: The content of 5-hydroxymethylphenidate and moulting ketones can be measured simultaneously at 2 wavelengths, the linear range of 5-hydroxymethylphenidate is 0.390 to 100 µg.mL⁻¹, and the linear range of moulting ketones is 3.125 to 100 mL⁻¹. The average recovery rates were 100.4 per cent and 99.0 per cent, respectively. Conclusion: This method is sensitive and accurate and provides a new content determination method for the quality control of the herbs of ox- knee.

Zhao B.T., Jeong S.Y., Moon D.C., Son K.H., Son K.H., Son J.K. and Woo M.H. (2012) High performance liquid chromatography used for quality control of *Achyranthis Radix*. Archives of Pharmaceutical Research 35(8), 1449-1455.

Abstract: To establish a standard of quality control and to identify reliable *Achyranthis Radix*, three phytoecdysones including ecdysterone (1), 25R-inokosterone (2) and 25S-inokosterone (3) were determined by quantitative HPLC/UV analysis. Three phytoecdysones were separated with an YMC J'sphere ODS C(18) column (250 mm × 4.6 mm, 4 µm) by isocratic elution using 0.1% formic acid in water and acetonitrile (85:15, v/v%) as the mobile phase. The flow rate was 1.0 mL/min and the UV detector wavelength was set at 245 nm. The standards were quantified by HPLC/UV from *Achyranthes bidentata* Blume and *Achyranthes japonica* Nakai, as well as *Cyathula capitata* Moq. and *Cyathula officinalis* Kuan, which are of a different genus but are comparative herbs. The method was successfully used in the analysis of *Achyranthis Radix* of different geographical origin or genera with relatively simple conditions and procedures, and the assay results were satisfactory for linearity, recovery, precision, accuracy, stability and robustness. The HPLC analytical method for pattern recognition analysis was validated by repeated analysis of eighteen *A. bidentata* Blume samples and ten *A. japonica* Nakai samples. The results indicate that the established HPLC/UV method is suitable for quantitation and pattern recognition analyses for quality evaluation of *Achyranthis Radix*.

Zhao H-li, Wang Y-z., Zhang L. and Hu H-l. (2017) Chemical constituents from *Achyranthes bidentata* Radix. Journal of Chinese Medicinal Materials (8), 1842-1845 [in Chinese].

Abstract: Objective: To study the chemical constituents from the *Achyranthes bidentata* Radix. Methods: The *A. bidentata* Radix were cut into pieces, and then extracted with 95% ethanol. The concentrated extracting solution was separated and purified by semipreparative HPLC, Sephadex LH-20, and silica gel column chromatography. Their structures were elucidated on the basis of spectral data and physicochemical properties. Results: Thirteen compounds were isolated. They were identified as β-ecdysterone (1), 25R-inokosterone (2), 25S-inokosterone (3), polypodine B (4), turkesterone (5), palythoalone B (6), uracil glucoside (7), methyl pyroglutamic acid (8), adenine (9), pedatisectine D (10), N-trans-feruloyltyramine (11), N-cis-feruloyltyramine (12), N-trans-feruloyl-3-methoxytyramine-4'-O-β-D-glucopyranoside (13). Conclusion: Compounds 5 ~ 10 are isolated from this plant for the first time.

Zhao M., Li Y.-m., Wang P.-f., Ju B.-y., Gao H.-m. and Wang Z.-m. (2018) Chemical constituents from the aerial parts of *Paris polyphylla* var. *chinensis*. Chinese Pharmaceutical Journal (16), 1342-1346 [in Chinese].

Abstract: OBJECTIVE: To study the chemical constituents from the aerial parts of *Paris polyphylla* var. *chinensis*. METHODS: The compounds were isolated and purified from the 75% ethanol extract by chromatography on HPD100 macroporous resin, silica gel, and Sephadex LH-20 as well as semi-preparative HPLC. Their structures were elucidated on the basis of spectral data. RESULTS: Eleven compounds were isolated and identified as corchionoside C (1), β -ecdysterone (2), coronasterone (3), kaempferol-3-O- β -D-galactopyranoside (4), astragalol (5), isorhamnetin-3-O- β -D-glucopyranoside (6), kaempferol-3-O- β -D-glucopyranosyl-(1 \rightarrow 2)- β -D-galactopyranoside (7), isorhamnetin-3-O- β -D-glucopyranosyl-(1 \rightarrow 2)- β -D-galactopyranoside (8), kaempferol-3-O- β -D-glucopyranosyl-(1 \rightarrow 2)- β -D-glucopyranoside (9), isorhamnetin-3-O- β -D-galactopyranosyl-(1 \rightarrow 6)- β -D-glucopyranoside (10), and isorhamnetin-3-O- β -D-gentiobioside (11). CONCLUSION: Compounds 1 and 3-11 are isolated from this plant for the first time and compounds 1,3-5 and 8-10 are isolated from *Paris* plants for the first time.

Zhao W.-t., Meng D.-l., Li X. and Li W. (2007) Chemical constituents of *Achyranthes bidentata* Bl. Journal of the Shenyang Pharmaceutical University (4), 207-210 [in Chinese].

Abstract: Objective: To study the chemical composition of the drug plant ox-knee.

Methods: The chemical composition in ox-knee of Chinese medicine was isolated by means of large-pore resin, silica column chromatography and preparative HPLC chromatography, and structural identification was carried out according to the physical and chemical properties of these compounds and the ¹H-NMR, ¹³C-NMR and other spectral data. Results: Five compounds were isolated from ox-knee, namely: stachysterone A (stachysterone A, 1), podocdysone C (2), β -moulting sterone (β -ecdysterone, 3), 25-R-ox-knee ketone (25R-inokosterone, 4), 25-S-ox-knee ketone (25S-inokosterone, 5). Conclusion: Compounds 1 and 2 are first isolated from ox-knee.

Zhao Y., Lu J., Chai R.-p., Lyu X.-k. and Chen X. (2019) Morphological and chemical differences between *Pfaffia glomerata* and *Pfaffia paniculata*. Modern Chinese Medicine (6), 758-763 [in Chinese].

Abstract: Objective: To investigate the morphological differences and contents differences of ecdysterone, total saponins and amino acids between *Pfaffia glomerata* and *P. paniculata*. Methods: The root microstructure of two medicinal herbs was observed by paraffin section. The contents of ecdysterone, total saponins in different extract parts, 15 hydrolyzed amino acids and total amino acids were determined by HPLC, UV and automatic amino acid analyzer, respectively. Result: There are significant differences in the microstructure of the roots of the two medicinal herbs. The content of ecdysterone in *P. glomerata* was higher than that in *P. paniculata*; the content of total saponins in water extract and 50% ethanol extract of *P. glomerata* was higher than that in *P. paniculata*; the content of total saponins in 75% ethanol extract was lower than that in *P. paniculata*; the content of amino acids and total amino acids in *P. paniculata* were higher than that in *P. glomerata*. Conclusion: There are obvious morphological differences between *P. glomerata* and *P. paniculata*. Both of them contain ecdysterone, saponin and amino acid, while the contents are significantly different, which provides a theoretical basis for the quality control and evaluation of *Pfaffia*.

Zhao Z., Gao W., Huang X., Zhao W. and Zhang Q. (2011) Chemical constituents in *Paris polyphylla* var. *pseudothibetica*. Zhongcaoyao 42(10), 1917-1920 [in Chinese].

Abstract: The chem. constituents in root and stem of *Paris polyphylla* var. *pseudothibetica* were studied. Silica gel chromatog., Sephadex LH-20 and chromatog. were used for isolation and purification, ¹H-NMR, ¹³C-NMR and ESI-MS were used for structure identification. Twelve compounds were isolated and identified as: diosgenin (1), diosgenin-3-O- α -L-arabinofuranosyl-(1 \rightarrow 4)[α -L-rhamnopyranosyl-(1 \rightarrow 2)]- β -D-glucopyranoside (2), pennogenin-3-O- α -L-arabinofuranosyl-(1 \rightarrow 4)- β -D-glucopyranoside (3), pennogenin-3-O- α -L-arabinofuranosyl-(1 \rightarrow 4)[α -L-rhamnopyranosyl-(1 \rightarrow 2)]- β -D-glucopyranoside (4), pennogenin-3-O- α -L-rhamnopyranosyl-(1 \rightarrow 4)- α -L-rhamnopyranosyl-(1 \rightarrow 4)-[α -L-rhamnopyranosyl-(1 \rightarrow 2)]- β -D-glucopyranoside (5), β -sitosterol (6), stigmaterol (7), daucosterol (8), stigmaterol-3-O- β -D-glucopyranoside (9), kaempferol (10), β -ecdysone (11) and sucrose (12). Compounds 1-9 were isolated from the plant for the first time.

Zheng M.S., Hwang N.K., Kim D.H., Moon T.C., Son J.K. and Chang H.W. (2008) Chemical constituents of *Melandrium firmum* Rohrbach and their anti-inflammatory activity. Archives of Pharmaceutical Research 31(3), 318-322.

Abstract: In our ongoing search for anti-inflammatory agents originating from Korean medicinal plants, we found that the hexane and BuOH fractions of the MeOH extract from the whole plants of *Melandrium firmum* Rohrbach inhibited 5-lipoxygenase (5-LOX) activity. By activity-guided fractionation, eleven compounds, alpha-spinaterol (1), ursolic acid (2), ergosterol peroxide (3), alpha-spinaterol glucoside (4), 2-methoxy-9-beta-D-ribofuranosyl purine (5), aristeromycin (6), ecdysterone (7), polypodaurein (8), (-)-bornesitol (9), mannitol (10) and cytoside (11) were isolated from the hexane and BuOH fractions using column chromatography. Compounds 2, 5, 6, 8, 9, 10 and 11

were isolated for the first time from this plant. Compounds 1, 3, 4 and 7 inhibited 5-LOX activity with IC₅₀ values of 21.04 microM, 42.30 microM, 32.82 microM, and 17.18 microM, respectively.

Zheng Y., Liu B., Chen M. and Chen T. (2008) Supercritical fluid extraction of ecdysterone from the roots of *Achyranthes bidentata* BL. *Journal of Separation Science* 31, DOI 10.1002/jssc.200700468.

Abstract: Ecdysterone has been found in a great many plants and animals and has some valuable pharmaceutical properties. The present study was conducted to investigate optimal conditions for the extraction of the compound by supercritical fluid extraction from the roots of *Achyranthes bidentata* BL. An orthogonal array design (OAD), OA(9)(3(4)), was employed as a chemometric method for optimization of the extraction of ecdysterone from the herbal medicine. Four parameters, namely, pressure and temperature of the supercritical fluid, the dynamic extraction time, and the flow rate of dimethyl sulfoxide, were studied and optimized by a three-level OAD. Determinations of the extracts were performed by high-performance liquid chromatography. The effects of the parameters were studied using analysis of variance. The results shown that the yield of ecdysterone could be influenced by the four parameters to a similar degree. The yield for DMSO-modified supercritical CO₂ was in the range from 0.65 to 1.03 mg/g under the selected conditions. In comparison with methanol-modified supercritical CO₂ and Soxhlet extraction, a higher yield was obtained when DMSO-modified supercritical CO₂ was used.

Zhou L. and Yang C. (1996) Root culture and β -ecdysone formation of *Cyanotis arachnoidea*. *Acta Botanica Yunnanica* 18(1), 99-104.

Abstract: The cultured roots have been induced from explants of stems, leaves and roots of *Cyanotis arachnoidea* using MS medium complemented with NAA and BAP. A liquid lift bioreactor with 2600 mL volume was tested for the root cultivation. It is noticed that farnesol and cholesterol have no obvious effect on the root growth. But β -ecdysone synthesis was stimulated by cholesterol with an appropriate concentration.

Zhou L., Cheng Z. and Chen D. (2012) Simultaneous determination of six steroidal saponins and one ecdysone in *Asparagus filicinus* using high performance liquid chromatography coupled with evaporative light scattering detection. *Acta Pharmaceutica Sinica B* 2(3) 267-273.

Abstract: A high-performance liquid chromatography coupled with an evaporative light scattering detector (HPLC-ELSD) has been developed to evaluate the quality of *Asparagus filicinus* through a simultaneous determination of six steroidal saponins and one ecdysone, including asparafiosides A, B, C, E, G, filiasparoside A and 20-hydroxyecdysone. With a C18 analytical column, the seven analytes were separated efficiently using acetonitrile-water as the mobile phase in a gradient program. The method limits of detection ranged 0.125-0.225 μ g, and the method limits of quantitation ranged 0.408-0.720 μ g, respectively. The intra- and inter-day precisions of the method were evaluated and were all less than 3%. All the recoveries for the spiked analytes ranged 95.16%-100.61%. The proposed method was successfully applied to quantify the seven components in thirteen samples from different localities in China

Zhou M-x., Chen S-f. And Zhu W-x. (2005) The study on the determination of phytoecdysone in *Folium Mori* by capillary electrophoresis. *Journal of Huaiyin Teachers' College (Natural Science Edition)* (4), 311-313 [in Chinese].

Abstract: A method for the determination of phytoecdysone in *Folium Mori* by capillary electrophoresis. The calibration curve of phytoecdysone showed good linearity in the range of 0.01-1.2 mg/mL with $r=0.9968$, and the RSD were of 6.5% for peak area. The method has the advantages of being simple convenient and sensitive. The method can be used for the routine analysis of phytoecdysone in *Folium Mori*.

Zhou R., Li B.-G. and Zhang G.-L. (2005) Chemical study on *Cyathula officinalis* Kuan. *Journal of Asian Natural Products Research* 7(3), 245-252.

Abstract: Four new compounds, 4-[(1-ethoxy-2-hydroxy)ethyl]phenol (**1**), 2,3-isopropylidene cyasterone (**2**), 24-hydroxycyasterone (**3**) and 2,3-isopropylidene isocyasterone (**4**), together with fourteen known compounds, have been isolated from the roots of *Cyathula officinalis* Kuan. Their structures have been elucidated predominantly by spectroscopic methods.

Zhu GD., Zhang YX., Je JY., Wu CL., Yu L. and Yang X. (2011) Effect of different size of ground *Cyanotis arachnoidea* particles on the release of β -ecdysone. *Hunan Academy of Agricultural Sciences, Changsa, China, Agricultural Sciences & Technology - Hunan* 12(9), 1318-1319 [in Chinese].

Abstract: [Objective] The aim was to investigate the influence of different size of ground *Cyanotis arachnoidea* C.B. Clarke particles on the release of β -ecdysone. [Method] The content of β -ecdysone extracted from *Cyanotis arachnoidea* powders of different sizes was detected through HPLC, under the chromatographic conditions: GRACE ODS C18 column (250 mm \times 4.6 mm, 5 μ m), mobile phase methanol:water = 40:60 (V/V, %), column temperature 26 $^{\circ}$ C, flow rate 1.0 ml/min and detection wavelength 248 nm. [Result] The extracted β -ecdysone from ultra-fine *Cyanotis arachnoidea* powder (particle size 10, 30 and 50 μ m) was as more than two fold as common powder (particle

size 180 μm). [Conclusion] The β -ecdysone in ultra-fine powder of *Cyanotis arachnoidea* was more easily soluble in methanol. The extraction rate of β -ecdysone was significantly higher than that of common powder. The result showed that the technology of ultra-fine pulverization markedly improved the dissolution of the β -ecdysone for increasing the cell breakage rate, and then the active ingredients could be extracted more easily.

Zhu GD., Zhang YX., Je JY., Wu CL., Yu L. and Yang X. (2011) Effect of different size of ground *Cyanotis arachnoidea* particles on the release of β -ecdysone. *Journal of Anhui Agricultural Sciences* (29), 17760-17761 [in Chinese]. [see previous entry]

Zhu N., Kikuzaki H., Vastano B.C., Nakatani N., Karwe M.V., Rosen R.T. and Ho C.-T. (2001) Ecdysteroids of quinoa seeds (*Chenopodium quinoa* Willd.). *Journal of Agricultural and Food Chemistry* 49, 2576-2578.
Abstract: Quinoa (*Chenopodium quinoa*) is a hardy and nutritious Latin American pseudo-cereal. Studies on the seeds led to the isolation of five ecdysteroids using column chromatography. Their structures were determined as ecdysterone, makisterone A, 24-*epi*-makisterone A, 24(28)-dehydromakisterone A, and 20,26-dihydroxyecdysone by spectroscopic methods. This study demonstrates that quinoa seeds are a source of ecdysteroids, which were reported to be molting hormones in insects.

Zhu T.T (also as Z-z.), Liang H., Zhao Y.Y. and Wang B. (2004) Isolation and structure identification of C-25 epimers of inokosterone from *Achyranthes bidentata* Blume. *Yao Xue Xue Bao (Acta Pharmaceutica Sinica)* 39(11), 913-916 [in Chinese].

Abstract: Purpose separation and determination of ox-knee plant (*Achyranthes bidentata* Blume.). The structure of the 25-bit differential isomer of steroids in the middle of the ox-knee. Methods: The 25-R/S-stereoisomers of purified ox-knee steroid was isolated by chromatography technology, and the structure was determined by spectral (IR, UV, MS, NMR) method and chemical method. Results: 3 compounds were isolated from the ethyl acetate extract of ox-knee, identified as 25S-ox-knee steroid (1; 25S-inokosterone), 25R-ox-knee steroid (2; 25R-inokosterone). Conclusion: compounds 1 and 2 were the first 25-stereoisomers isolated from ox-knee, the C-25 absolute configuration was determined for the first time, and the ^{13}C NMR data of the 25-R/S-isomers are identified and published.

Zhu W-M., Yang X-S., He H-P. and Hao X-J. (2000) Phytoecdysones from *Porana discifera*. *Yunnan Zhiwu Yanjiu (Acta Botanica Yunnanica)* 22(3), 351-357 [in Chinese, with an English abstract].

Abstract: Ten phytoecdysones were isolated from the aerial parts of *Porana discifera*. They had no anti-inflammatory, analgesic, sedative, anticonvulsant or anticerebral hypoxic activity in animal testing.

Zhu X-d. and An Y-l. (2003) A study on the technology for extracting pure β -ecdysone from *Cyanotis arachnoidea*. *Journal of Southwest Forestry College* vol? Pages?

Zia-Ul-Haq M., Riaz M. and De Feo V. (2012) *Ipomea hederacea* Jacq.: a medicinal herb with promising health benefits. *Molecules* 17, 13132-13145 (doi: 10.3390/molecules171113132).

Abstract: *Ipomea hederacea* Jacq. (kaladana or ivy leaf morning-glory), a member of the family *Convolvulaceae*, is used primarily for its seeds and recognized for its medicinal properties, especially in Asian countries. This medicinal herb contains various valuable chemical constituents such as ecdysteroids, steroidal glycosides, aromatic acids, triterpenes, amino acids, organic acids, mineral elements and vitamins. A number of pharmacological properties such as diuretic, anthelmintic, blood purifier, deobstruent, laxative, carminative and anti-inflammatory actions have been ascribed to this plant, besides its use to treat abdominal diseases, fevers, headache and bronchitis. This review focuses on compositional, medicinal and therapeutic properties of this plant, as a potential sources of bioactive molecules for medicinal and nutraceutical applications.

Zibareva L.N. (1995) Study of peculiarities to accumulate ecdysteroids in plants of the *Silene* L. genus. In: Abstracts of the International Conference "Fundamental and Applied Problems of Environmental Protection", Vol. 2 p.97

Zibareva L.N. (1997) The prognosis of the presence of ecdysteroids among the species *Silene* L. and *Chenopodium* L. to their contents in the seeds. *Rastitelny Resursy* 33(1) 89-92 [in Russian, with an English abstract].

Zibareva L.N. (1999) Occurrence of phytoecdysteroids in *Silene* L. genus and dynamics of their contents" *Rastitelny Resursy* 35(1), 79-87 [in Russian, with an English abstract].

Zibareva L. (2000) Distribution and levels of phytoecdysteroids in plants of the genus *Silene* during development. *Archives of Insect Biochemistry and Physiology* 43(1) 1-8.

Abstract: The purpose of the present work is the study of ecdysteroid distribution in annual and perennial *Silene* species during development. The experimental approach included the measurement of ecdysteroid levels in different plant organs and an evaluation of the contribution of individual organs to the total amount of 20-hydroxyecdysone (20E) produced by the plant. The highest concentrations of 20E were observed in reproductive organs. High levels were also found in leaves with lower levels in stems. Maximal ecdysteroid content for aerial parts was observed during periods of intense growth: at budding or flowering in annual species, and during vegetative growth or budding in perennial species. The contribution of the different organs to the overall ecdysteroid content changes during plant development. Leaves represent the main part of plant mass and 20E content. Reproductive organs represent a relatively small mass, but they contain high concentrations of ecdysteroids and, in terms of the amount of 20E they contain, their contribution is equal to that of stems.

Zibareva L.N. and Sviridova T.P. (1989) The dynamics of 20-hydroxyecdysone content in *Lychnis chalconica* L. grown in the Siberian Botanical Garden (Tomsk). *Rastitelny Resursy* 25(4), 561-564 [in Russian].

Zibareva L.N., Yeryomina V.I. and Zibarev P.V. (1997) The method of detection and quantity definition of phytoecdysteroids in plant objects. Patent (Russia). N° 2082168 [in Russian]

Zibareva L.N., Baltaev U.A., Revina T.A. and Abubakirov N.K. (1991a) Phytoecdysteroids in plants of the genus *Lychnis*. *Khimiya Prirodnikh Soedinenii* (4), 584-585 [in Russian].
No Abstract available

Zibareva L.N., Saatov Z. and Abubakirov N.K. (1991b) Stachysterone D, viticosterone E and ecdysone from *Lychnis chalconica*. *Khimiya Prirodnikh Soedinenii* (4), 585-586 [in Russian]/*Natural Product Chemistry* (1992) 514-515 [in English].
No Abstract available

Zibareva L.N., Baltaev U.A., Sviridova T.P., Saatov Z and Abubakirov N.K. (1995) Species of the genus *Lychnis* L. - potential sources of ecdysteroids. *Rastitelny Resursy* 31(4), 1-8 [in Russian].

Zibareva L.N. and Yeryomina V.I. (1996) Dynamics of the contents of ecdysteroids in species of the genus *Silene* L. grown in the Siberian botanical garden (Tomsk city). *Rastitelny Resursy* 32(1/2), 106-110 [in Russian, with an English abstract].

Zibareva L.N., Yeryomina V.I. and Ivanova N.A. (1997) New ecdysteroidiferous species of the genus *Silene* L and the dynamics of ecdysteroid contents in them. *Rastitelny Resursy* 33(3), 73-76 [in Russian, with an English abstract].

Zibareva L., Volodin V., Saatov Z., Savchenko T., Whiting P., Lafont R. and Dinan L. (2003) Distribution of phytoecdysteroids in the Caryophyllaceae. *Phytochemistry* 64, 499-517.

Abstract: Certain genera within the Caryophyllaceae (especially *Silene* and *Lychnis*) have received a significant amount of attention with regard to the isolation and identification of ecdysteroids. However, the taxonomy of this family is difficult. Hence, the occurrence of phytoecdysteroids in members of the Caryophyllaceae is presented, and combined with new data on ecdysteroid agonist (phytoecdysteroid) and antagonist activities, in order to survey the distribution of phytoecdysteroid-containing species within this large family, and to assess the utility of phytoecdysteroids as chemotaxonomic markers. The new data presented (representing ca. 110 species) have been obtained by the application of sensitive biological/biochemical methods for the detection of ecdysteroid agonists and antagonists, using *Drosophila melanogaster* B_{II} bioassay and ecdysteroid-specific immunoassays. In the antagonist version of the B_{II} bioassay, only weak ecdysteroid antagonist activities were detected in a few of the extracts. From both new and previously available data, it was found that phytoecdysteroids were present predominantly in the Genera *Lychnis*, *Petrocoptis*, *Sagina* and *Silene*. Comparison of ecdysteroid occurrence with a molecular phylogeny for the tribe *Sileneae* [Taxon 44 (1995) 525] revealed close association of ecdysteroid occurrence with certain groups of this tribe. In 14 species of *Silene* examined, there is a reasonable, but not absolute, relationship between the presence of ecdysteroids in the seeds and in other plant parts. Where ecdysteroids are present in the plant, highest concentrations are generally present in the roots.

The distribution of ecdysteroids in members of the Caryophyllaceae is surveyed and their value as chemotaxonomic markers assessed.

Zibareva L.N., Dinan L. and Yeryomina V.I. (2007) Screening of Caryophyllaceae species for phytoecdysteroid presence. *Rastitelnye Resursy* 43(4), 66-75 (2007a) [in Russian].

Zibareva L.N., Lafont R. and Dinan L. "TITLE" in: *Actual Ecology Problems of Siberia in a Global Context* (Ed. Kirlotina S.I.), Tomsk University Press, pp. 132-135 (2007b) [in Russian].

Zibareva L.N., Yeriomina V.I., Munkhjargal N., Girault J.-P., Dinan L. and Lafont R. (2009) The phytoecdysteroid profiles of 7 species of *Silene* (Caryophyllaceae). *Archives of Insect Biochemistry and Physiology* **72**(4) 234-248. **Abstract:** The phytoecdysteroid profiles of extracts of aerial parts of flowering plants of 7 ecdysteroid-containing species in the genus *Silene* (Caryophyllaceae; *S. fridvaldszkyana* Hampe, *S. gigantea* L., *S. graminifolia* Otth, *S. mellifera* Boiss. & Reuter, *S. repens* Patr., *S. schmuckeri* Wettst., and *S. sendtneri* Boiss.) have been examined and identified by HPLC and, in the case of two new compounds, by mass spectrometry and NMR. *S. fridvaldszkyana* was found to contain predominantly 20-hydroxyecdysone (20E), with smaller amounts of 2-deoxyecdysone (2dE), 2-deoxy-20-hydroxyecdysone (2d20E), polypodine B (polB), integristerone A (IntA), 26-hydroxypolypodine B (26polB), and 20,26-dihydroxyecdysone (20,26E). Additionally, a new minor ecdysteroid, 26-hydroxyintegristerone A, has been identified from this species. *S. gigantea* contains 3 major ecdysteroids (2dE, 2d20E, and 20E) and much smaller amounts of IntA and 2-deoxy-20-hydroxyecdysone 25-beta-D-glucoside, which is a new ecdysteroid. Ecdysteroids in the other 5 species have been identified by co-chromatography with reference compounds on RP- and NP-HPLC systems. There is considerable variability with regard to ecdysteroid profiles within the genus *Silene*. The chemotaxonomic value of ecdysteroid profiles within the genus *Silene* is discussed.

Zibareva L.N., Seliverstova A.A., Suksamrarn A., Morozov S.V. and Chernyak E.I. (2014) Phytoecdysteroids from the aerial part of *Silene colpophylla*. *Chemistry of Natural Compounds* **50**(3) 571-572. The genus *Silene* L. includes about 700 species according to current data [1] and represents the richest source of ecdysteroids. Greater than 100 species of this genus have now been found to produce ecdysteroids [2]. Greater than 80 of the 476 identified ecdysteroids are synthesized by *Silene* species [3, 4]. The goal of the present work was to study the chemical composition of ecdysteroids from the aerial part of *S. colpophylla* Wrigley, which we recommend for the first time as a source of ecdysteroids. Ecdysteroids were isolated from the aerial part of *S. colpophylla* collected during flowering in 2009–2012. Starting extracts were obtained by exhaustive extraction of ground air-dried raw material by EtOH (70%). Lipophilic substances were removed by hexane. Subsequent multiple extraction by n-BuOH extracted terpenoid and phenolic compounds. The concentrated residual was fractionated successively using CHCl₃-EtOH (9:1), EtOH (70%), and H₂O. The CHCl₃-EtOH fraction of the BuOH extract was sep ...

Zibareva L., Athipornchai A., Wonganan O. and Suksamrarn A. (2017) Application of ultrasound to extraction of biologically active substances of some *Serratula* species. *International Journal of Food and Biosystems Engineering* **5**(1), 31-37.

Zimmer A.R., Bruxel F. Bassani V.L. and Gosman G. (2006) HPLC method for the determination of ecdysterone in extractive solution from *Pfaffia glomerata*. *Journal of Pharmaceutical and Biomedical Analysis* **40**, 450-453. **Abstract:** A RP-LC method was developed and validated to quantify ecdysterone in extractive solution from subterranean parts of *Pfaffia glomerata*. The analysis was performed using a RP-18 column with acetonitrile:water isocratic elution and the detection was carried out by UV at 242 nm. The standard curve for ecdysterone was linear over the range of 5.2-41.6 microg/ml (R²=0.9995). The extractive solution showed linear response in the range of 25.05-175.35 microg/ml (R²=0.9977). This method showed excellent repeatability (relative standard deviation, R.S.D.<2.0%), intermediary precision (R.S.D.=2.13%) and accuracy (101.04; R.S.D.=1.51%). The limit of detection (LOD) was 0.036 microg/ml and the limit of quantification (LOQ) was 0.110 microg/ml, demonstrating the sensitivity of the method. This assay can be readily utilized as quality controlled method for *P. glomerata* preparations.

Zughdani M., Yusufoglu H.S., Ekiz G. and Linden A. (2020) Ecdysteroids from the underground parts of *Rhaponticum acaule* (L.) DC. *Phytochemistry* **180**, 112530. **Abstract:** In addition to two known ecdysteroids, 20-hydroxyecdysone and turkesterone, three previously undescribed stigmastane-type ecdysteroids were isolated from the underground parts of *Rhaponticum acaule* (L.) DC. by chromatographic techniques (CC, VLC, MPLC). The structures of the compounds were established by chemical (acetylation) and spectroscopic methods including UV, IR, HRMS, 1D-NMR: ¹H-NMR, ¹³C-NMR, DEPT-135. and 2D-NMR: COSY, NOESY, HSQC, HMBC. Two compounds were isolated as an isomeric mixture and each of them was purified and converted to the corresponding acetylated derivative. Based on all of the evidence, the structures of three undescribed stigmastane-type ecdysteroids were established as 2β,3β,11α,20β,22α,24,28-heptahydroxy-6-oxo-stigmast-7-en-25,29-lactone and the cyclic 22,29-hemiacetals 22*R* and 22*S* stigmast-7-en-29-al,2β,3β,11α,20α,22,28-hexahydroxy-6-oxo, and the trivial names acaulesterone and rhapocasterones A and B are suggested, respectively. The structures and absolute configurations of 20-hydroxyecdysone and cyclic-22,29-hemiacetal-22*R*-stigmast-7-en-29-al,2β,3β,11α,20α,22,28-hexahydroxy-6-oxo were confirmed by X-ray crystal-structure analyses of their acetyl derivatives.

Zuo JY. (2006) Isolation and structural modification of 20-hydroxyecdysone. Master's Thesis, Shenyang Pharmaceutical University, Shenyang, China.

1c. Associated References/Reviews

Abat J.K., Kumar S. and Mohanty A. (2017) Ethnomedicinal, phytochemical and ethnopharmacological aspects of four medicinal plants of Malvaceae used in Indian traditional medicines: a review. *Medicines* 4, 75 (doi: 10.3390/medicines-4040075).

Abstract: The ethnomedicinal values of plants form the basis of the herbal drug industry. India has contributed its knowledge of traditional system medicines (Ayurveda and Siddha) to develop herbal medicines with negligible side effects. The World Health Organization has also recognized the benefits of drugs developed from natural products. *Abutilon indicum*, *Hibiscus sabdariffa*, *Sida acuta* and *Sida rhombifolia* are ethnomedicinal plants of Malvaceae, commonly used in Indian traditional system of medicines. Traditionally these plants were used in the form of extracts/powder/paste by tribal populations of India for treating common ailments like cough and cold, fever, stomach, kidney and liver disorders, pains, inflammations, wounds, etc. The present review is an overview of phytochemistry and ethnopharmacological studies that support many of the traditional ethnomedicinal uses of these plants. Many phytoconstituents have been isolated from the four ethnomedicinal plants and some of them have shown pharmacological activities that have been demonstrated by *in vivo* and/or *in vitro* experiments. Ethnomedicinal uses, supported by scientific evidences is essential for ensuring safe and effective utilization of herbal medicines.

Abdillahi H.S., Stafford G.I., Finnie J.F. and Van Staden J. (2010) Ethnobotany, phytochemistry and pharmacology of *Podocarpus sensu latissimo* (s.l.). *South African Journal of Botany* 76, 1-24.

Abstract: The genus *Podocarpus sensu latissimo* (s.l.) was initially subdivided into eight sections. However, based on new information from different morphological and anatomical studies, these sections were recognised as new genera. This change in nomenclature sometimes is problematic when consulting ethnobotanical data especially when selecting plants for pharmacological screening, thus there is a need to clear any ambiguity with the nomenclature. Species of *Podocarpus* s.l. are important timber trees in their native areas. They have been used by many communities in traditional medicine and as a source of income. *Podocarpus* s.l. is used in the treatment of fevers, asthma, coughs, cholera, distemper, chest complaints and venereal diseases. Other uses include timber, food, wax, tannin and as ornamental trees. Although extensive research has been carried out on species of *Podocarpus* s.l. over the last decade, relatively little is known about the African species compared to those of New Zealand, Australia, China and Japan. Phytochemical studies have led to the isolation and elucidation of various terpenoids and nor- and bis-norditerpenoid dilactones. Biflavonoids of the amentoflavone and hinokiflavone types have also been isolated. Nor- and bis-norditerpenes are said to be taxonomic markers for this genus. Recent *in vitro* and *in vivo* studies have shown antitumor, antimicrobial, anti-inflammatory, antioxidant, larvicidal, plant and insect growth regulation activities. Various studies have yielded important natural bioactive products and two of them are worth mentioning. Taxol, a significant anticancer agent has been isolated from *Podocarpus gracilior* and totarol, a diterpenoid isolated from various species and now commercially produced as a potent antibacterial and antioxidant agent. Findings from this review supports the use of an ethnobotanical and chemotaxonomical approach in selecting plants for pharmacological screening since most of the species in the different morphological groups have similar uses. Also the isolated compounds have chemotaxonomic value amongst the groups. Some of the biological activities identified from extracts and compounds isolated from *Podocarpus* s.l. support the rationale behind the medicinal uses of these species.

Abubakirov N.K. (1975) Ecdysteroids: what is their use? *Chimia i Zhyzn* (11). 57-62 [in Russian].

Abubakirov N.K. (1980) New phytoecdysone. In: *Frontiers of Bioorganic Chemistry and Molecular Biology* (Ed: Ananchenko S.N.), Pergamon Press, pp 257-259.

Abubakirov NK (1981) Ecdysteroids of flowering plants (Angiospermae). *Khimiya Prirodnykh Soedinenii* 685-702 [in Russian]/*Chemistry of Natural Products* 489-503 (1982) [in English].

Abstract: A review is given of the ecdysone-like substances found in recent years in flowering plants. The structures of new compounds are presented. The role of molting hormones in the ecological interrelationships between plants and insects is discussed.

Adler J.H. and Grebenok R.J. (1997) Occurrence, biosynthesis and putative role of ecdysteroids. In: *Biochemistry and function of sterols*, 1st Edition. CRC Press (ISBN: 9781003068754), pp. 12.

The biosynthesis of ecdysteroids in plants and their distribution within the plant is well understood. The function of this review is to bring together the limited information available on these processes, and to attempt

a synthesis and an integration of our knowledge of plant biosynthetic capabilities with the assumed function of these phytoecdysteroids as defense compounds against insect herbivory. The distribution of phytoecdysteroids within an individual plant is related to the organ type and position of organ on the plant as well as the state of development of the organ. The presence of phytoecdysteroids within a specific organ may fluctuate over the time course of growth, depending upon cycles of biosynthesis and transport. Few analyses of the fluctuation of ecdysteroids between plant parts were reported in the early period of phytoecdysteroid discovery, since the major emphasis was on isolation and structural characterization.

Adler J.H. and Grebenok R.J. (1999) Occurrence, biosynthesis, and putative role of ecdysteroids in plants. *Critical Reviews in Biochemistry and Molecular Biology* **34**(4), 253-264.
No Abstract.

Adler J.H. and Salt T.A. (1987) Phytosterol structure and composition in the chemosystematics of the Caryophyllales. In: *The Metabolism, Structure, and Function of Plant Lipids* (Eds. Stumpf P.K., Mudd J.B. and Nes W.D.) Plenum Publishing Corp., pp. 119-121.

Abstract: Angiosperms synthesize a diverse array of 4-desmethylsterols which vary primarily in position and number of double bonds as well as stereochemistry at C-24^{1,2}. The high degree of structural chemistry^{1,2} associated with sterols, their essentiality for proper eucaryotic cell function^{3,4,5}, and their ubiquitous distribution in the plant kingdom^{1,2} make phytosterols potentially useful molecules for the characterization of taxonomically related organisms. Most angiosperms produce 24 α -alkyl- Δ^5 -sterols with relatively few species producing 24-alkyl- Δ^7 -sterols as dominant sterols^{1,2}.

Ahmed B., Masoodi M.H., Khan S., Habibullah (2008) *Lychnis coronaria* Linn. A review. *Natural Products* **4**(1), 22-25.

Abstract: *Lychnis coronaria* has been used for centuries for curing various ailments in the form of extracts and has been reported to possess potent pharmacological actions against leprosy, diarrhoea, lungs, liver and also as a remedy for beri-beri etc. Different chemical constituents from the plant such as ecdysterone 22-O- β -D-glucopyranoside, stigmast-5-ene-3-one, taraxerol, α -tocopherol and dehydrodiconiferyl alcohol-4-O- β -D-glucopyranoside, epoxyactinidionoside and many others have been isolated from the plant. The present article reviews the pharmacological and phytochemical work done on the plant.

Al-Easa H.S., Rizk A.M. and Abdel-Bari E.M. (2003) Chemical constituents and nutritive values of range plants in Qatar. Scientific and Applied Research Center, University of Qatar, Doha-Qatar, pp. 386.

Al Naggar Y., Ghorab M. and Mohamed K. (2017) Phytoecdysteroids: isolation and biological applications. *American Journal of Life Sciences* **5**(1), 7-10.

Abstract: Phytoecdysteroids are analogues of arthropod steroid hormones found in plants, where they deter predation by non-adapted predators. Their discovery in several plant species displayed a wide array of rather beneficial agricultural impact. An overview is given on both well-known and recently discovered phytoecdysteroids including a sophisticated isolation scheme and notable physiological and pharmacological effects of ecdysteroids on vertebrates.

Aminah N.S., Laili E.R., Rafi M., Rochman A., Insanu M. and tun K.N.W. (2021) Secondary metabolite compounds from *Sida* genus and their bioactivity. *Heliyon* **7**, article e06682.

Abstract: Plants are the key source for the production of novel therapeutic products for new medicines. The biological properties of the plant species used world wide are mainly accountable for their secondary metabolites obtained from plants. The goal of this analysis is to summarize the chemical composition and biological effects of the genus *Sida* (Malvaceae) to identify potential research opportunities. This analysis draws on the literature review of scientific journals, and books from libraries, and electronic sources like ScienceDirect, Springer, PubMed, ResearchGate, Google Scholar, and the Website. Some groups of secondary metabolite compounds isolated from the genus *Sida* include alkaloids, flavonoids, coumarin, and others. Pharmacological experiments found that there are a wide variety of biological activities in extracts and compounds isolated from the genus *Sida* comprising antimalarial, antiplasmodial, antimicrobial, analgesic, antibacterial, antioxidant, vasorelaxant, wound healing, antifungal activities, the inhibition of quinone reductase, and mouse mammary organ culture.

Arfan M., Khan G.A. and Ahmad N. (1996) Steroids and terpenoids of the genus *Ajuga*. *Journal of the Chemical Society of Pakistan* **18**(2), 170-174.

Bajguz A. (2000) Ecdysteroids in plants. *Kosmos (Krakow)* **49**(1/2), 169-178 [in Polish].

Bajguz A., Bakała I. and Talarek M. (2015) Chapter 5 - Ecdysteroids in Plants and their Pharmacological Effects in Vertebrates and Humans. In: *Studies in Natural Products Chemistry*, Vol. 45 (Editor: Atta-ur-Rahman), Elsevier, pp. 121-145.

Abstract: Ecdysteroids, the molting hormones of arthropods, have been found in many plant species.

Phytoecdysteroids belong to a large family of ecdysteroids that comprises more than 250 representatives. To date, ecdysteroids or ecdysteroid-like compounds have been found in gymnosperms, angiosperms, fungi, algae, and certain marine organisms. Ecdysteroids accumulate in various plant organs, including fruits, seeds, flowers, anthers, leaves, and roots, during various phases of their growth. Ecdysteroid concentrations in plants may be very large, i.e., several orders of magnitude higher than in most arthropods. The chemical structure of phytoecdysteroids is very varied. Plants may contain C27, C28, or C29 type of ecdysteroids either in the free form or as various polar or apolar conjugates. They may also contain C24, C21, or C19 compounds resulting from various side chain cleavage reactions. The presence of ecdysteroids and their conjugates in plants is suggested to be a factor which may alter development of nonphytoecdysteroid-adapted insect species. Phytoecdysteroids induce abnormal molting in many arthropods with lethal effect. Hence these compounds may potentially prove useful to control insect herbivory on agriculturally important plants. However, many functions of ecdysteroids in plants are still unknown. At present, our knowledge of the effects of phytoecdysteroids in animals or human is still rather fragmentary. Phytoecdysteroids from *Ajuga decumbens* showed strong inhibitory effect on early induction and potent antitumor-promoting activities of Epstein–Barr virus on a mouse skin. It suggests that *A. decumbens* may be valuable as a source of potential cancer chemopreventive agent. Phytoecdysteroids have an antifungal and antibacterial activity. They are apparently nontoxic to mammal ecdysteroids. A very low toxicity (LD50>6g/kg) of ecdysteroids in vertebrates is observed. The wide range of beneficial pharmacological (adaptogenic, anabolic, antidiabetic, hepatoprotective, immunoprotective, wound-healing, anti-inflammatory, and perhaps even antitumor) activities is claimed for ecdysteroids. In particular, this has led to a large (and unregulated) market for ecdysteroid-containing preparations for bodybuilders, sportsmen, and pets, among others. Ecdysteroids are also being considered as nutraceutical additives to food products. They have antioxidative and antifree-radical properties.

Baltaev U.A. (2000) Phytoecdysteroids: structure, sources and biosynthesis in plants. *Russian Journal of Bioorganic Chemistry* 26(12), 799-831 [in English]/ *Bioorganicheskaya Khimiya* 26(12), 892-925 [in Russian].

Abstract: Data on the content of ecdysteroids in plant sources are given and the ecdysteroid biosynthesis and role in plants are discussed.

Barboza G.E., Cantero J.J., Nunez C., Pacciaroni A. and Espinar L.A. (2009) Medicinal plants: a general review and a phytochemical and ethnopharmacological screening of the native Argentine flora. *Kurtziana* 34(1/2), 7-365.

Abstract: A review and a checklist based on empirical evidence of the therapeutic properties of the native medicinal flora from Argentina are presented. The chemical constituents and biological activity of each species, when known, are also provided. Medicinal flora comprises 1,529 taxa of vascular plants (Pteridophyta: 56; Gymnospermae: 13; Monocotyledoneae: 152; Dicotyledoneae: 1,308), including 115 endemic species. In addition, the distribution of these species is analyzed, and the endemic areas are also stated.

Báthori M. (2002) Phytoecdysteroids effects on mammals, isolation and analysis. *Mini Reviews in Medicinal Chemistry* 2, 285-293.

Abstract: Ecdysteroids are known insect moulting hormones, regulating the insects' metamorphosis. At the same time, ecdysteroids reveal beneficial effects on humans and animals alike. Medicinal plants have been subjected to an intensive research, addressing the presence of ecdysteroids. The possible utilization of medicinal plant deals with their use as raw materials for health preparations and also for the isolation of new phytoecdysteroids. Research on the plant ecdysteroids involves two basic lines. Isolation of major compounds and scout their physiological effects; and isolation of minor ecdysteroids to find new compounds. This review summarizes the recent efforts in the ecdysteroid research including their indication as health improvement preparations, chromatography of ecdysteroids and certain methods for identification of new ecdysteroids

Báthori M., Kálmán A., Argay G. and Kalász H. (2000) The analysis and crystallographic characterization of 20-hydroxyecdysone. *Current Medicinal Chemistry* 7, 1305-1312.

Abstract: 20-Hydroxyecdysone (20E) is an insect molting hormone that is also widely spread in various plants. Many chromatographic methods can be used to identify and/or determine 20E content in samples of biological origin and various spectroscopic methods serve to identify its structural elements. We have utilized X-ray crystallography to reveal the stereostructures of 20E. Our data demonstrates that 20E exists in two different crystalline forms that are both orthorhombic modifications. One form is homo-molecular, with a limited freedom of internal rotation of the side chain around the C23-C24 bond and the other, which is a clathrate formed with methanol and water, which minimize the conformational freedom of the side chain.

Báthori M. and Pongrácz Z. (2005) Phytoecdysteroids - from isolation to their effects on humans. *Current Medicinal Chemistry* 12(2), 153-172.

Abstract: An overview is given on both well-known and recently discovered phytoecdysteroids including a sophisticated isolation scheme and notable physiological and pharmacological effects of ecdysteroids on vertebrates. The isolation of pure ecdysteroids has been improved by the use of low-pressure reversed-phase chromatography. An optimized combination of preliminary purification and chromatographic separations results in pure ecdysteroids. Structural elucidation has been done using spectroscopic methods, however, the final proof of the steric structure is rendered using x-ray crystallography. Ecdysteroid containing preparations show a boom and both OTC products and numerous preparation techniques can be found using the Internet. This paper will give a review on the kaleidoscope of pharmacological effects attributed to the ecdysteroids, such as: An increase of protein synthesis (for body-building, AIDS, patients with neoplasm disease, etc.), and other body functions; Antidepressant effect; Shielding the body from stress, and improve the physical and sexual performance; Prevention from infections and certain diseases. A list of recent offers of ecdysteroid-containing products will also be given. The perspective use of ecdysteroids is promising in genetics. Steroid regulation of programmed cell death during development and differentiation has recently come to the limelight. Murine model of human diseases and its influencing with ecdysteroids are detailed.

Batista D.S., Koehler A.D., Romanel E., de Souza V.C., Silva T.D., Almeida M.C., Maciel T.E.F., Ferreira P.R.B., Felipe S.H.S., Saldanha C.W., Maldaner J., Dias L.L.C., Festucci-Buselli R.A. and Otoni W.C. (2018) De novo assembly and transcriptome of *Pfaffia glomerata* uncovers the role of photoautotrophy and the P450 family genes in 20-hydroxyecdysone production. *Protoplasma* (doi: 10.1007/s00709-018-1322-1).

Abstract: *Pfaffia glomerata* is a medically important species because it produces the phytoecdysteroid 20-hydroxyecdysone (20-E). However, there has been no ready-to-use transcriptome data available in the literature for this plant. Here, we present de novo transcriptome sequencing of RNA from *P. glomerata* in order to investigate the 20-E production as well as to understand the biochemical pathway of secondary metabolites in this non-model species. We then analyze the effect of photoautotrophy on the production of 20-E genes phylogenetically identified followed by expression analysis. For this, total messenger RNA (mRNA) from leaves, stems, roots, and flowers was used to construct indexed mRNA libraries. Based on the similarity searches against plant non-redundant protein database, gene ontology, and eukaryotic orthologous groups, 164,439 transcripts were annotated. In addition, the effect of photoautotrophy in two genes putatively involved in the 20-E synthesis pathway was analyzed. The Phantom gene (CYP76C), a precursor of the route, showed increased expression in *P. glomerata* plants cultured under photoautotrophic conditions. This was accompanied by increased production of this metabolite indicating a putative involvement in 20-E synthesis. This work reveals that several genes in the *P. glomerata* transcriptome are related to secondary metabolism and stresses, that genes of the P450 family participate in the 20-E biosynthesis route, and that plants cultured under photoautotrophic conditions promote an upregulated Phantom gene and enhance the productivity of 20-E. The data will be used for future investigations of the 20-E synthesis pathway in *P. glomerata* while offering a better understanding of the metabolism of the species.

Borin M.R. de M.B and Gottlieb O.R. (1993) Steroids, taxonomic markers? *Plant Systematics and Evolution* 184, 41-76.

Abstract: The use of steroids as taxonomic markers of the angiosperms was evaluated. To this end evolutionary advancement parameters, based on oxidation state and skeletal specialization of each steroid were calculated. The averages of these values for the steroids registered for a particular plant group were assumed to represent the evolutionary parameters of the plant group. Positive correlations of these chemical parameters and of morphological Sporne indices, as well as of herbacity indices, were observed to constitute a general trend. Furthermore, steroids are not uniformly distributed within a morphologically homogeneous plant group, but are replacement characters. Thus, in spite of their functional importance, steroids can be considered, analogously to many other classes of secondary metabolites, for chemosystematic purposes.

Camps F. and Coll J. (1993) Insect allelochemicals from *Ajuga* plants. *Phytochemistry* 32(6), 1361-1370.

Abstract: Clerodane diterpenoids and phytoecdysteroids with potential insect antifeedant and moulting hormone activities, respectively, have been isolated from *Ajuga* plants. Some clerodanes were active against larvae of Egyptian cotton leafworm, *Spodoptera littoralis*, when present in the diet at 3 ppm doses. Structure-antifeedant activity relations were established. Likewise, first stage larvae of the greenhouse whitefly, *Trialeurodes vaporariorum*, exhibited complete mortality when fed on *A. reptans*. This effect was mainly originated by 29-norsengosterone and ajugalactone, two phytoecdysteroids occurring in this plant. For biotechnological production of phytoecdysteroids its total content in different parts of normally grown or in vitro micropropagated *A. reptans* plants was studied. Great quantitative and qualitative differences were observed. For comparison of these qualitative differences, a dealkylation ratio ($Dr = C_{28} / C_{29}$ phytoecdysteroid content) and a C-5 hydroxylation ratio ($5Hr = 5-OH / 5-H$ phytoecdysteroid content) were established. The 5Hr values appeared to be quite constant ranging from 0.2

to 0.4, whereas Dr values oscillated from 2.3 in whole plants to 12 in root cultures. Production of phytoecdysteroids was highest (≈ 5000 ppm/dry wt) in cultures of roots in an hormone supplemented solid medium.

Canonica L. (1976) Phytoecdysones: environmental degradation. In: Natural Products and the Protection of Plants. (Ed. Marini-Bettolo G.B.) Elsevier, Amsterdam, pp. 211-224.

Chandra S and Rawat D.S. (2015) Medicinal plants of the family Caryophyllaceae: a review of ethno-medicinal uses and pharmacological properties. Integrative Medicine Research 4, 123-131.

Abstract: Several species of the family Caryophyllaceae are widely used by many ethnic communities as traditional medicine throughout the world. The highest number of plants of the family are used in Chinese traditional medicine. The ethnopharmacological studies of this family indicate that plants of the family possess anticancer, antibacterial, antifungal, antiviral, antioxidant, and anti-inflammatory properties. Other miscellaneous properties reported are ribosome inactivation properties, inhibition of prostatic enlargement in rats, and inhibition of intestinal enzyme carboxylase in rats, cerebro-protective activity, and antiobesity in rats. Few reviews have been published yet, providing information regarding medicinal plants of the family and their biomedical properties. All published reviews have focused either on a particular taxa or a few species. The present review is focused on the traditional medicinal uses of the plants of the family Caryophyllaceae along with phytochemical and pharmacological studies of the family. A study of the literature revealed significant traditional medicinal importance of the family. Major chemical constituents of Caryophyllaceae are saponins, Phytoecdysteroids, benzenoids, phenyl propanoids, and nitrogen containing compounds. The most important property of plants of the family is anticancer activity and is shown by the large number of plant species studied. This review of traditional medicinal and pharmacological uses of plants of the family, provide a ground for future research in the family.

Chaubey M.K. (2018) Role of phytoecdysteroids in insect pest management: a review. Journal of Agronomy 17, 1-10.

Abstract: Background and Objective: Climate change influence crops through its effects on the growing, development and yield. Research on climate change risk in staple food crops has been implemented in South Sumatra, Indonesia. The objective of this study was to assess level of climate change risk in staple food crops production (paddy, corn and soybean). Materials and Methods: This study used data of temperature, rainfall, sea level rise, hythergraph, irrigation, production of staple food crops and farmer's socio-economic. Methods of this study were descriptive explanatory that using risks-vulnerability conceptual framework and risks were a function hazard and vulnerability. Results: The results of this study indicate that staple food crops were vulnerable to the impacts of climate change, indicated by hazards such as decreasing production of paddy, corn and soybean due to air temperature increase and rainfall change. Some areas of South Sumatra experienced the high risk of decreasing production of staple food crops. Generally, South Sumatra had decreasing wetland of paddy, upland paddy, corn and soybean production, respectively with an average 9.44, 22.00, 10.7 and 10.10% per year. Conclusion: South Sumatra Indonesia experienced the high risk of decreasing production of staple food crops due to climate change and had potency to disrupt food security in Indonesia.

Cheng Q., Li F., Yan X., He J., Zhang H., Wang C., He Y. and Li Z. (2021) Phytochemical and pharmacological studies on the genus Arcangelisia: a mini review. Arabian Journal of Chemistry 14, article 103346, pp.18.

Abstract: Only 3 plants in the genus Arcangelisia, belonging to the family Menispermaceae, are used as folk medicines for the treatment of various diseases by local residents. Alkaloids are main compounds of berberine analogues found in this genus and they demonstrate a wide range of pharmacological activities. The aim of this review is to compile the phytochemical progress including all the compounds isolated from this genus, their pharmacological activities together with the ^{13}C NMR spectral data of the main bioactive components, which will bring more attention of other researchers to this genus for further study to find new active compounds.

Coll J. (2003) Ecdysteroid biosynthesis in plants. Chemicke Listy 97(S) s249-s251.

Coll Toledano J. (1998) Old and new ecdysteroids in *Ajuga* species: an overview. Russian Journal of Plant Physiology 45(3), 310-315.

Das N., Mishra S.K., Bishayee A., Ali E.S. and Bishayee A. (2021) The phytochemical, biological, and medicinal attributes of phytoecdysteroids: an updated review. Acta Pharmaceutica Sinica B 11(7), 1740-1766 (doi.org/10.1016/j.apsb.2020.10.012).

Abstract: The phytoecdysteroids (PEs) comprise a large group of biologically-active plant steroids, which have structures similar to those of insect-molting hormones. PEs are distributed in plants as secondary metabolites that offer protection against phytophagous (plant-eating) insects. When insects consume the plants containing these chemicals, they promptly molt and undergo metabolic destruction; the insects eventually die.

Chemically, **ecdysteroids** are a group of polyhydroxylated **ketosteroids** that are structurally similar to androgens. The carbon skeleton of ecdysteroids is termed as cyclopentanoperhydro-phenanthrene with a β -side chain at carbon-17. The essential characteristics of ecdysteroids are a *cis*-(5 β -H) junction of rings A and B, a 7-en-6-one chromophore, and a *trans*-(14 α -OH) junction of rings C and D. Plants only synthesize PEs from **mevalonic acid** in the mevalonate pathway of the plant cell using acetyl-CoA as a precursor; the most common PE is 20-hydroxyecdysone. So far, over 400 PEs have been identified and reported, and a compilation of 166 PEs originating from 1998 has been previously reviewed. In the present review, we have summarized 212 new PEs reported between 1999 and 2019. We have also critically analyzed the biological, pharmacological, and medicinal properties of PEs to understand the full impact of these phytoconstituents in health and disease.

Deans B.J., de Salas M., Smith J.A. and Bissember A.C. (2018) Natural products isolated from endemic Tasmanian vascular plants. *Australian Journal of Chemistry* **71**, 756-767.

Abstract: Tasmania is the south-eastern island state of Australia. It is geographically isolated and is recognised for both its rich diversity of plant species and high degree of endemism. Although 530 endemic Tasmanian vascular plant species are known, natural products have only been isolated from 27 of these species (~5.1 %), representing 3 classes (Dicotyledonae, Monocotyledonae, and Gymnospermae), 12 families, and 14 genera. Terpenoids, flavonoids, and alkaloids are the major classes of compound that have been isolated from these species. This report provides the first review of the natural products isolated from endemic Tasmanian plant species and covers ~70 years of research in this area.

Dinan L. (1998) A strategy towards the elucidation of the contribution made by phytoecdysteroids to the deterrence of invertebrate predators on plants. *Russian Journal of Plant Physiology* **45**(3), 296-305 [in English]/ *Fiziologiya Rastenii* **45**(3), 347-359 [in Russian].

Dinan L. (2001) Phytoecdysteroids: biological aspects. *Phytochemistry* **57**(3), 325-339.

Abstract: Phytoecdysteroids are a family of about 200 plant steroids related in structure to the invertebrate steroid hormone 20-hydroxyecdysone. Typically, they are C27, C28 or C29 compounds possessing a 14 α -hydroxy-7-en-6-one chromophore and A/B-*cis* ring fusion (5 β -H). In the present review, the distribution, biosynthesis, biological significance and potential applications of phytoecdysteroids are summarised.

Dinan L. (2003) Isoprenoids as ecdysteroid receptor agonists and antagonists. *Chemicke Listy* **97**(S), s255-s258.

Dinan L., Savchenko T., Whiting P. and Sarker S.D. (1999) Plant natural products as insect steroid receptor agonists and antagonists. *Pesticide Science* **55**, 331-335.

Abstract: Findings to date on plant secondary compounds which possess ecdysteroid-like or anti-ecdysteroid activities in an efficient and effective bioassay based on an ecdysteroid-responsive insect cell-line are summarised. Several novel antagonists have been identified, among which the cucurbitacins are the best characterised and have been shown to compete with ecdysteroids for the ligand binding site of the insect steroid hormone receptor. Certain withanolides, limonoids and resveratrol derivatives also antagonise 20-hydroxyecdysone action. Additionally, several new phytoecdysteroids have been isolated and identified. In common with all other ecdysteroids, these possess agonistic activity in the B II bioassay. Extensive SAR studies based on the potencies of a large number of purified ecdysteroids have been performed and molecular (CoMFA) modelling used to characterise ecdysteroid binding to the ligand binding site of the receptor complex.

Dinan L., Mamadalieva N.Z. and Lafont R. (2020) Dietary phytoecdysteroids. In: *Handbook of Dietary Phytochemicals* (Eds Xiao J. et al.) pp 54 (doi: [org/10.1007/978-981-13-1745-3_35-1](https://doi.org/10.1007/978-981-13-1745-3_35-1)).

Abstract: Phytoecdysteroids are polyhydroxylated steroids which are widely distributed in the plant world and are present in significant amounts in 5–6% of plant species. Their major role in the plant is probably to deter invertebrate predators, but ecdysteroids also have many beneficial effects in mammals and are attracting attention as therapeutic and nutraceutical agents. Four hundred analogues have been identified so far from plant sources, but 20-hydroxyecdysone is the most frequently encountered and is often the major analogue present. Here we consider the occurrence of phytoecdysteroids in food plants and the human diet and how this might change in the future against the backdrop of what we currently know about biosynthesis of these compounds in plants and their bioavailability, metabolism, and biological activities in mammals. Finally, we discuss the medical and pharmaceutical potential of these molecules, particularly in the area of muscle wasting diseases and diabetes, and indicate which areas of fundamental research require focused study.

Egamberdieva D., Mamedov N., Ovidi E., Toezi A. and Craker L. (2016) Phytochemical and pharmacological properties of medicinal plants from Uzbekistan: a review. *Journal of Medicinally Active Plants* **5**(2), 59-75.

Abstract: Medicinal plants are a reservoir of biologically active compounds with therapeutic properties that over time have been discovered and used by diverse groups of people for treatment of various ailments. In this regard, Uzbekistan has an excellent historic research base of herbal medicines with about 70% of Uzbek households using medicinal plants to meet their health needs for several centuries. The flora of Uzbekistan includes more than 4500 species of vascular plants and over 600 of these plants are used in traditional or conventional medicines, significantly surpassing other areas by the absolute number of endemics and the percentage of endemism. The plants are a reservoir of secondary metabolites, suitable for use in pharmacological studies with a high possibility of observing biologically active constituents. The present review provides an up-to-date report on the phytochemicals and pharmacological activity of the medicinal plants widely used in Uzbekistan. As a country, Uzbekistan appears to be a source of novel herbal drugs that have not yet been fully evaluated. We trust the present report will be useful for further investigations on the medicinal activity of indigenous plant species in Uzbekistan.

Egamberdieva D. and Jabborova D. (2018) Medicinal plants of Uzbekistan and their traditional uses. In; Vegetation of Central Asia and Environs (Eds Egamberdieva D. and Öztürk M.) Springer Nature, Switzerland, Chapter 8, p211-237.

Abstract: Uzbekistan is known throughout the world for its rich history of herbal plants and their use in traditional medicine. Uzbekistan significantly surpasses other regions of the world in terms of the endemism and species richness found in this country. These plants are a source for biologically active compounds that are used in pharmacological tests, with a high probability of new drug discovery. Pharmacological studies have confirmed that plant extracts and individual compounds have various biological activities such as hepatoprotective, dermatological, antimicrobial, antiviral, anti-ulcer, antioxidant, neuroprotective, and anti-inflammatory properties. In the present review, an attempt was made to provide an up-to-date report on the diversity of medicinal plants widely used in Uzbekistan and their pharmacological properties. The present report can be useful for the continuous investigation of the Uzbek with the aim of obtaining new biologically active compounds.

Festucci-Buselli R.A., Contim L.A.S., Barbosa L.C.A., Stuart J. and Otoni W.C. (2008) Biosynthesis and potential functions of the ecdysteroid 20-hydroxyecdysone – a review. *Botany* **86**, 978-987.

Abstract: The ecdysteroid 20-hydroxyecdysone (20E) is a steroid hormone found in arthropods and plants. It is suspected to have agrochemical, biotechnological, medicinal, and pharmaceutical applicability. In insects, 20E controls or elicits molting and other developmental processes, and several characterized P450 enzymes are involved in its biosynthesis. In plants, it may act as a defensive substance against insects and nematodes. It is suspected that 20E, being a physiologically active compound, may affect morphological and physiological processes in plants and that C₂₇ phytosterols may be its precursors. However, neither its precise function nor its mechanism of biosynthesis in plants is fully understood. Here, the importance of 20E and current understanding of its structure, potential functions, and biosynthesis in both plants and insects are reviewed.

Fujimoto Y., Ohyama K., Nomura K., Hyodo R., Takahashi K., Yamada J. and Morisaki M. (2000) Biosynthesis of sterols and ecdysteroids in *Ajuga hairy* roots. *Lipids* **35**(3), 279-288.

Abstract: Hairy roots of *Ajuga reptans* var. *atropurpurea* produce clerosterol, 22-dehydroclerosterol, and cholesterol as sterol constituents, and 20-hydroxyecdysone, cyasterone, isocyasterone, and 29-norcyasterone as ecdysteroid constituents. To better understand the biosynthesis of these steroidal compounds, we carried out feeding studies of variously ²H- and ¹³C-labeled sterol substrates with *Ajuga hairy* roots. In this article, we review our studies in this field. Feeding of labeled desmosterols, 24-methylenecholesterol, and ¹³C₂-acetate established the mechanism of the biosynthesis of the two C₂₉-sterols and a newly accumulated codisterol, including the metabolic correlation of C-26 and C-27 methyl groups. In *Ajuga hairy* roots, 3 α -, 4 α -, and 4 β -hydrogens of cholesterol were all retained at their original positions after conversion into 20-hydroxyecdysone, in contrast to the observations in a fern and an insect. Furthermore, the origin of 5 β -H of 20-hydroxyecdysone was found to be C-6 hydrogen of cholesterol exclusively, which is inconsistent with the results in the fern and the insect. These data strongly support the intermediacy of 7-dehydrocholesterol 5 α ,6 α -epoxide. Moreover, 7-dehydrocholesterol, 3 β -hydroxy-5 β -cholest-7-en-6-one (5 β -ketol), and 3 β ,14 α -dihydroxy-5 β -cholest-7-en-6-one (5 β -ketodiol) were converted into 20-hydroxyecdysone. Thus, the pathway cholesterol \rightarrow 7-dehydrocholesterol \rightarrow 7-dehydrocholesterol 5 α ,6 α -epoxide \rightarrow 5 β -ketol \rightarrow 5 β -ketodiol is proposed for the early stages of 20-hydroxyecdysone biosynthesis. 3 β -Hydroxy-5 β -cholestan-6-one was also incorporated into 20-hydroxyecdysone, suggesting that the introduction of a 7-ene function is not necessarily next to cholesterol. C-25 Hydroxylation during 20-hydroxyecdysone biosynthesis was found to proceed with *ca.* 70% retention and 30% inversion. Finally, clerosterol was shown to be a precursor of cyasterone and isocyasterone.

Galal A., Raman V. and Khan I.A. (2015) *Sida cordifolia*, a traditional herb in modern perspective – a review. *Current Traditional Medicine* **1**, 5-17.

Abstract: *Sida cordifolia* (Malvaceae) is a highly reputable medicinal herb in the Ayurveda and other traditional systems of medicine in India and various other countries. In the Ayurvedic system of medicine it is used as antirheumatic, analgesic, antipyretic, antiasthmatic, nasal anticongestant, antiviral, laxative, diuretic, aphrodisiac, hypoglycaemic, hepatoprotective and in the treatment of Parkinson's disease. In order to evaluate this traditional plant in a modern perspective, the current review presents essential aspects of *S. cordifolia* including taxonomy, uses in disciplined traditional medicines, geographical distribution, chemical constituents, pharmacological studies on plant extracts and on single entity constituents, toxicity, and standardization. The chemical composition of this herb comprises of alkaloids, flavonoids, phytoecdysteroids, sterols and fatty acids. The problem of plant misidentification, due to confusion with other related species, is discussed. This paper reviews the conflicting reports regarding the presence or absence of ephedrine and discusses the claimed utility of this herb as a weight loss aid on the basis of ephedrine purported to be present in this species.

Galbraith M.N., Horn D.H.S., Hocks P., Schulz G. and Hoffmeister H. (1967) The identity of 20-hydroxy-ecdysone from various sources. *Die Naturwissenschaften* 54(17), 471-472.

George J., Bais H.P. and Ravishankar G.A. (2000) Biotechnological production of plant-based insecticides. *Critical Reviews in Biotechnology* 20(1), 49-77.

Abstract: The demand for natural and nonpersistent insecticides is increasing day by day. Plant cell cultures could be an alternative to conventional methods of production of insecticides from field-grown plants. In vitro cultured plant cells produce a wide array of insecticides as a part of their secondary metabolism. Their ability to synthesize key enzymes and the manipulation of these could lead to the enhanced production of many insecticides of industrial importance. The development of a high-yielding hairy root culture system for thiophenes, nicotine, and phytoecdysones is of considerable interest. In this article, the current literature on various factors that influence the growth, production, and secretion of six insecticidal compounds, namely, pyrethrins, azadirachtin, thiophenes, nicotine, rotenoids, and phytoecdysones which have been prospects for the scale-up of cell cultures, genetic engineering to obtain transgenic plants, and metabolically engineered plants for increased production of bio-molecules, has been discussed. Environmental safety clearance and the future prospects of application of biomolecules for plant-derived insecticides are presented.

Ghosh D. and Laddha K.S. (2006) Plant steroids to phytoecdysteroids; a new chemical entity overviewed with extraction, purification, and analysis. *Pharmacognosy Magazine* 2(5), 15-26.

Abstract: The phytochemical investigation of ecdysteroids involves the following: extraction of the plant material; separation and isolation of the constituents of interest; characterization of the isolated compounds and quantitative evaluations. Emphasis is placed more on chromatographic procedures and structural elucidations.

Ghosh D. and Laddha K.S. (2006) Extraction and monitoring of phytoecdysteroids through HPLC. *Journal of Chromatographic Science* 44(1), 22-26.

Abstract: The size of the phytoecdysteroids family is rapidly growing. Recent data shows over 250 ecdysteroid analogs have been identified so far in plants. It is theorized that there are over 1000 possible structures, which might occur in nature, but it is a fact that ecdysteroids usually occur in plants as a complex cocktail of structurally different compounds. Among these compounds, the major component is usually the common ecdysteroid-like 20-hydroxyecdysone. Ecdysteroids are polar steroids, almost sugar-like in their solubility properties. Extraction and purification of ecdysteroids (polyhydroxy steroids) is complicated by their polar nature and poor crystallizing properties. These properties make them difficult to separate from other polar plant constituents. Besides, this plant extract is very often processed by multistep procedures to isolate the major and minor ecdysteroids from the new or existing sources. A simplified scheme consisting of a few extraction steps for the purification of ecdysteroid from plants is in great demand. A quantitative approach through high-performance liquid chromatography has been initiated for developing an easy method for the extraction of ecdysteroids from *Ipomoea hederacea* (kaladana) seeds.

Glazowska J., Kaminski M.M. and Kaminski M. (2018) Chromatographic separation, determination and identification of ecdysteroids: focus on Maral root (*Rhaponticum carthamoides*, *Leuzea carthamoides*). *Journal of Separation Science* (doi: 10.1002/jssc.201800506).

Abstract: The review presents general principles for choosing optimal conditions for ecdysteroid separation, identification, and isolation using HPLC/TLC techniques in RP, NP-HILIC or NP modes. Analytics of ecdysteroids pose a still insufficiently resolved problem. Plant-derived ecdysteroids are a point of interest of pharmaceutical industry and sport medicine due to their postulated adaptogenic and anabolic properties. In insects, ecdysteroids regulate larval transformation. Maral root (*Rhaponticum carthamoides*, *Leuzea carthamoides*), traditional Siberian folk-medicine plant used as stimulant to boost overall health and fitness, is a particularly rich source of a wide variety of phytoecdysteroids. The similarity of molecular structures of ecdysteroids present in its extracts together with high content of unrelated compounds of similar chromatographic characteristics makes optimization of

separation, identification and isolation of ecdysteroids a difficult analytical task. In that respect, two-dimensional separations, two-dimensional separations, 2D HPLC or 2D TLC, could be of use. For identification, the hyphenated techniques are particularly important. Thus, comprehensive overview of MS spectral parameters of ecdysteroids is provided. Described principles could easily be applied for separation of ecdysteroids in extracts from other sources. They are also useful for development of separation procedures for isolation of ecdysteroids in preparative-scale applications.

Grzelak A. and Janiszowska W. (2003) Triterpenoid occurrence and *in vitro* biosynthesis in cultures. *Postępy Biologii Komórki* **30**(2), 229-241 [in Polish, with an English abstract].

Guillen P.O., Jaramillo K.B., Genta-Jouve G. and Thomas O.P. (2019) Marine natural products from zoantharians: bioactivity, biosynthesis, systematics, and ecological roles. *Natural Product Reports* (doi: 10.1039/c9np00043g).
Abstract: Zoantharians, also improperly known as zoanths or colonial anemones, are well known by aquarists because of their ease of use in aquaria but also because of their splendid colours. However, high concentrations of the highly toxic palytoxin found in some species of zoantharians maintained in reef aquaria has raised some issues recently, unveiling at the same time a rather unknown chemical diversity hidden in these marine beauties. Herein, we report the structure of the metabolites described in all species of zoantharians up to the end of 2018 and their associated biological activities. As sessile invertebrates, zoantharians harbour a rich diversity of micro-organisms that can play a role in the biosynthesis of these natural products and we detail the current hypotheses on the metabolic pathways leading to the identified ecdysteroids, zoanthoxanthins, zoanthamines, palytoxins and others. Finally, we assess the possible use of these metabolites in the systematics of such a complex group of marine invertebrates and we discuss their possible ecological roles. Altogether, this review brings some insights into the rich chemical diversity of zoantharians and their potential for marine biodiscovery and marine ecology.

Heftmann E. (1970) Insect molting hormones in plants. *Recent Advances in Phytochemistry* **3**, 211-227.

Herout V., Reinhold L. and Liwshitz Y. (1970) Some relations between plants, insects and their isoprenoids. *Progress in Phytochemistry* **2**, 143-202.

Herout V. (1980) Extant flora with respect to its chemistry. In: *Frontiers of Bioorganic Chemistry and Molecular Biology* (Ed: Ananchenko S.N.), Pergamon Press, pp265-275.

Abstract: An attempt is made to follow the conditions of plant species existence with respect to the content of their secondary products. According to present data, an evaluation of the phytochemical contribution to the phylogeny of some developmental lines of the present-day flora is presented (e. g., of the Marchantiopsida, etc.). This synthesis is concentrated mainly to plant-plant, plant-microorganisms, and plant-animal interactions.

Hetru C. and Horn D.H.S. (1980) Phytoecdysteroids and zooecdysteroids. In: *Developments in Endocrinology 7; Progress in Ecdysone Research* (Ed. Hoffmann J.A.), Elsevier/North Holland Biomedical Press, pp. 13-28.

Hikino H. and Hikino Y. (1970) Arthropod molting hormones. *Fortschritte der Chemie Organischer Stoffe* **28**, 256-312.

Abstract: While much is known about the hormones of vertebrates, knowledge of the hormones of invertebrates is far less complete. However, the chemistry of the molting hormones and the juvenile hormones of insects has made surprisingly rapid advances during the past few years and has now become a subject of research which is attracting the interest of both chemists and biologists. This review article is an attempt to summarize recent developments in our knowledge regarding the chemistry, synthesis and metabolism of the arthropod molting hormones, but will in the main exclude consideration of biological properties which have been frequently the object of excellent reviews (71).

Hikino, H. and Takemoto, T. (1972) Arthropod moulting hormones from plants, *Achyranthes* and *Cyathula*. *Die Naturwissenschaften* **59**(3), 91-98.

Abstract: Isolation, chemistry (including structure determination), synthesis, and metabolism of the arthropod moulting hormones (ecdysterols) from *Achyranthes* and *Cyathula* plants belonging to amaranthaceous family are reviewed. The physiological properties of the ecdysterols in higher animals as well as their biological activities in arthropods (and insects in particular) are also described.

Hikino H. and Takemoto T. (1974) Ecdysones of plant origin. In: *Invertebrate Endocrinology and Hormonal Heterophylly* (Ed. Burdette W.J.) Springer-Verlag, pp. 185-203.

Abstract: It is well known that the first insect molting hormone, α -ecdysone, was isolated from the silkworm, *Bombyx mori*, by Butenandt and Karlson in 1954 with a background of pioneering work performed by a

number of researchers involved in the purification of the hormone. Since then six additional molting hormones (zooecdysones) have been isolated from a variety of arthropods, insects, and crustaceans. The content of these zooecdysones in animals is only 10^{-5} – 10^{-9} per cent; the compound occurring most commonly is β -ecdysone. The structure of α -ecdysone was first elucidated in 1965, and the structures of another five zooecdysones were successively determined thereafter.

Hikino H, Okuyama T, Konno C and Takemoto T (1975) Carbon-13 nuclear magnetic resonance spectra of phytoecdysones. *Chemical and Pharmaceutical Bulletin* **23**(1), 125-132.

Abstract: The natural-abundance ^{13}C nuclear magnetic resonance spectra of certain phytoecdysones and their selected derivatives have been measured at 25 MHz. With the aid of complete noise, off-resonance, and single-frequency proton decoupling techniques, and the shifts which occur on formation of specific derivatives, it has been possible to make assignments of the resonances for the phytoecdysones.

Horn D.H.S. (1971) The ecdysones. In: *Naturally occurring insecticides* (Eds. Jacobson M and Crosby D.G.) Marcel Dekker Inc., pp. 333-459.

Huang Y., Wang S., Liu L., Wang J., Song Y., Yuan Q. Yuan X. and Wu C. (2019) Review of traditional uses, botany, chemistry, pharmacology, pharmacokinetics, and toxicology of Radix Cyathulae. *Chinese Medicine* **14**, 17 (doi.org/10.1186/s13020-019-0237-x).

Abstract: Cyathulae Radix (CR), also known as "Chuanniux" is a well-known traditional Chinese herbal medicine that has been used in China for thousands of years. The present work reviewed advances in traditional uses, origin, chemical constituents, pharmacology, pharmacokinetics, and toxicity studies of CR. This work aims to provide more up-to-date references for modern study and application of this plant. Furthermore, the possible trends and perspectives for future research of this plant are also discussed. In China, the roots of CR have been widely used in clinical practice to treat orthopedic, gynecological, and urologic diseases. Currently, over 59 compounds have been isolated and identified from CR, including alkaloids and flavonoids. The extracts and compounds from CR have many pharmacological activities both in vivo and in vitro. They provide beneficial effects on the hematological system and anti-inflammatory properties. However, few studies have investigated the pharmacokinetics and toxicity of CR. Further studies should be undertaken to investigate the clinical effects, toxic constituents, and pharmacokinetics of CR; perform quality evaluation; and establish quality criteria for processed *C. officinalis*. Furthermore, studying the changes of raw and processed CR and the variety of this plant between different cultivated areas and cultivars will be interesting.

Ikekawa N., Hattori F., Rubio-Lightbourn J., Miyazaki H., Ishibashi M. and Mori C. (1972) Gas chromatographic separation of phytoecdysones. *Journal of Chromatographic Science* **10**, 233-242.

Abstract: Preparation of derivatives of the insect moulting hormones ecdysone and phytoecdysones for use in GLC separation was studied. Ecdysones are partially silylated with trimethylsilylimidazole at room temperature and completely silylated at 100°. Both derivatives show single sharp peaks, and the structure of the derivatives were determined by mass spectrometry. Heptafluorobutyryl derivatives of ecdysones can be prepared by exchange reaction between trimethylsilyl and heptafluorobutyryl moieties, which can be identified and determined by GLC at the nanogram or picogram level. GLC analysis of these derivatives are useful for investigation of those hormones in plants and also in biological systems.

Jain A., Choube S., Singour P.K., Raiak H. and Pawar R.S. (2011) *Sida cordifolia* (Linn) – an overview. *Journal of Applied Pharmaceutical Science* **1**(2), 23-31.

Abstract: Our world is fulfilled by various medicinal plants which have widely been used in treatment of various diseases since ancient time. Medicinal plants still play an important role in emerging and developing countries. They also generate income to people of many Asian countries who can earn their livelihood from selling collected materials from the forest or by cultivating on their farms. Thus the medicinal plants constitute very important rational resources. In India plants have been traditionally used for human and veterinary health care needs. This reflects that medicinal plant and their products have taken an increasing demand. Herbs are staging a comeback and herbal 'renaissance' is happening all over the globe. The herbal products today symbolize safety in contrast to the synthetics that are regarded as unsafe to human and environment. Although herbs had been priced for their medicinal, flavoring and aromatic qualities. Malvaceae, commonly known as Bala, is an Ayurvedic medicine that is used to treat bronchial asthma, cold and flu, chills, lack of perspiration, head ache, nasal congestion, aching joints and bones, cough and wheezing, and edema. The root infusion is given in nervous and urinary diseases and also in disorders of the blood and bile. *Sida cordifolia* has been reported to possess analgesic, anti-inflammatory and hypoglycemic activities as well as hepatoprotective activity. Traditionally the plant *Sida cordifolia* (Linn) syn has been used as CNS depressant, fat lose, analgesics, anti-inflammatory, hypotensive, hepatoprotective. Presence of ephedrine has highlighted the utility of this plant. Various ayurvedic preparation of this plant used in asthma diseases, fat lose and

increase energy, Oil preparation is also cure pain, swelling disorder, and Gritha cures Heart diseases. This plant has great potential to develop the Ayurvedic, modern medicine and athletic supplements by pharmaceutical industries. The present review is highlights the traditional uses, Ayurvedic preparation, chemical constituents and pharmacological properties of *Sida cordifolia* (linn) syn. Country Mallow

Jayasinghe L. (1992) Ecdysteroids. Chemistry in Sri Lanka 43-45.

Jefferson R.G. and Walker K..J. (2017) Biological flora of the British Isles: *Serratula tinctoria*. Journal of Ecology 105, 1438-1458.

Summary:

1. This account presents information on all aspects of the biology of *Serratula tinctoria* L. that are relevant to understanding its ecological characteristics and behaviour. The main topics are presented within the standard framework of the *Biological Flora of the British Isles*: distribution, habitat, communities, responses to biotic factors, responses to environment, structure and physiology, phenology, floral and seed characters, herbivores and disease, history and conservation.
2. *Serratula tinctoria* is a gynodioecious perennial forb of open or semi-shaded habitats including grassland, heath, mire, open woodland and scrub and their ecotones. It is found throughout most of England and Wales but it is very rare in Scotland and absent from Ireland. *Serratula tinctoria* is found in Europe as far north as southern Sweden and Norway. It is absent from much of the Boreal biogeographical zone, including most of Fennoscandia and northern Poland, Russia and the Baltic States, and from the lowland Mediterranean zone.
3. *Serratula tinctoria* occurs on soils overlying a wide variety of superficial deposits and sedimentary, metamorphic and igneous rocks, including ultramafic rocks. The pH range of these soils is wide, ranging from acidic to moderately alkaline but with nutrient status classed as oligotrophic, or more rarely mesotrophic. In mire or fen habitats, it occurs in topogenous or soligenous situations with a similar soil water pH range. It is able to tolerate a wide spectrum of soil water-table conditions, ranging from very dry to flooded.
4. *Serratula tinctoria* is pollinated by various insects of the orders Hymenoptera, Diptera and Lepidoptera, particularly bumblebees, hoverflies and butterflies. It produces phytoecdysteroids, which mimic a hormone that regulates insect development.
5. The seed has a feathery pappus for wind dispersal and establishment is primarily from seed. However, it has strong dispersal limitation with slow migration rates into restored grasslands of 1 m year⁻¹. The species has a short-term persistent seed bank. Seedling recruitment is enhanced by disturbance that creates bare ground, increasing light flux and lower cover of bryophytes and litter, and reduced by higher nutrient availability and lack of management.
6. Formerly the source of a yellow dye, *S. tinctoria* has declined in Britain since at least the late 19th Century primarily due to a combination of drainage, ploughing and agricultural improvement and, conversely, lack of management by cutting and grazing in its grassland, heath and mire habitats. There is evidence that the decline slowed or stabilised between the late-1980s and the mid-2000s, in part due to the positive impact of conservation via statutory protection on designated sites and wider measures delivered through agri-environment schemes.

Jones C.G. and Firn R.D. (1991) On the evolution of plant secondary chemical diversity. Philosophical Transactions of the Royal Society, London B 333, 273-280.

Abstract: A common-sense evolutionary scenario predicts that well-defended plants should have a moderate diversity of secondary compounds with high biological activity. We contend that plants actually contain a very high diversity of mostly inactive secondary compounds. These patterns result because compounds arising via mutation have an inherently low probability of possessing any biological activity. Only those plants that make a lot of compounds will be well defended because only high diversity confers a reasonable probability of producing active compounds. Inactive compounds are retained, not eliminated, because they increase the probability of producing new active compounds. Plants should therefore have predictable metabolic traits maximizing secondary chemical diversity while minimizing cost. Our hypothesis has important implications to the study of the evolution of plant defence.

Kalász H., Báthori M. and Csermely T. (2000) Planar versus microcolumn chromatography. American Laboratory 32(19), 28-31.

Karlson P. (1976) Animal hormones as secondary plant products and their ecological relevance. Deutsche Akademie der Naturforschung Nova Acta Leopoldina Supplement 7, 423-432.

Karlson P. (1996) On the hormonal control of insect metamorphosis. A historical review. International Journal of Developmental Biology 40, 93-96.

Kayani W.K., Hasan H. and Mirza B. (2018) Advances in genetic engineering of *Ajuga* species. In: Kumae N (Ed.) Biotechnological approaches for Medicinal and Aromatic Plants. Springer Nature, Singapore, Chapter 27, pp. 599-629.

Abstract: *Ajuga* genus is among one of the more than 250 genera of Labiatae, cosmopolitan in distribution, and comprises of more than 70 species with the remarkable therapeutic importance. Many species of *Ajuga* including *A. bracteosa*, *A. reptans*, *A. Chamaepitys*, etc. have been used in the traditional system of medicine and are also in use for making formulations in modern medicines. *Ajuga* species offer anticancer, antibacterial, antifungal, antidiabetic, anabolic, antileishmanial, anti-inflammatory, hepatoprotective, immunomodulatory, antimalarial, astringent, anthelmintic, and diuretic properties and are used in the treatment of rheumatism, palsy, and gout. They hold a large number of secondary metabolites which are active principles to combat the foresaid diseases including phytoecdysteroids, withanolides, iridoid glycosides, neo-clerodane di- and triterpenoids, sterols, and a large range of flavonoid and phenolic compounds. Some of the species of *Ajuga* are genetically modified for some of these compounds including *A. bracteosa*, *A. reptans*, *A. multiflora*, etc. Latest development made in the exploration of these compounds is yet dealing with the transformation of rol genes and some stress and feeding experiments. We precisely discuss here the details of biotechnological progress that has been made in *Ajuga* species so far.

Kelayeh T.P.S, Abedinzade M. and Ghorban A. (2019) A review on biological effects of *Lamium album* (white dead nettle) and its components. Journal of Herbmed Pharmacology 8(3), 185-193.

Abstract: *Lamium album*, commonly known as white dead nettle, is a plant in the family of Lamiaceae. This plant is distributed all over Asia, Europe, and Africa. In the traditional medicine of Asia, it has been used for the treatment of a number of diseases such as trauma, fracture, paralysis, leucorrhoea, hypertension women's pain, uterine hemorrhage, menorrhagia, vaginal and cervical inflammation. In recent years, *L. album* has been the subject of intensive experimental studies to evaluate its traditional use to reveal new biological properties. A wide range of pharmacological effects, including antimicrobial, anti-inflammatory, anticancer, and antidiabetic properties have been reported by these studies. This review presents an up-to-date overview of the current literature on the pharmacological and physiological effects of *L. album*. Also, phytochemical constituents responsible for the biological properties of *L. album* are presented and discussed.

Kholodova Yu. D. (1979) Phytoecdysteroids - biologically active polyhydroxysteroids. Ukrainskii Biochimiiyeskii Zhurnal 51(5), 560-575 [in Russian, with an English abstract].

Kholodova Y.D. (1987) Phytoecdysteroids. Biokhimiia Zhivotnykhi Cheloveka 11, 27-41 [in Russian].

Klein R. (2004) Phytoecdysteroids. Journal of the American Herbalists Guild (Fall/Winter) 18-28.

Koami T., Ohyama K. and Fujimoto Y. (2002) Mechanism of clerosterol biosynthesis in *Ajuga* hairy roots: stereochemistry of C-28 methylation of 24-methylene sterol. Tetrahedron Letters 43(31), 5479-5481.

Abstract: Biosynthesis of clerosterol, (24S)-ethylcholesta-5,25-dien-3 β -ol (1), involves transfer of the methyl group from S-adenosylmethionine to the C-28 position of a 24-methylene-sterol precursor. The resulting C-24 cationic species undergoes migration of hydrogen from C-25 to C-24, followed by deprotonation from C-26 to form 1. We have now investigated the steric course of the methylation in hairy roots of *Ajuga reptans* var. *atropurpurea*. Feeding of [28E2 H]- and [28Z2 H]-24-methylenecholesterols and 2 H NMR analysis of clionasterol obtained by partial hydrogenation of the biosynthesized clerosterol have revealed that the methylation takes place from the 28-si face.

Kokanova-Nedialkova Z., Nedialkov P.T. and Nikolov S.D. (2009) The genus *Chenopodium*: phytochemistry, ethnopharmacology and pharmacology. Pharmacognosy Reviews 3(6), 280-306.

Abstract: The review includes 154 references on the genus *Chenopodium* covered up to December 2008 and has been compiled using references mainly from Chemical Abstracts and Pubmed. This article briefly reviews the phytochemistry, ethnopharmacology and pharmacology of *Chenopodium* genus. Three hundred seventy nine compounds isolated from different species are reported. Fenolics, flavonoids, saponins, ecdysteroids and triterpenoids were the major classes of phytoconstituents of this genus. The detailed distribution of these compounds among the different *Chenopodium* species with the related references is given in tables. In addition, this review discusses the traditional medicinal uses of different *Chenopodium* species as well as recent developments done in this aspect.

Krasnov Y.A. (2006) Flora of the Siberia is the source of biologically active preparations and drugs.***** (2), 11-18.

Kreis W. and Mueller-Uri F. (2010) Biochemistry of sterols, cardiac glycosides, brassinosteroids, phytoecdysteroids and steroid saponins. Annual Plant Reviews 40, 304-363.

Abstract: Phytosterols are synthesized via the mevalonate pathway of terpenoid formation and arise from the initial cyclization of 3*S*-squalene-2,3-epoxide. Plant sterols are derived from sterols and comprise steroid saponins, steroid alkaloids, pregnanes, androstanes, estranes, ecdysteroids, with anolides and cardiac glycosides. The typical route of sterol and steroid biosynthesis follows the cycloartenol pathway, whereas the lanosterol route seems to be operative mainly in fungi and animals. It was demonstrated, however, that both sterol pathways can be operative in higher plants. Crucial steps in the conversion of cycloartenol/lanosterol to sterols are the events leading to the removal of the methyl groups at C-4 and C-14. Meanwhile, all steps in the sterol pathway have been elucidated and the respective enzymes/genes characterized. The biosynthetic pathway leading from phytosterol precursors to the cardiac glycosides – important compounds in the treatment of cardiac insufficiency in humans – was basically deduced from studies using radiolabelled precursors. The more recent identification and characterization of several enzymes/genes involved in pregnane and cardenolide metabolism, such as 3 β -hydroxysteroid dehydrogenase and progesterone 5 β -reductase, have further clarified the pathway. Brassinosteroids (BRs) are hydroxylated derivatives of cholestane and they are specific plant steroid hormones that are essential for normal plant development. The biosynthesis of BRs has mainly been studied in *Arabidopsis thaliana*. Many of the genes encoding biosynthetic enzymes have been cloned using mutants of *Arabidopsis thaliana*, pea, tomato and rice which revert to a wild-type phenotype following treatment with exogenous BRs. Phytoecdysteroids are related in structure to the invertebrate steroid hormones. Their biological significance in plants is still under discussion. The understanding of the biosynthetic pathway(s) for phytoecdysteroids is very limited. Steroid saponins constitute a vast group of glycosides present almost exclusively in the monocotyledonous angiosperms, and occurring in only a few dicotyledonous families. As far as enzymatic and genetic aspects are concerned, the biosynthesis of steroid saponins (including the steroid alkaloids) has not been studied extensively. The with anolides are C28-steroids and biogenetically related to the steroid saponins in that they are derived from ergostane-type sterols. These compounds appear to be specific for the Solanaceae and their biosynthesis has not yet been studied at the enzyme/gene level.

Krepinsky J., Findlay J.A., Danieli B., Palmisano G., Beynon P. and Murakami S. (1977) Concerning Carbon-13 n.m.r. spectroscopy of 5 β -hydroxylated phytoecdysones. *Organic Magnetic Resonance* **10**, 255-257.

Abstract: The ¹³C n.m.r. spectra of the 5 β -hydroxylated phytoecdysones polypodine B, muristerone A and kaladasterone are presented and briefly discussed together with the spectrum of makisterone A. Comparisons with previously reported spectra of ecdysone, ecdysterone and poststerone are made and allow a correction for the C-20/C-24 assignment for ecdysone.

Kubo I. (1992) Recent examples of natural products isolation by countercurrent chromatographic methods. *Journal of Liquid Chromatography* **15**(15/16), 2843-2855.

Abstract: The efficient isolation of two steroidal glycoalkaloids from the Andean potato “papa negra”, three bitter quassinoids from the bark of *Castela tortuosa*, and several phytoecdysteroids from the root barks of *Vitex strickeri* was accomplished by countercurrent chromatographic methods.

Kubo I., Matsumoto A. and Hanke F.J. (1985) The ¹H-NMR assignment of 20-hydroxyecdysone. *Agricultural and Biological Chemistry* **49**(1), 243-244.

No Abstract.

Kuroda M., Mimaki Y. and Sashida Y. (1999) Cholestane rhamnosides from the bulbs of *Ornithogalum saundersiae*. *Phytochemistry* **52**(3), 445-452.

Abstract: Phytochemical examination of the bulbs of *Ornithogalum saundersiae* yielded six cholestane rhamnosides, two of which had previously been isolated from the same plant material. However, detailed spectroscopic analysis of the aglycone led us to revise the configuration of the C-11 hydroxyl group of the latter two and reassign their structures as (2*S*)-cholest-5-ene-3 β ,11 α ,16 β ,22-tetrol 16-O- α -L-rhamnopyranoside and (2*S*)-cholesta-5,24-diene-3 β ,11 α ,16 β ,22-tetrol 16-O- α -L-rhamnopyranoside, respectively. The other four are new naturally occurring constituents and their structures were determined to be (2*S*)-cholest-5-ene-3 β ,11 α ,16 β ,22-tetrol 16-O-(2,3-di-O-acetyl- α -L-rhamnopyranoside), (2*S*)-cholest-5-ene-3 β ,11 α ,16 β ,22-tetrol 16-O-{2-O-acetyl-3-O-(3,4,5-trimethoxybenzoyl)- α -L-rhamnopyranoside}, (2*S*)-cholest-5-ene-3 β ,11 α ,16 β ,22-tetrol 16-O-{2-O-acetyl-3-O-(*p*-methoxybenzoyl)- α -L-rhamnopyranoside} and (2*S*)-cholesta-5,24-diene-3 β ,11 α ,16 β ,22-tetrol 16-O-(2,3-di-O-acetyl- α -L-rhamnopyranoside), respectively. The isolated compounds were evaluated for their cytostatic activity against leukemia HL-60 cells.

Lafont R. (1998) Phytoecdysteroids in world flora: diversity, distribution, biosynthesis and evolution. *Russian Journal of Plant Physiology* **45**(3), 276-295.

Lafont R. (2003) Ecdysteroids and related molecules in animals and plants. *Chemicke Listy* **97** (S) s280-s281.

Lafont R. and Horn D.H.S. (1989) Phytoecdysteroids: structure and occurrence. In: Ecdysone: from chemistry to mode of action (Ed. Koolman J), Thieme Verlag, Stuttgart, pp. 39-64.

Lafont R.D. and Wilson I.D. (1996) The Ecdysone Handbook, 2nd Ed. The Chromatographic Society, Nottingham, pp. 525

Lafont R., Bouthier A. and Wilson I.D. (1991) Phytoecdysteroids: structures, occurrence, biosynthesis and possible ecological significance. In: Insect Chemical Ecology (Ed. Hrdý I.) Academia Prague and SPB Academic Publishers The Hague, pp. 197-214.

Lafont R., Morgan E.D. and Wilson I.D. (1994) Chromatographic procedures for phytoecdysteroids. *Journal of Chromatography A* **658**, 31-53.

Abstract: The complexity of the mixtures of ecdysteroids in plants and the close similarities in their chemical structures have challenged chemists to find suitable ways to separate and identify them. Great ingenuity has been applied to these problems and consequently a wide range of separation and methods are available today. These methods have been reviewed with assessment of their strengths and limitations, with the intention to guide investigators towards the methods most useful to their purpose.

Lafont R., Harmatha J. and Dinan L. (2003) Ecdybase [<http://ecdybase.org>] - the 2003 update of the natural ecdysteroid database. *Chemicke Listy* **97**(S) s269.

Lafont R., Harmatha J., Marion-Poll F., Dinan L. and Wilson I. (2003) Ecdybase - The Ecdysone Handbook, 3rd Edition, Cybersales, Prague [<http://ecdybase.org>].

Lafont R., Blais C., Harmatha J. and Wilson I.D. (2000) Ecdysteroids: chromatography. In: *Encyclopaedia of separation science*. Academic Press, pp. 2631-2643.

Abstract: Ecdysteroids represent a large family of polyhydroxylated steroids found in plants and in animals. Many different chromatographic procedures can be used with these molecules, among which thin-layer chromatography and high performance liquid chromatography are the most popular ones. They can be separated by using either normal-phase or reversed-phase chromatographic systems, and their detection is best performed by diode-array detectors and mass spectrometry. In the case of insect or arthropod extracts, specific methods (nano LC-MS/MS or the use of derivatives) have been developed allowing accurate measurements of trace amounts in small samples (e.g., in a few or even a single *Drosophila*).

Lapenna S., Gemen R., Wollgast J., Worth A., Maragkoudakis P. and Caldeira S. (2015) Assessing herbal products with health claims. *Critical Reviews in Food Science and Nutrition* **55**, 1918-1928.

Abstract: Herbs, herbal extracts, or phytochemicals are broadly used as foods, drugs, and as traditional medicines. These are well regulated in Europe, with thorough controls on both safety and efficacy or validity of health claims. However, the distinction between medicines and foods with health claims is not always clear. In addition, there are several cases of herbal products that claim benefits that are not scientifically demonstrated. This review details the European Union (EU) legislative framework that regulates the approval and marketing of herbal products bearing health claims as well as the scientific evidence that is needed to support such claims. To illustrate the latter, we focus on phytoecdysteroid (PE)-containing preparations, generally sold to sportsmen and bodybuilders. We review the limited published scientific evidence that supports claims for these products in humans. In addition, we model the *in silico* binding between different PEs and human nuclear receptors and discuss the implications of these putative bindings in terms of the mechanism of action of this family of compounds. We call for additional research to validate the safety and health-promoting properties of PEs and other herbal compounds, for the benefit of all consumers.

Le Bizec B., Antignac J.-P., Monteau F. and Andre F. (2002) Ecdysteroids: one potential new anabolic family in breeding animals. *Analytica Chimica Acta* **473**(1/2), 89-97.

Abstract: Ecdysteroids, polyhydroxylated Δ^7-8-6 -ketosteroids, are widespread in plants and invertebrates, especially arthropods. As early as the 1960s, several studies underlined the potential of ecdysterone to enhance the rate of protein synthesis in mammalian tissue. For these suspected anabolic properties, ecdysteroids such as ecdysone or ajugasterone, are widely diffused on the Internet and are presented as powerful growth promoters without any hormonal consequences. Because these compounds would be potentially used as anabolizing agents in food producing animals, it was necessary to develop an efficient analytical method dedicated to their analysis in biological matrices. Gas chromatography-mass spectrometry (GC-MS) was compared to liquid chromatography (LC)-MS for this purpose. Ecdysteroids, because of their numerous alcohol functions produced, after derivatisation, various by-products, making the GC-MS interpretation difficult. LC-MS was more appropriate, especially when positive

electrospray ionization (ESI) and MS/MS acquisition were used; detection and identification (4 MRM transitions) limits were determined in the pg range.

Lefèvre G. and Rivière C. (2019) Amaranthaceae halophytes from the French Flanders coast of the North Sea: a review of their phytochemistry and biological activities. *Phytochemistry Reviews* (doi.org/10.1007/s11101-019-09636-w).

Abstract: Halophytes are plant species that tolerate high salinity levels. To adapt to these particular abiotic conditions, they develop multiple physiological, biochemical and molecular mechanisms, including the biosynthesis of osmolytes, enzymes and specialized metabolites. The French Flanders coast of the North Sea is an ideal environment for this kind of plant. Amaranthaceae is one of the most represented botanical families of halophytes present on this coast, with 15 species belonging to 7 genera, namely *Atriplex*, *Beta*, *Halimione*, *Kali*, *Oxybasis*, *Salicornia* and *Suaeda*. Some of these species are well known as wild edible plants, and some are used in traditional medicine. This review examines the chemistry of these species and their potential for human health.

Lehman A.D., Dunkel F.V., Klein R.A., Ouattara S., Diallo D., Gamby K.T. and N'Diaye M. (2007) Insect management products from Malian traditional medicine - establishing systematic criteria for their identification. *Journal of Ethnopharmacology* **110**, 235-249.

Abstract: In material-resource poor countries like Mali, traditional practices incorporate the use of plants for medicinal purposes. Ethnobotanical research has documented traditional uses of plants, while concomitant studies by natural product chemists, ethnobotanists, and microbiologists have verified the efficacy of using traditional medicinal plants that have proven antimicrobial activity. These plants may also be used to protect agricultural crops pre-harvest and post-harvest from insect herbivory. In Mali, subsistence farmers, regional scientists, and extension specialists rely on local plants for many medicinal needs and are amenable to using traditional plant materials for insect pest management. The goal of this research was to develop Integrated Pest Management (IPM) strategies using Malian traditional medicine as a discovery lead. The discovery premise was based on identifying plants through a matrix approach utilizing agricultural scientists, traditional practitioners, and subsistence farmers. We hypothesized that plants used in traditional medicine with antimicrobial activity lead to potential insect pest management agents. To test our hypothesis, we developed a four-step process for selecting Malian plant species. Seven criteria were selected to create a systematic matrix to identify the most promising plant materials for practical, affordable, ecologically-sound insect management by Malian farmers. In the first step of the process, we developed a list of 294 medicinal Malian plant species which were evaluated using the matrix. Sixty-seven plant species met our main criteria. After the environmental soundness of these species was evaluated using four minor criteria, 50 species emerged from this pre-chemical, pre-bioassay process for further consideration in IPM programs in Mali.

Li J., Wang C., Han X., Qi W., Chen Y., Wang T., Zheng Y. and Zhao X. (2016) Transcriptome analysis to identify the putative biosynthesis and transport genes associated with the medicinal components of *Achyranthes bidentata* Bl. *Frontiers in Plant Science* **7**: 1860 (doi: 10.3389/pls.2016.01860).

Abstract: *Achyranthes bidentata* is a popular perennial medicine herb used for 1000s of years in China to treat various diseases. Although this herb has multiple pharmaceutical purposes in China, no transcriptomic information has been reported for this species. In addition, the understanding of several key pathways and enzymes involved in the biosynthesis of oleanolic acid and ecdysterone, two pharmacologically active classes of metabolites and major chemical constituents of *A. bidentata* root extracts, is limited. The aim of the present study was to characterize the transcriptome profile of the roots and leaves of *A. bidentata* to uncover the biosynthetic and transport mechanisms of the active components. In this study, we identified 100,987 transcripts, with an average length of 1146.8 base pairs. A total of 31,634 (31.33%) unigenes were annotated, and 12,762 unigenes were mapped to 303 pathways according to the Kyoto Encyclopedia of Genes and Genomes pathway database. Moreover, we identified a total of 260 oleanolic acid and ecdysterone genes encoding biosynthetic enzymes. Furthermore, the key enzymes involved in the oleanolic acid and ecdysterone synthesis pathways were analyzed using quantitative real-time polymerase chain reaction, revealing that the roots expressed these enzymes to a greater extent than the leaves. In addition, we identified 85 ATP-binding cassette transporters, some of which might be involved in the translocation of secondary metabolites.

Lin M., Han P., Li Y., Wang W., Lai D. and Zhou L. (2019) Quinoa secondary metabolites and their biological activities or functions. *Molecules* **24**, 2512 (doi: 10.3390:molecules24132512).

Abstract: Quinoa (*Chenopodium quinoa* Willd.) was known as the “golden grain” by the native Andean people in South America, and has been a source of valuable food over thousands of years. It can produce a variety of secondary metabolites with broad spectra of bioactivities. At least 193 secondary metabolites from quinoa have been identified in the past 40 years. They mainly include phenolic acids, flavonoids, terpenoids, steroids, and nitrogen-containing compounds. These metabolites exhibit many physiological functions, such as insecticidal, molluscicidal

and antimicrobial activities, as well as various kinds of biological activities such as antioxidant, cytotoxic, anti-diabetic and anti-inflammatory properties. This review focuses on our knowledge of the structures, biological activities and functions of quinoa secondary metabolites. Biosynthesis, development and utilization of the secondary metabolites especially from quinoa bran were prospected.

Liu M. and Li X. (1997) Spectral rules of phytoecdysteroids. *Natural Product Research and Development* 10(4), 5-11 [in Chinese, with English abstract].

Liu M. and Li X. (1998) Spectral specificity of Ajugasterone C. *Chinese Journal of Magnetic Resonance* 15(6), 539-542.

Luan F., Han K., Li M., Zhang T., Liu D., Yu L and Lv H. (2019) Ethnomedicinal uses, phytochemistry, pharmacology, and toxicology of species from the genus *Ajuga* L.: a systematic review. *The American Journal of Chinese Medicine* 47(5), 959-1003.

Abstract: The present review is aimed at providing a comprehensive summary of the botanical characteristics, ethnomedicinal uses, phytochemical, pharmacological, and toxicological studies of the genus *Ajuga* L. The extensive literature survey revealed *Ajuga* L. species to be a group of important medicinal plants used for the ethnomedical treatment of rheumatism, fever, gout, sclerosis, analgesia, inflammation, hypertension, hyperglycemia, joint pain, palsy, amenorrhea, etc., although only a few reports address the clinical use and toxicity of these plants. Currently, more than 280 chemical constituents have been isolated and characterized from these plants. Among these constituents, *neo*-clerodane diterpenes and diterpenoids, phytoecdysteroids, flavonoids, and iridoids are the major bioactive compounds, possessing wide-reaching biological activities both *in vivo* and *in vitro*, including anti-inflammatory, antinociceptive, antitumor, anti-oxidant, antidiabetic, antimicrobial, antifeedant, antidiarrhoeal, hypolipidemic, diuretic, hypoglycaemic, immunomodulatory, vasorelaxant, larvicidal, antimutagenic, and neuroprotective activity. This review is aimed at summarizing the current knowledge of the ethnomedicinal uses, phytochemistry, biological activities, and toxicities of the genus *Ajuga* L. to reveal its therapeutic potentials, offering opportunities for future researches. Therefore, more focus should be paid to gathering information about their toxicology data, quality-control measures, and the clinical application of the bioactive ingredients from *Ajuga* L. species.

Luu, B. and G. Ourisson (1989) Stérols et triterpènes polyoxygénés: une famille de produits à large spectre d'activités biologiques [Polyoxygenated sterols and triterpenes: a family of natural products with manifold bioactivities]. *Médecine/Sciences* 5(6), 403-407 [in French, with an English abstract].

Summary: Cholesterol and the other sterols are precursors of many hormones of vertebrates (pregnane, androstane, estrane derivatives) or invertebrates (ecdysteroids) or even plants (brassinoids). The intermediates between sterols and these essential bioactive metabolites can display themselves two types of important biological activities: they may regulate the biosynthesis of sterols by interacting with some of the enzymatic steps of the biosynthesis, or they may exert some action on other individuals either of the same species (pheromonal activity), or of other species (ecological activities). Examples are given of cytotoxic and tumour-modulating substances, which are simple hydroxylated derivatives of cholesterol normally present in animal cells, and may be useful therapeutically.

Maior M.C. and Dobrota C. (2013) Natural compounds with important medical potential found in *Helleborus* sp. *Central European Journal of Biology* 8(3), 272-285.

Abstract: *Helleborus* (family Ranunculaceae) are well-known as ornamental plants, but less known for their therapeutic benefits. Over the past few years, *Helleborus* sp. has become a subject of interest for phytochemistry, pharmacology and other medical research areas. On the basis of their usefulness in traditional medicine, it was assumed that their biochemical profile could be a source of metabolites with the potential to overcome critical medical issues. There are studies involving natural extracts from these species which demonstrate that *Helleborus* plants are a valuable source of chemical compounds with great medical potential. Some phytochemicals produced by these species have been separated and identified a few decades ago: hellebrin, deglucohellebrin, 20-hydroxyecdysone and protoanemonin. Lately, many other active compounds have been reported and considered as promising remedies for severe diseases such as cancer, ulcer, diabetes and also for common medical problems such as toothache, eczema, low immunity and arthritis. This paper is an overview of the *Helleborus* genus focusing on some recently discovered compounds and their potential for finding new drugs and useful biochemicals derived from these species.

Mamadaliyeva N.Z. (2012) Phytoecdysteroids from *Silene* plants: distribution, diversity and biological (antitumour, antibacterial and antioxidant) activities. *Boletín Latinoamericano y del Caribe de Plantas Medicinales y Aromáticas* 11(6), 474-497.

Abstract: *Silene* is a genus of the Caryophyllaceae family, contains more than 700 species, which are widely distributed in Northern Hemisphere, but also in Africa, Asia and South American. Phytochemical investigations of *Silene* species have revealed that many components from this genus are highly bioactive. More than 400 compounds has been isolated, among them major are phytoecdysteroids. The paper reviews the biological (antitumour, antibacterial and antioxidant) activities and the phytoecdysteroids of genus *Silene*. We summarized the phytoecdysteroids content referring to 171 species from the genus *Silene* and list 93 phytoecdysteroids isolated over the past few decades. There are also reports on the mentioned folk and traditional effects of *Silene* plants.

Mamrasulov B., Davranov K. and Jabbarova D. (2020) Phytochemical, pharmacological and biological properties of *Ajuga turkestanica* (Rgl.) Brig (Lamiaceae). *Annals of Phytomedicine* 9(1), 44-57.

Abstract: *Ajuga turkestanica* (Rgl.) Brig (Lamiaceae) is a medicinal, herbaceous flowering species which has been traditionally used in Uzbekistan for cure of various human diseases like, heart disease, muscle aches and stomach problems. This plant possesses diverse pharmacological activities, antibacterial activity, hypoglycemic activity, hypolipidemic action, anabolic activity: growth promotion, increase in protein synthesis in skeletal muscle cells, hepatoprotection activities. Several compounds have been isolated from *A. turkestanica*, which display a wide spectrum of biological and pharmacological activities. This review provides scientific information on the biologically active substances of the medicinal plant

Meena A.K., Singh U., Yadav A.K., Singh B. and Rao M.M. (2010) Pharmacological and phytochemical evidences for the extracts from plants of the genus *Vitex* – a review. *International Journal of Pharmaceutical and Clinical Research* 2(1), 1-9.

Abstract: The scientific basis for the statement that plants and their active constituents play an important role in the prevention diseases is continuously advancing. In fact, the origin of many therapeutic substances from the genus *Vitex*. The genus *Vitex* contains 270 species distributed around the world. It is an interesting source of potential bioactive molecules, as iridoids compounds, flavonoids, diterpenoids derivatives, phytosteroids, with antioxidant, anti-inflammatory, antimicrobial, Hepatoprotective activity, analgesic and antihistamine, Anti-implantation, antiasthmatic activities. This work reviews the pharmacological evidence of extracts of plants from the genus *Vitex*, giving an overview of the most studied biological effects and the known phytochemical composition. Although more studies are necessary, *Vitex* exhibits proven potential to become of important pharmacological interest.

Milano F., Mussi F., Fornaciari S., Altunoz M., Forti L., Arru L. and Buschini A. (2019) Oxygen availability during growth modulates the phytochemical profile and the chemo-protective properties of spinach juice. *Biomolecules* 9, 53 (doi: 10.3390/biom9020053).

Abstract: Fruits and vegetables are a good source of potentially biologically active compounds. Their regular consumption in the human diet can help reduce the risk of developing chronic diseases such as cardiovascular diseases and cancer. Plants produce additional chemical substances when subject to abiotic stress or infected by microorganisms. The phytochemical profile of spinach leaves (*Spinacia oleracea* L.), which is a vegetable with widely recognized health-promoting activity, has been affected by applying root hypoxic and re-oxygenation stress during plant growth. Leaf juice at different sampling times has been subject to liquid chromatography mass spectrometry (LC-MSn) analysis and tested on the human colorectal adenocarcinoma cell line HT29 by using the Comet assay. The cells were previously treated with H₂O₂ to simulate the presence of an oxidative stress (as in colon cancer condition) and the leaf juice application resulted in a significant antioxidant and protective in vitro effect. The duration of the hypoxic/re-oxygenation stress imposed on the plant reflects the antioxidant leaf juice content. After hypoxic stress (24 hours) and reoxygenation (2 hours), we show a decrease (50%) of the relative abundance of the principal identified antioxidant molecules but a higher antioxidant activity of the spinach juice on HT29 cells (20%). Data shows a complex relation between plant growing conditions and the modulation of secondary metabolites content in leaf juice that results in different chemo-protective activities in colon cancer cells.

Nakajima N., Fujioka S., Tanaka T., Takatsuto S. and Yoshida S. (2002) Biosynthesis of cholestanol in higher plants. *Phytochemistry* 60, 275-279.

Abstract: To understand the early steps of C₂₇ brassinosteroid biosynthesis, metabolic experiments were performed with *Arabidopsis thaliana* and *Nicotiana tabacum* seedlings, and with cultured *Catharanthus roseus* cells. [26, 28-²H₆]Campestanol, [26-²H₃]cholesterol, and [26-²H₃]cholestanol were administered to each plant, and the resulting metabolites were analyzed by gas chromatography–mass spectrometry. In all the species examined, [²H₃]cholestanol was identified as a metabolite of [²H₆]campestanol, and [²H₃]cholest-4-en-3-one and [²H₃]cholestanol were identified as metabolites of [²H₃]cholesterol. This study revealed that cholestanol (C₂₇ sterol) was biosynthesized from both cholesterol (C₂₇ sterol) and campestanol (C₂₈ sterol). It was also demonstrated that cholestanol was converted to 6-oxocholestanol, and campestanol was converted to 6-oxocampestanol.

Nakanishi K. (1971) The ecdysones. *Pure and Applied Chemistry* 25(11), 167-195.

Abstract: The discovery of ecdysones, moulting hormones of insects and crustaceans, from plants in late 1966 has resulted in subsequent isolation of a total of about 40 new phytoecdysones. This has contributed a great deal to the understanding of the chemistry and physicochemical data of ecdysones. A general account of these aspects is given, together with results of preliminary bio-organic studies concerning the metabolic fate of exogenous ecdysones and the site of biosynthesis of ecdysones in the silkworm. Finally, very recently several plants have been discovered to contain substances which inhibit the moulting as bioassayed by the topical dipping method. The structure of the first to be elucidated, ajugalactone, is discussed. The second inhibitor to be isolated is interesting because it is a non-steroid. Another intriguing aspect of these inhibitors is their specificity towards the ecdysone structure.

Nakanishi K. (1977) Insect growth regulators from plants. In: Natural products and the protection of plants. (Ed. Marini-Bettolo G.B.) Elsevier, pp. 185-210.

Nakanishi K. (1992) Past and present studies with ponasterones, the first insect molting hormones from plants. *Steroids* **57**, 649-657.

Abstract: The search for antitumor compounds from Southeast Asian plants led to ponasterones, the first phytoecdysteroids, just a year after structure determination of ecdysone and 20-hydroxyecdysone. An independent study of Chinese herb constituents by Takemoto et al. at Tohoku University led to the simultaneous and totally independent discovery of phytoecdysteroids. These findings greatly facilitated research in insect and crustacean physiology. The original structural studies on various phytoecdysteroids have led to interdisciplinary bioorganic studies in the area related to ecdysone receptor, ecdysone biosynthetic precursor (or its storage form), crustacean molt inhibitory factors, and so on.

Nakanishi K. (2006) Studies in microbial and insect natural products chemistry. *Journal of Natural Medicine* **60**, 2-20.

Abstract: This article reviews selected studies performed over the years on natural products other than those of marine origin. The topics are: microbial products (monascorubrin and monascoflavin, rapamycin, intramolecular symbionts); ginkgolides and bilobalide; taxine and taxicin; phytoalexins—ipomeamarones; phytoalexins—cabenegrins; ocular pigment A2E and bilberry; and absolute configuration studies of monoalcohols and primary monoamines by zinc porphyrin host—anisotomonic acid.

Nakanishi K., Koreeda M. and Schooley D.A. (1974) Recent studies on ecdysones. In: *Invertebrate Endocrinology and Hormonal Heterophyly* (Ed. Burdette W.J.), Springer-Verlag, pp. 204-217.

Naz H., Nawaz H., Ayub M.A., Mushtaq A. and Khan S. (2016) A review on phytopharmacological properties of Bisffajj [*Polypodium vulgare*]. *International Journal of Chemical and Biochemical Sciences* **9**, 111-115.

Abstract: *Polypodium vulgare* L. is a perennial fern belonging to Polypodiaceae family. It has been cultivated throughout the world and used for essential oil application, aroma flavor and in traditional medicine. Mostly *Polypodium vulgare* contain polypody rhizome having saponins (polypodosapogenin), ecdysteroids, phloroglucins, volatile oil, fixed oil, and tannins. The extent of each of these chemical constituents varies depending on the type of species or cultivars as well as cultivation conditions such as spore type, weather, artificial condition (green house). It is an essential component of several industrial applications that ranges from food to pharmaceutical application. More uses and applications of *Polypodium Vulgare* by products are continuously added. Further research to maximize yield per hectare and optimum preservation and oil extraction methods are needed, particularly in the developing world where basil leaf and flower harvesting and postharvest processing methods are much traditional.

Ogawa S., Nishimoto N. and Matsuda H. (1974) Source and availability of ecdysterones. In: *Invertebrate Endocrinology and Hormonal Heterophyly* (Ed. Burdette W.J.), Springer-Verlag, pp. 218-232.

Okauchi T. (1969) Recent studies on phytoecdysones. *Botyu-Kagaku* **34**(3), 140-156 [in Japanese].

Okuzumi K., Hara N., Fujimoto Y., Yamada J., Nakamura A., Takahashi K. and Morisaki M. (2003) Biosynthesis of phytoecdysteroids in *Ajuga hairy* roots: clerosterol as a precursor of cyasterone, isocyasterone and 29-norcyasterone. *Tetrahedron Letters* **44**(2), 323-326.

Abstract: Feeding studies of six ¹³C-labeled sterols, including clerosterol, to hairy roots of *Ajuga reptans* var. *atropurpurea* have established that clerosterol is a precursor of three phytoecdysteroids, cyasterone, isocyasterone and 29-norcyasterone.

Olatunde O.Z., Yang Y., Yong J. and Lu C. (2019) Advance of the chemical components and biological activities of *Ajuga decumbens* Thunb. *Biomendical Journal of Scientific and Technical Research* (doi: 10.26717/BJSTR.2019.12.002314).

Abstract: *Ajuga decumbens* Thunb is an annual herbaceous flowering plant, distributed in some parts of Asia. It is adequately used as folk medicines for the treatment different ailments. It contained important bioactive compounds (diterpenoids, iridoid glycosides, phytoecdysteroids, flavonoids and sesquiterpenoids) which made it to possess a wide spectrum of biological activities such as anti-cancer, antioxidant, Antifeedant, antibacterial, anti-inflammatory, antihyperlipidemic, anti-cholinesterase and cytotoxicity activities. This review gives an update about its phytochemicals and biological activities.

Olennikov D.N. and Kashchenko N.I. (2018) *Rhaponticum uniflorum*: chemical components and biological activity. *Khimiya Rastitel'nogo Syr'ya* (2), 5-220 [in Russian, with English Abstract].

Abstract: *Rhaponticum uniflorum* (L.) DC. is a plant species of the Asteraceae family, widely used in traditional medicine in the Eastern Asia. Currently *R. uniflorum* is a subject of scientific interest of the chemists, biologists, pharmacologists, and others. This review includes scientific data from 1991 to 2017. The investigation of the chemodiversity of *R. uniflorum* showed the presence of more than 100 compounds, including sesquiterpenes, diterpenes, ecdysteroids, triterpenes, sterols, thiophenes, flavonoids, amino acids, fatty acids, etc. Ecdysteroids and triterpenes (more than 40 compounds) are the most studied groups of substances of *R. uniflorum*. The data about the methods of chromatographic analysis of terpenoids and phenolic compounds, as well as the quantitative content of some compounds in various organs of *R. uniflorum* are summarized in this paper. It is shown that the extracts and some compounds of *R. uniflorum* possess a wide spectrum of biological activity, including anxiolytic, stress-protective, actoprotective, antihypoxic, anabolic, hepatoprotective, inhibiting PPAR γ receptors, anti-inflammatory, antitumor, immunostimulating, antiatherosclerotic, hypolipidemic effects. However, in spite of the known information about the pharmacological activity of *R. uniflorum*, the data of clinical trials are absent, thereby further investigations of this plant species is necessary.

Orlova I.V., Zacharchenko N.S., Semenyuk E.G., Nosov A.M., Volodin V.V. and Bur'yanov Y.I. (1998) The initiation of transformed root culture from *Rhaponticum carthamoides*. *Russian Journal of Plant Physiology* 45(3), 339-341 [in English]/*Fisiologiya Rastenii* 45(3), 397-400 [in Russian].

Orlova I.V., Semenyuk E.G., Volodin V.V., Nosov A.M. and Burjanov Ya.I. (2000). The development of the system of regeneration and genetic transformation of *Rhaponticum carthamoides* - ecdysteroids producers. *Russian Journal of Plant Physiology* 47(3), 355-359 [in English]/*Fisiologiya Rastenii* 47(3), 402-407 [in Russian].

Abstract: An efficient system for plant regeneration from leaf explants has been worked out. Gene transformation of a medicinal plant *Rhaponticum carthamoides* (Willd.) Iljin with a recombinant strain of *Agrobacterium tumefaciens* GV 3101 containing plasmid with the gene *rolC* was performed for the first time. PCR analysis of DNA from transformed shoots showed that the genome comprised the copies of *rolC* and *nptII* genes.

Pal A., Jadon M., Katare Y.K., Singour P.K., Rajak H., Chaurasiya P.K., Patil U.K. and Pawar R.S. (2011) *Ajuga bracteosa* Wall: a review on its ethnopharmacological and phytochemical studies. *Pelagia Research Library* 2(2), 1-10.

Abstract: The aim of the study is an attempt to investigate the phytochemical and ethnopharmacological application of *Ajuga bracteosa* wall benth. to treat cancer, diabetes, gastrointestinal disorders, worm infestations, urinary disorders, fungal infections, inflammation and tuberculosis in folk medicines. Data was collected from various internet websites and libraries of Council of Scientific and Industrial Research Institutes as CIMAP (Central Institutes of Medicinal and aromatic plant) Lucknow, CDRI (Central Drug Research Institute) Lucknow, NBRI (National Botanical Research Institute) Lucknow, NML (National Medical Library) New Delhi and Pharmacy Department, GGDU, Bilaspur, India. Various plant parts such as leaves, bark, stem and roots of *Ajuga bracteosa* have been used in ethno medicine to exploit its medicinal properties including astringent, hypoglycemia, gastrointestinal disorders, as anthelmintic, diuretic, antifungal, anti-inflammatory and antimycobacterial agents. Chemical compounds isolated from *Ajuga bracteosa* wall Ex. Benth have been proven to be pharmacological active against several major diseases including cancer, hypoglycemia and protozoal diseases. Preclinical studies indicate the therapeutic potential of crude extracts of *Ajuga bracteosa* wall Ex. Benth in the treatment of many microbial diseases, spasmodic activity and gastric ulcer. This review covers the pharmacological activities of some isolated chemical constituents of *Ajuga bracteosa* and preclinical studies on some crude extracts and pure compounds to explore novel bioactive compounds for therapeutic application.

Qing X., Yan H-M., Ni Z-Y., Vavricka C.J., Zhang M-L., Shi Q-S., Gu Y-C. and Kiyota H. (2017) Chemical and pharmacological research on the plants from the genus *Ajuga*. *Heterocyclic Communications* 23(4), 245-268.

Abstract: The genus *Ajuga*, a member of the Lamiaceae family, is comprised of more than 300 species of annual and perennial herbaceous flowering plants mainly distributed throughout the temperate regions of Asia, Europe, Australia, North America and Africa. These plants are used as folk medicines effective for rheumatic fevers, dysentery, malaria, hypertension, diabetes and gastrointestinal disorders, as well as anthelmintic, astringent, febrifuge diuretic, antifungal and anti-inflammatory agents. A variety of constituents has been isolated from these

plants. This review summarizes the phytochemical progress of the genus *Ajuga* and lists the compounds isolated up to 2014.

Qiu M. (1996?) Exploring the diversity of plant secondary metabolic products from phytoecdysones. Proceeding of a conference, sponsored by the Biodiversity Committee of the Chinese Academy of Sciences [all further information is in Chinese] 54-62. [in Chinese, with an Abstract in English].

Qiu M. and Nie R. (1987) Interrelations of phytoecdysones on their chemical structure in same plant and their biosynthetic pathway in plant. *Acta Botanica Yunnanica* 9(1) 119-128 [in Chinese, with English abstract].

Abstract: In this review, in nine plants the interrelations of phytoecdysones on their chemical structure and their biosynthetic pathway have been discussed. We propose the general character of C27, C28, C29 phytoecdysones in their biosynthetic processes, which based on the biogenesis of C27 phytoecdysones was expounded, and consider the common biogenetic precursor existed, and suggest the biosynthetic mechanism of some special phytoecdysones, and predict some special new phytoecdysones (Fig. 9) possibly existed in the special plant.

Qui M. and Nie R. (1988) The dehydration of ajugasterone C under base conditions. *Acta Botanica Yunnanica* 10(2), 219-222 [in Chinese, with English abstract].

Abstract: The base-catalyzed dehydration of hydroxyl compounds is unusually studied in dehydration reactions. A special phytoecdysone, carrying a α -hydroxyl group on C11-position, ajugasterone C on treatment with 5% KOH-MeOH solution, eliminated a H₂O, then afforded a product dactryhainansterone (IV) in over 80% yield at room temperature.

Ramazanov N.S. (2005) Phytoecdysteroids and other biologically active compounds from plants of the genus *Ajuga*. *Chemistry of Natural Compounds* 41(4), 361-369 [in English]/*Khimiya Prirodnykh Soedinenii* (4), 293-299 [in Russian].

Abstract: Literature data on the structures of phytoecdysteroids and other biologically active compounds and their biological activities were reviewed.

Ramazanov N.S., Bolaev I.D., Syrov V.N., Serdyllaev X.X. and Mamatchanov A.Y. (2016) *Chemistry, Biology and Technology of Phytoecdysteroids*. Fan va Texnologiya, Tashkent, pp. 260 (ISBN 978-9943-4679-2-7) [book in Russian].

Rates S.M.K. and Gosmann G. (2002) *Pfaffia*: available chemical and pharmacological data and their implications for its therapeutical use. *Revista Brasileira de Farmacognosia* 12(2), 85-93 [in Portuguese, with an English Abstract].

Abstract: This review summarizes available chemical and pharmacological data about *Pfaffia* genre. Scientific studies about these species are still lacking, and their therapeutical rational use depends on the availability of more informations about their pharmacological properties and the development of production and control quality technologies.

Repyakh S.M., Yushkova E.V., Velichko N.A. (1996) The effect of chemical and physical factors on the growth and development of callus tissues of *Rhaponticum carthamoides*. *Biotekhnologiya* (8), 45-49 [in Russian]/*Russian Biotechnology* (8), 45-49 [in English].

Rharrabe K., Alla S., Maria A., Sayah F. and Lafont R. (2007) Diversity of detoxification pathways of ingested ecdysteroids among phytophagous insects. *Archives of Insect Biochemistry and Physiology* 65, 65-73.

Abstract: The metabolic pathways of ingested ecdysteroids have been investigated in three insect species, the aphid *Myzus persicae* and two Lepidoptera, *Plodia interpunctella* and *Ostrinia nubilalis*. *M. persicae* produces mainly a 22-glucoside conjugate, whereas *P. interpunctella* eliminates a mixture of 20E and its 3-oxo and 3-epi derivatives, both in free form and as conjugates with various fatty acids. *O. nubilalis* only produces fatty acyl ester conjugates. These data point out the great diversity of detoxification mechanisms used by phytophagous insects in order to overcome the potential harmful effects of ecdysteroids present in their food.

Rimpler H. (1970) Phytoecdysone, Insektenhäutungshormone aus Pflanzen [Phytoecdysones, insect moulting hormones from plants]. *Preparative Pharmazie* 6(6), 101-108 [in German]

Robbins W.E., Kaplanis J.N., Thompson M.J., Shortino T.J., Cohen C.F. and Joyner S.C. (1968) Ecdysone and analogs: effects on development and reproduction of insects. *Science* 161, 1158-1160.

Abstract: Ingestion of certain synthetic ecdysone analogs inhibited larval growth and development in several species of insects, whereas 20-hydroxyecdysone was inactive or considerably less active. Natural 20-hydroxyecdysone and

ponasterone A, and a synthetic ecdysone analog inhibited ovarian maturation and egg production in the adult housefly. These effects appeared to be related to hormonal activity.

Robins R.J. (1998) The application of root cultures to problems of biological chemistry. *Natural Product Reports* 549-570.

Roy S., Dutta A.K. and Chakraborty D.P. (1982) Amasterol, an ecdysone precursor and a growth inhibitor from *Amaranthus viridis*. *Phytochemistry* 21, 2417-2420.

Abstract: A sterol isolated from the roots of *Amaranthus viridis* has been assigned the structure 24-methylene-20-hydroxycholest-5,7-en-3 beta -ol. The compound (amasterol) had allelopathic effects on lettuce seed germination.

Russell J. and Cohn R. (Eds.) (2012) 20-Hydroxyecdysone. Bookvika Publishing (Book on Demand Ltd.) ISBN-13: 978-5510990621, pp. 17.

Saad M.I., Kovalenko P.G., Zayets B.N., Korniets G.V., Shatursky Y.P., Kholodova Y.D. and Galkin A.P. (1992) A comparative analysis of proteins in the cell culture and in field plants of *Serratula coronata* L., a producer of ecdysteroids. *Ukrainian Biochemical Journal* 64(6), 84-86 [in Russian, with English abstract].

Abstract: Differences in the composition and amount of proteins synthesized in the cell culture and leaves of field plants *Serratula coronata* have been shown. They proceed from differences in intensity of synthesis of secondary metabolites, ecdysteroids, whose content in the cell culture is considerably lower.

Saatov Z., Agzamkhodzhaeva D.A. and Syrov V.N. (1999) Distribution of phytoecdysteroids in plants of Uzbekistan and the possibility of using drugs based on them in neurological practice. *Chemistry of Natural Compounds* 35(2), 186-191 [in English]/*Khimiya Prirodnikh Soedinenii* (2), 209-215 (1999) [in Russian].

Abstract: The distribution of phytoecdysteroids in plants of the families Labiatae, Compositae, and Caryophyllaceae growing in Uzbekistan has been analyzed. It has been shown that some of them possess a capacity for lowering the levels of urea and residual nitrogen in the blood and for improving the functional state of the kidneys in various pathological states. The dependence of biological activity on the structure of the compounds is discussed. The possibility of broadening the indications for the use of the drug ékdisten, the first to have been created from compounds of this class, in complications affecting the eyes of patients suffering from chronic glomerulonephritis has been substantiated experimentally and clinically.

Salehi B., Armstrong L., Rescigno A., Yeskaliyeva B., Seitimova G., Beyatli A., Sharmeen J., Mahomoodally M.F., Sharopov F., Durazzo A., Lucarini M., Santini A., Abenavoli L., Capasso R. and Sharifi-Rad J. (2019) *Lamium* plants – a comprehensive review on health benefits and biological activities. *Molecules* 24, 1913 (doi: 10.3390/molecules24101913).

Abstract: This work is an updated snapshot of *Lamium* plants and their biological activities. The main features of the plant are described and the components of its essential oils are summarized. The traditional medicinal uses of *Lamium* plants has been reported. The presence of these chemicals i.e., hydroxycinnamic acids, iridoids, secoiridoids, flavonoids, anthocyanins, phenylpropanoids, phytoecdysteroids, benzoxazinoids, betaine can provide biological activities. After the discussion of antioxidant properties documented for *Lamium* plants, the biological activities, studied using in vitro models, antimicrobial, antiviral, anti-inflammatory, anti-nociceptive activity, and pain therapy and cytotoxicity and cytoprotective activity are here described and discussed. Finally, targeted examples of in vivo studies are reported.

Sardini D. and Krepinsky J. (1974) La determinazione densitometrica degli ecdisoni [Densitometric determination of ecdysones]. *Il Farmaco Edizione Pratica* 29(12), 723-731 [in Italian].

Satoh H., Koshino H., Uzawa J. and Nakata T. (2003) CAST/CNMR: highly accurate C-13 NMR chemical shift prediction system considering stereochemistry. *Tetrahedron* 59(25), 4539-4547.

Abstract: Accurate, practical prediction of ¹³C NMR chemical shifts has been achieved with a new system, CAST/CNMR, taking account of stereochemistry. The CAST/CNMR system has solved the critical problem of the accurate distinction of differences and similarities in stereochemical structures around a specific carbon, which has not yet been achieved by any other database-oriented system for prediction of ¹³C NMR chemical shifts. CAST/CNMR uses a three-dimensional structural database together with a ¹³C NMR spectral database. Absolute/relative configurational and conformational structural information are described by the CAST (CAnonical-representation of STereochemistry) coding method. This paper provides an overview of the CAST/CNMR system, and describes its application to two natural products as examples.

Semwal P., Painuli S., Painuli K.M., Antika G., Tumer T.B., Thapliyal A., Setzer W.N., Martorell M., Alshehri M.M., Taheri Y., Dastan S.D., Ayatollahi S.A., Petkoska A.T., Sharifi-Rad J. And Cho W.C. (2021) *Diplazium esculentum* (Retz.) Sw.: ethnomedicinal, phytochemical, and pharmacological overview of the Himalayan ferns. *Oxidative Medicine and Cellular Longevity* 2021, article 1917890, pp15.

Sena Filho J.G., Durringer J., Maia G.L.A., Tavares J.F., Xavier H.S., Sobral da Silva M., da Cunha E.V.L. and Barbosa-Filho J.M. (2008) Ecdysteroids from *Vitex* species: distribution and compilation of their ¹³C-NMR spectral data. *Chemistry & Biodiversity* 5, 707-713.

Abstract: Iridoids and ecdysteroids are found in some genera of the family Verbenaceae. In such cases, they are used as chemotaxonomic markers for the difficult task of taxonomic identification by using morphological characteristics of plants belonging to this family. The present work describes the distribution of ecdysteroids in plants from the genus *Vitex* from a review of previous work on seventeen *Vitex* species. In addition, (13)C-NMR data of the main ecdysteroids found in this genus are described. This study attempted to summarize previous research on ecdysteroids distribution in *Vitex* species with the addition of (13)C-NMR analysis to further refine the characterization of these compounds in the Verbenaceae family.

Sharma P., Dwivedee B.P., Bisht D., Dash A.K. and Kumar D. (2019) The chemical constituents and diverse pharmacological performance of *Tinospora cordifolia*. *Heliyon* 5, e02437.

Abstract: *Tinospora cordifolia* is a popular medicinal plant which is used in several traditional medicines to cure various diseases. The common names are Amrita and Guduchi and belong to the family of Menispermaceae. It is considered an essential herbal plant of Indian system of medicine (ISM) and has been used in the treatment of fever, urinary problem, dysentery, skin diseases leprosy, diabetes, and many more diseases. The plant reported containing chemical compound including Alkaloids, Terpenoids, Lignans, Steroids and others that establish the phytochemistry and pharmacological activity of *Tinospora cordifolia*. The present review highlights the pharmacological importance viz antioxidant activity, antimicrobial activity, antibacterial activity, antifungal activity, anti-diabetic activity, antistress activity, hypolipidaemic effect, hepatic disorder, anticancer anti HIV potential, antiosteoporotic effects, antitoxic effects, wound healing, anticomplementary activity, and immunomodulating activity, systemic infection and Parkinson's disease.

Sharma V., Reddy K.R.C. and Singh Gautam DN. (2018) Phytochemistry and pharmacology of *Sida spinosa*. *International Journal of Green Pharmacy* 12 (Special Issue), S109-S115.

Abstract: *Sida spinosa* Linn. (Malvaceae) is an erect, perennial shrub found throughout the hotter parts of India. The tribes used this for the treatment of ulcers, pain, asthma, burning sensation, skin diseases, snake bite, gonorrhoea, diarrhoea, and dysentery. Phytochemical investigations indicate that 26 compounds reported from the plant belong to various chemical category, namely, aliphatics, ecdysteroids, alkaloids, steroid, and other compounds. Pharmacological activities of different parts of the plant reported include antioxidant, antipyretic, antidiabetic, antihyperlipidemic, antimicrobial, antiulcer, wound healing, and diuretic activity. In the present review, the literature data on the phytochemical and biological investigations on the *S. spinosa* are summarized up to December 2017.

Shikov A.N., Narkevich I.A., Flisyuk E.V., Luzhanin V.G. and Pozharitskaya O.N. (2021) Medicinal plants from the 14th edition of the Russian Pharmacopoeia, recent updates. *Journal of Ethnopharmacology* 268, article 113685 (doi: 10.1016/j.jep.2020.113685).

Abstract:

Ethnopharmacological relevance

Herbal medicine in Russia has a long history starting with handwritten herbalist manuscripts from the Middle Ages to the officinal Pharmacopoeia of the 21st century. The “herbophilous” Russian population has accumulated a lot of knowledge about the beneficial effects of local medicinal plants. Yet, for a long time, Russian traditional and officinal herbal medicine was not well known to the international audience. In our previous comprehensive review, we discussed the pharmacological effects of specific plants included in the 11th edition of the Pharmacopoeia of the USSR, which was also for a while used in Russia. The 14th edition of the Russian Federation's State Pharmacopoeia was implemented in 2018.

Aim of the review

The aims of the present review are: (i) to trace the evolution of medicinal plant handling from handwritten herbalist manuscripts to Pharmacopoeias; (ii) to describe the modern situation with regulatory documents for herbal medicinal products and their updated classification; (iii) to summarize and discuss the pharmacology, safety, and clinical data for new plants, which are included in the new edition of the Pharmacopoeia.

Methods

New medicinal plants included in the 14th edition of the Russian Federation's State Pharmacopoeia were selected. We carefully searched the scientific literature for data related to traditional use, pharmacological, clinical application,

and safety. The information was collected from local libraries in Saint-Petersburg, the online databases E-library.ru, Scopus, Web of Science, and the search engine Google scholar.

Results

Investigating the evolution of all medicinal plants referred to in the Russian Pharmacopoeias led us to the identification of ten medicinal plants that were present in all editions of civilian Russian Pharmacopoeias starting from 1778. In the 14th edition of the modern Russian Pharmacopoeia, medicinal plants are described in 107 monographs. Altogether, 25 new monographs were included in the 14th edition, and one monograph was excluded in comparison to the 11th edition. Some of the included plants are not endemic to Russia and do not have a history of traditional use, or on the other hand, are widely used in Western medicine. For 15 plants, we described the specificity of their application in Russian traditional medicine along with the claimed dosages and indications in officinal medicine. The pharmacology, safety, and clinical data are summarized and assessed for nine plants, underlining their therapeutic potential and significance for global phytopharmacotherapy.

Conclusions

In this review, we highlight the therapeutical potential of new plants included in the modern edition of the Russian Pharmacopoeia. We hope that these plants will play an imperative role in drug development and will have a priority for future detailed research.

Shivhare M.K., Singour P.K., Chaurasiya P.K. and Pawar R.S. (2012) *Trianthema portulacastrum* Linn. (Bishkhapra). *Pharmacognosy Reviews* 6(12), 132-140.

Abstract: World Health Organization (WHO) has recommended that traditional health and folk medicine systems are proved to be more effective in health problems worldwide. *Trianthema portulacastrum* Linn. is a herb used in Ayurvedic medicine. The principal constituent of *T. portulacastrum* Linn. is ecdysterone and the other constituents are trianthenol, 3-acetylaleuritic acid, 5,2'-dihydroxy-7-methoxy-6,8-dimethylflavone, leptorumol, 3,4-dimethoxy cinnamic acid, 5-hydroxy-2-methoxybenzaldehyde, p-methoxybenzoic acid, and beta cyanin. Different parts of *Trianthema portulacastrum* Linn. are traditionally used as analgesic, stomachic, laxative, treatment of blood disease, anemia, inflammation, and night blindness. Laboratory investigations on extracts of the plant have demonstrated significant pharmacological activities, such as antioxidant, diuretic, analgesic, hepatoprotective, and anticarcinogenic. This article compiles all updated information related to *T. portulacastrum* Linn. Scientifically proved activities are co-related with traditional concepts. Scientific evidence exists with respect to their major and minor constituents. The novelty and applicability of *T. portulacastrum* are hidden. Such things should be overcome through modern scientific concepts.

Singh D. and Chaudhuri P.K. (2017) Chemistry and pharmacology of *Tinospora cordifolia*. *Natural Product Communications* 12(2), 299-308.

Abstract: *Tinospora cordifolia* (Menispermaceae) is an Ayurvedic medicinal plant distributed throughout the Indian subcontinent and China. The whole plant is used in folk and the Ayurvedic system of medicine alone and in combination with other plants. Due to its commercial importance, *T. cordifolia* has been of intense research interest for the last four decades with the isolation of diverse compounds such as alkaloids, sesquiterpenoids, diterpenoids, phenolics, steroids, aliphatic compounds and polysaccharides, along with the discovery of a wide spectrum of pharmacological properties like immunomodulation, anticancer, hepatoprotective and hypoglycemic. Although pharmacological activities of extracts and compounds of *T. cordifolia* have been studied both in vitro and in vivo, only few mechanisms of action have been explored and need further elaboration. In the present review, the pharmacological activities of compounds and different extracts of *T. cordifolia* are highlighted, along with those of the marketed products, showing the relevance of phytochemicals and the standardization of the marketed products for medicinal use. This compilation of the extensive literature of *T. cordifolia* here will be a referral point for clinical study and the development of standardized phytomedicines in healthcare.

Singh S.S., Pandey S.C., Srivastava S., Gupta V.S., Patro B. and Ghosh A.C. (2003) Chemistry and medicinal properties of *Tinospora cordifolia* (Guduchi). *Indian Journal of Pharmacology* 35, 83-91.

Abstract: *Tinospora cordifolia* (Guduchi) is a widely used shrub in folk and ayurvedic systems of medicine. This review presents a detailed survey of the literature on chemistry and medicinal properties of *Tinospora cordifolia*. The chemical constituents reported from this shrub belong to different classes such as alkaloids, diterpenoid lactones, glycosides, steroids, sesquiterpenoid, phenolics, aliphatic compounds and polysaccharides. The notable medicinal properties reported are anti-diabetic, anti-periodic, anti-spasmodic, anti-inflammatory, anti-arthritic, anti-oxidant, anti-allergic, anti-stress, anti-leprotic, anti-malarial, hepatoprotective, immunomodulatory and anti-neoplastic activities.

Sinlaparaya D., Duanghaklang P and Panichajakul S. (2007) Enhancement of 20-hydroxyecdysone production in cell suspension culture of *Vitex glabrata* R.Br. by precursors feeding. *African Journal of Biotechnology* 6(14), 1639-1642.

Abstract: The effect of ecdysteroid precursors feeding on cell growth and 20-hydroxyecdysone production of *Vitex glabrata* suspension cultures were studied. On the addition of cholesterol, there was no apparent increase of 20-hydroxyecdysone while growth was partially inhibited at higher levels. Feeding of 7-dehydrocholesterol and ergosterol did not affect the cell growth. Both precursors effectively increased production of 20-hydroxyecdysone. Feeding of 7-dehydrocholesterol as a precursor was most effective. The maximum 20-hydroxyecdysone productivity of about 1.31 mg/L/day was observed in culture with 10 mg/L 7-dehydrocholesterol. This data is the first indication that 7-dehydrocholesterol and ergosterol feeding are effective precursors for 20-hydroxyecdysone formation in plant cell suspension culture.

Skala E., Grabkowska R., Sitarek P. Kuzma L., Blauz A. and Wysokinska H. (2015) *Rhaponticum carthamoides* regeneration through direct and indirect organogenesis, molecular profiles and secondary metabolite production. *Plant Cell Tiss Organ Cult* **123**, 83-98.

Abstract: The organogenic competence of different explants of *Rhaponticum carthamoides* was investigated on MS agar medium supplemented with BA, IBA or NAA at concentrations of 0.2 and 0.5 mg L⁻¹. Adventitious shoot formation was obtained through direct organogenesis using leaves of in vitro cultures as explants and through indirect organogenesis when seedling explants (hypocotyl, cotyledon and root) were used for regenerative callus initiation. The shoots were rooted on half-strength MS medium (½ MS) without auxin or containing IBA (0.2–2.0 mg L⁻¹). The plantlets regenerated through direct and indirect organogenesis were transferred into pots and grown in the greenhouse for 3 months. Significant differences in morphology, accumulation of chlorogenic acid and 20-hydroxyecdysone (20-HE) as well as in genetic profile were observed between these plants. UHPLC analysis showed that the highest level of chlorogenic acid (12 mg g⁻¹ DW) was found in leaves of plants developed directly from explants, whereas leaves of plants developed via callus tissue accumulated the highest amount of 20-HE (7.4 mg g⁻¹ DW). Its level exceeded that detected in leaves of 3-month-old plants obtained from seeds (2.4 mg g⁻¹ DW). Genetic variations of *R. carthamoides* regenerated plants were evaluated by flow cytometry and RAPD and ISSR methods. Flow cytometry confirmed similar ploidy level in the mother plant and plants regenerated through direct and indirect organogenesis. Genetic similarity values calculated on the basis of RAPD and ISSR data among regenerated in vitro plants to the mother plant were ranged from 0.765 to 0.941 and 0.647 to 0.947, respectively.

Skzypczak-Pietraszek E. and Grzybek J. (1997) Secondary metabolites of tissue cultures of Lamiaceae species. *Biotechnologia* (2), 93-110 [in Polish].

Sláma K. (1969) Plants as a source of materials with insect hormone activity. *Entomologia Experimentalis et Applicata* **12**, 721-728.

Abstract: Certain plants contain substances with insect hormone like activity. These hormone analogues may influence insect-plant interactions like some other factors such as nutritional requirements, chemoreception, toxicity, etc. Experimental evidence indicates that an external supply of the hormonal substances, which comes from the plant without respect to the physiological control mechanisms, disturbs the precise synchronization of insect development and leads to the appearance of malformed creatures unable to survive and reproduce. We distinguish two types of hormonally active plant substances; a) Those with insect juvenile hormone activity, which are mainly aliphatic or monocyclic sesquiterpenes of farnesane or bisabolane types, and b) Substances with moulting hormone activity, which are polyhydroxy-steroid compounds related to ecdysone. The isoprenoid compounds with juvenile hormone activity occur in various plants. One of the most common ecdysone analogues of plant origin seems to be the 20-hydroxy ecdysone or ecdysterone. Hormonal activity of the plant substances is similar to, or higher than that of ecdysone. Whole series of ecdysone analogues have been isolated from various unrelated plants. In some cases they occur in extremely large quantities. There are some indications that they do act when fed in the diet. Plants which are known to contain analogues of insect hormones are more or less resistant, especially against susceptible insects. It does not seem impossible that certain plants have interacted in their evolution with insect feeders by using hormonomimetic materials -- a method which we have not appreciated until now.

Sláma K. (1978) The principles of antihormone action in insects. *Acta Entomol. Bohemoslov.* **75**, 65-82.

Abstract: The possible consequences of antihormonal action on insect development have been calculated on the basis of the available information concerning the function of the neuroendocrine system. The effects have been classified as antineurotropic, antiadenotropic, antihormonal in the strict sense, and antihomeostatic, according to the expected location of the inhibition within the neuroendocrine action chain. It has been extremely difficult to identify the specifically antineuroendocrine nature of various developmental inhibitions, which can also be nonspecifically produced by general inhibitors of insect growth and development. Special emphasis is given to the effects of antifeeding due to their interference with the nutritional links involved in the activation of endocrine glands. Results of some experiments with compounds that cause antijuvenile-like prothelias in insects have been described.

Sláma K. (1987) Insect hormones and bioanalogs in plants. In: *Insects-Plants; Proceedings of the 6th International Symposium on Insect-Plant Relationships (Pau, 1986)* (Eds. Labeyrie V., Fabres G. and Lachaise D.) Dr. W. Junk Publishers, Dordrecht, pp. 9-16.

Sláma K. (1993) Ecdysteroids - insect hormones, plant defensive factors, or human medicine. *Phytoparasitica* 21(1), 3-8.

Abstract: The history of insect hormone research is closely associated with plants. The first such substance of known structure was the rather common plant constituent farnesol, which was thought for some time to be the true juvenile hormone of insects. Since then more than 4000 natural or synthetic mimics of juvenile hormone have been uncovered, some of them being 100 million times more effective than farnesol (14). As to the historical background of ecdysteroids, which are related to insect molting hormone: Kopeč discovered the presence of molt-inducing substances in insect brain in the 1920s; Bounhiol and others described the presence of a special thoracic developmental center for regulation.

Sláma K., Romanuk M. and Šorm F. (1974) Chemistry and physiology of ecdysoids. In: Sláma K., Romanuk M. and Šorm F., *Insect Hormones and Bioanalogs*. Springer Verlag, Wien, New York, pp 333-387.

Abstract: The history of the discovery of insect molting hormone began with papers by the Polish entomologist KOPEČ [172, 173] which demonstrated for the first time that the molting process was controlled by a humoral factor. Further significant contributions to the physiology of the molting process came from WIGGLESWORTH [323]. Of great importance for the chemistry of molting hormones was the observation that the molting process is controlled by a factor present in the prothoracic glands (FUJKUDA [68–70]). PLAGGE and BECKER [14, 234] discovered that molting hormone could be extracted from blowfly (*Calliphora erythrocephala*) larvae with boiling ethanol.

Speranza A. (2010) Into the world of steroids: a biochemical “keep in touch” in plants and animals. *Plant Signaling & Behavior* 5(8), 940-943.

Abstract: Evolution of steroids such as sex hormones and ecdysteroids occurred independently in animal and plant kingdoms. Plants use phytoecdysteroids (PEs) to control defence interactions with some predators; furthermore, PEs can exert beneficial influence on many aspects of mammalian metabolism. Endocrine disrupting compounds such as the estrogen agonist bisphenol A (BPA) are widespread in the environment, posing a potential hormonal risk to animals and plants. Adverse BPA effects on reproductive development and function are coupled with other toxic effects. BPA bioremediation techniques could be developed by exploiting some tolerant plant species.

Stránský K., Němec V. and Sláma K. (1998) Lipid composition of the seeds of an ecdysteroid-containing plant, *Leuzea carthamoides* (Willd.) D.C. (Asteraceae). *Russian Journal of Plant Physiology* 45(3), 333-338.

Abstract: Lipid extract of the seeds of *Leuzea carthamoides* (Willd.) DC (Asteraceae) (19.78% of the dry seeds) was separated by preparative TLC into 6 fractions: (I) Hydrocarbons, 0.063% of total lipid; (II) Sterol esters, 0.13%; (III) Triacylglycerols, 15.51%; (IV) Free fatty acids, 0.26%; (V) Free sterols, 0.66% and (VI) Polar lipids, 3.12%. Fractions I to V were further analyzed by GC. There was a homologous series of free fatty acids with maxima at C16:0 (palmitic), C18:1 ω 9 (oleic) and C18:2 ω 6 (linoleic) acids. The fatty acids obtained from sterol esters and triacylglycerols by transesterification showed a distribution spectrum similar to that of free fatty acids. C29 stigmastane-type sterols were the most abundant, β -sitosterol being the predominating free sterol (60%), followed by β -sitostanol (13.2%) and stigmasterol (4.94%). The esterified sterols were similar in their composition to the free sterols. There was a very small amount of C27 cholestane-type sterols (cholesterol) and C28 ergostane-type sterols (campesterol). The quite small amounts of free cholesterol (0.4% of free sterols; 0.0026% of dry seeds) have special significance in relation to the extremely high amounts of ecdysteroids (the content of 20-hydroxyecdysone in dry seeds was as high as 2%). 20-Hydroxyecdysone is a polyhydroxylated, partly water soluble derivative of cholesterol. Thus, paradoxically, the seeds of this plant contain almost three orders of magnitude (750-fold) more cholesterol hidden in this hydrophilic, polyhydroxylated form. The proportions between the ecdysteroid-bound and the free sterols have been completely reversed in the case of C29 stigmastane-type sterols. The corresponding C29 ecdysteroid, makisterone C, is present only as 0.02% of the dry seeds, which is 26 times less than free C29 sterols. These facts provide evidence that cholesterol is preferentially hydroxylated by plant tissues, which results in its selective removal from the pool of lipid-soluble, free phytosterol.

Sukhanov A.E., Buyuklinskaya O.V. and Koptyaeva R.G. (2017) Steroid connections of the phytogenesis: phytoecdysones (phytoecdysteroids). *Scientometric research of data of scientific and practical literature*. Belgorod State University Scientific Bulletin: Medicine, Pharmacy. 40(26), 123-136 [in Russian, with an Abstract in English].

Swevers L., Kravariti L., Ciolfi S., Xenou-Kokoletsi M., Ragoussis N., Smagghe G., Nakagawa Y., Mazomenos B. and Iatrou K. (2004) A cell-based high-throughput screening system for detecting ecdysteroid agonists and

antagonists in plant extracts and libraries of synthetic compounds. FASEB Journal 18, 134-136 (summary of on-line publication: <http://www.fasebj.org/cgi/doi/10.1096/fj.03-0627fje>).

Abstract: Screening systems for ecdysteroid mimetic or antiecdysteroid substances in plant extracts or libraries of synthetic compounds are commonly based on the observation of morphological and/or growth responses in insect cell lines. Because these responses are slow and require careful monitoring, existing screening systems are considered limited regarding their applicability to analysis in high-throughput (HT) formats. Here we describe the generation of transformed silkworm (*Bombyx mori*) cell lines that respond to the addition of ecdysone-like substances through the expression of the green fluorescent protein (GFP) and the appearance of green fluorescence. Because tests consist of three simple steps, i.e., 1) distribution of transformed cells in microtiter plates; 2) addition of compounds/extracts at different concentrations; and 3) quantification of fluorescence intensity by a fluorescence plate reader, they can be performed quickly and be easily adapted to a HT format. The generated reporter cell lines are used for the screening of extracts from available plant collections for the presence of compounds with ecdysone mimetic or antagonistic activities as well as for monitoring subsequent activity during enrichment and purification steps. The same cell lines are also used here for the determination of structure-activity relationships among available synthetic dibenzoylhydrazine derivatives. Finally, for the identified agonists, we show that their activity as determined by the cell-based screening assays parallels their bioactivity in growth inhibition and toxicity assays carried out on live insects.

Tabasum A., Zahida S. and Rabiah B. (2019) A review on phytochemical and ethnopharmacological studies of *Ajuga bracteosa* Wall. ex Benth. Journal of Drug Delivery and Therapeutics 9(2), 489-492.

Abstract: Herbal medicines as the major remedy in traditional system of medicine have been used in medical practices since antiquity. The plants of genus *Ajuga* are evergreen, clump-forming rhizomatous perennial or annual herbaceous flowering species, with *Ajuga* being one of the 266 genera of the family Lamiaceae. There are at least 301 species of the genus *Ajuga* with many variations. *Ajugabracteosa* Wall. ex Benth (*A. bracteosa*) is an important medicinal plant of Himalaya regions. Medicinal potential is due to presence of various pharmacologically active compounds such as neo-clerodane diterpenoids, flavonol glycosides, iridoid glycosides, ergosterol-5,8- endoperoxide and phytoecdysones. The aim of this review article was to gather information about *A. bracteosa* which is currently scattered in form of various publications. This review article tried to attract the attention from people for therapeutic potential of *A. bracteosa*. The present review comprises upto date information of, traditional uses, botanical aspects, active ingredients and pharmacological activities such as antitumor, antimicrobial, antimalarial, anti-inflammatory, cardiotoxic activity, antiarthritic activity, antioxidant activity . A large variety of compounds have so far been isolated from *Ajuga bracteosa*.

Takahashi K., Hashimoto K., Fujiyama A., Yamada J., Kobayashi N., Morisaki M., Nakanao S., Hara N. and Fujimoto Y. (2003) Stereochemistry of the C-24-,25 double bond in the conversion of desmosterol into cholesterol. Tetrahedron Letters 44, 341-344.

Abstract: Feeding of the chemically prepared [24-¹³C, 24-²H]desmosterol to cell-free systems derived from rat liver and silkworm gut and to cultured cells of *Oryza sativa* followed by deuterium-decoupled ¹H, ¹³C shift correlation NMR analysis of the biosynthesized cholesterol revealed the stereospecific incorporation of hydrogen atoms from the *re*-face of the C-24 position of desmosterol.

Takemoto T., Ogawa S. and Nishimoto N. (1967) Studies on the constituents of *Achyranthis radix*. I. Oleanolic acid bisdesmoside from the root. Yakugaku Zasshi 87(12) 1463-1468 [in Japanese, with an English abstract].

Abstract: A new triterpenoidal saponin was isolated from the methanol extract of *Achyranthes fauriei* roots (Amaranthaceae). The structure of this saponin was elucidated as 3-β-D-glucopyranosyl-olean-12-en-28-O-β-D-glucopyranosyl ester.

Tarkowska D. And Strnad M. (2016) Plant ecdysteroids: plant sterols with intriguing distributions, biological effects and relations to plant hormones. Planta (DOI: 10.1007/s00425-016-2561-z).

Abstract: The present review summarises current knowledge of phytoecdysteroids' biosynthesis, distribution within plants, biological importance and relations to plant hormones. Plant ecdysteroids (phytoecdysteroids) are natural polyhydroxylated compounds that have a four-ringed skeleton, usually composed of either 27 carbon atoms or 28-29 carbon atoms (biosynthetically derived from cholesterol or other plant sterols, respectively). Their physiological roles in plants have not yet been confirmed and their occurrence is not universal. Nevertheless, they are present at high concentrations in various plant species, including commonly consumed vegetables, and have a broad spectrum of pharmacological and medicinal properties in mammals, including hepatoprotective and hypoglycaemic effects, and anabolic effects on skeletal muscle, without androgenic side-effects. Furthermore, phytoecdysteroids can enhance stress resistance by promoting vitality and enhancing physical performance; thus, they are considered adaptogens. This review summarises current knowledge of phytoecdysteroids' biosynthesis, distribution within plants, biological importance and relations to plant hormones.

Thiem B., Kikowska M., Malinski M.P., Kruszka D., Napierala M. and Florek E. (2017) Ecdysteroids: production in plant in vitro cultures. *Phytochemical Reviews* **16**, 603-622 (DOI: 10.1007/s11101-016-9483-z).

Abstract: Ecdysteroids are secondary metabolites, widely distributed in the animal and plant kingdoms. They have a wide range of pharmacological effects in vertebrates, including mammals, most of which are beneficial for humans. Therefore, they have become compounds of interest for the pharmaceutical industry due to their adaptogenic, anabolic, hypoglycaemic, hypocholesterolaemic and antimicrobial activities, which are still being researched. Nowadays, ecdysteroids are present as active ingredients in bodybuilding supplements. Because of their complex structures, their chemical synthesis seems unprofitable and impractical. Due to high content of ecdysteroids in many plants, they are primarily obtained by extraction of the plant material. Plant in vitro cultures provide an alternative source of these compounds, helping to avoid problems associated with field production—such as variable yield or dependence on environmental factors, as well as limited availability of natural resources. Plant cell and tissue cultures may be suggested as alternatives for the production of plant biomass rich in pharmaceutically active ecdysteroids. Moreover, the use of common biotechnological strategies, such as elicitation or precursor feeding, may further increase the yield and improve production of these compounds. In this paper, we describe general information about ecdysteroids: their structure, biosynthesis, distribution, role in plants, and we review recent studies on micropropagation of ecdysteroid-producing plants and cell cultures, and potential ability of ecdysteroids enhancement in in vitro cultures.

Timofeev N.P. (2007a) Achievements and problems in investigation of biology in medicinal herbs of *Rhaponticum carthamoides* (Willd.) Ilijin and *Serratula coronata* L. *Agricultural Biology* (3), 3-17 [in Russian, with an English abstract].

Timofeev N.P. (2007b) Industrial ecdysteroid sources. Part II. Ecdysterone: plants from the genera *Rhaponticum*. *Collection of Scientific Papers, Moscow* **15**, 8-49 [in Russian].

Tom W-M., Abul-Hajj Y.J. and Koreeda M. (1975) Microbial oxidation of ecdysones. A convenient preparation of rubrosterone. *Journal of the Chemical Society, Chemical Communications* 24-25.

Abstract: Ponasterone A(I) is converted by the microorganism *Fusarium lini* ATCC 9593 into rubrosterone (IV) in 15% yield.

Tomas-Barberan F.A. (1995) Capillary electrophoresis: a new technique in the analysis of plant secondary metabolites. *Phytochemical Analysis* **6**(4), 177-192.

Abstract: The possibilities and applications of capillary electrophoresis (CE) as an analytical technique for plant secondary metabolites are reviewed. Applications of both capillary zone electrophoresis and micellar electrokinetic capillary chromatography for this purpose are described. CE is compared with high pressure liquid chromatography (HPLC) as an analytical technique, and the advantages and criticisms of CE are described. The effects of the applied voltage, capillary temperature, electrolyte concentration and nature (complexing or non-complexing buffers), buffer pH, micelle concentration and nature (sodium dodecyl sulphate or cetyltrimethylammonium bromide) and the addition of organic modifiers to the running buffer (organic solvents, cyclodextrins, urea, cholate) on the parameters of separation and resolution of different secondary metabolites are discussed. The applications of CE to the analysis of flavonoid aglycones and glycosides, phenolic acids, quinones, coumarins, alkaloids, capsaicinoids, glucosinolates, polyamines, monoterpenes, diterpenes, phytoecdysteroids, cardiac glycosides and saponins are shown. It is concluded that CE is a very promising analytical technique in the analysis of plant secondary metabolites, and it will become an indispensable tool, together with HPLC and gas liquid chromatography, in phytochemical laboratories since these techniques are in many ways complementary, and problems that are difficult to solve by HPLC can often be solved using CE.

Volodin V.V., Chadin I.F., Dinan L. and Lafont R. (2004) Phytoecdysteroids - plant analogues of insect moulting hormones. *Rastitelnye Resursy* **40**(2), 1-18 [in Russian, with an English abstract].

Volodin V.V. and Volodina S.O. (2015) Floristic and molecular phylogenetic analysis of the distribution of phytoecdysteroids among plants of North-east Russia (Asteraceae and Caryophyllaceae). *Biology & Medicine* **7**(1), Article ID: BM-064-15, pp. 23.

Abstract: The review is devoted to analysis of the exploration degree of the world flora for the presence of phytoecdysteroids which are plant analogs of molting and metamorphosis hormones in insects. The results of long-term screening of the flora of the European North-East of Russia for ecdysteroids presence are summarized. The highest number of ecdysteroid-containing species has been found in the families Asteraceae and Caryophyllaceae belonging to the leading families of the flora. Combination of floristic and molecular-phylogenetic approaches has allowed to prove phylogenetic relations between producers on the intrafamilial level. The belonging of a

considerable number of the species to the southern and polyzonal latitudinal groups testifies to the existence of an ecological-geographic factor determining the distribution of ecdysteroids in the world flora. An algorithm of chemotaxonomic forecast of phytoecdysteroids discovering in unstudied regional floras is proposed.

Wang Q.J., Zheng L.P., Zhao P.F., Zhao Y.L. and Wang J.W. (2014) Cloning and characterization of an elicitor-responsive gene encoding 3-hydroxy-3-methylglutaryl coenzyme A reductase involved in 20-hydroxyecdysone production in cell cultures of *Cyanotis arachnoidea*. *Plant Physiology and Biochemistry* 84, 1-9.

Abstract: *Cyanotis arachnoidea* contains a rich source of bioactive phytoecdysteroids (i.e. analogues of insect steroid hormones). 3-Hydroxy-3-methylglutaryl coenzyme A reductase (HMGR) supplies mevalonate for the synthesis of many secondary metabolites including 20-hydroxyecdysone (20E), one of metabolism-enhancing phytoecdysteroids. In this study, in order to develop a sustainable source of 20E, cell suspension cultures were established from shoot cultures of *C. arachnoidea*, and a full length cDNA encoding HMGR (designated as CaHMGR) was cloned and characterized. The cDNA contained 2037 nucleotides with a complete open reading frame (ORF) of 1800 nucleotides, which was predicted to encode a peptide of 599 amino acids. Expression analysis by real-time PCR revealed that CaHMGR mRNA was abundant in *C. arachnoidea* stems, roots and leaves. When cultivated in Murashige & Skoog medium supplemented with 0.2 mg L⁻¹ 1-naphthylacetic acid (NAA) and 3.0 mg L⁻¹ 6-benzyladenine (6-BA), *C. arachnoidea* cells in suspension culture grew rapidly, yielding 20E (124.14 µg L⁻¹) after 12 days. The content of 20E in cell cultures elicited by 0.2 mM methyl jasmonate (MeJA), 100 mg L⁻¹ yeast elicitor (YE) or 25 µM AgNO₃ was increased 8-, 2-, and 6-fold over the control, respectively. Quantitative real-time PCR analysis showed that CaHMGR was expressed at a higher level under the treatment of MeJA or Ag⁺ elicitor. Our results suggested that 20E accumulation may be the result of the expression up-regulation of CaHMGR involved in the biosynthesis under the treatment of various elicitors.

Wikipedia (2007) Phytoecdysteroids. <http://en.wikipedia.org/wiki/Phytoecdysteroids>

Williams C.M. (1970) Hormone interactions between plants and animals. In: *Chemical Ecology*, pp. 103-132.

Williams D.H., Stone M.J., Hauck P.R. and Rahman S.K. (1989) Why are secondary metabolites (natural products) biosynthesized? *Journal of Natural Products* 52(6), 1189-1208.

Abstract: We adopt the definition of a natural product as a substance that has no known role in the internal economy of the producing organism. The literature abounds with conflicting views for the existence of such natural products. We propose that all such structures serve the producing organisms by improving their survival fitness. We argue that this conclusion is necessitated by the fact that natural products are normally complex structures, whose biosynthesis is programmed by many kilobases of DNA. If it were otherwise, the pressures of Darwinian natural selection would have precluded the expenditure of so much metabolic energy in their construction and the development of such complexity. We further conclude that a natural product improves the producer's survival fitness by acting at specific receptors in competing organisms. Current studies of natural products interacting with receptors support this view, in terms of both the sophistication of the molecule/molecule recognition and the mechanistic details of physiological action. By the application of Occam's razor and general weaknesses of other hypotheses, these other hypotheses are rejected. It is a consequence of our proposal that natural product/receptor interactions of sophistication comparable to enzyme/substrate interactions will be commonplace. Additionally, structures that are candidates to interact with known receptors (e.g., double helical DNA) can on occasion be suggested by inspection of the structures. A range of evidence to support the general conclusions is presented.

Yagi T., Morisaki M., Kushiro T., Yoshida H. and Fujimoto Y. (1996) Biosynthesis of 24 β -alkyl- Δ^{25} -sterols in hairy roots of *Ajuga reptans* var. *atropurpurea*. *Phytochemistry* 41(4), 1057-1064.

Abstract: A hairy root culture of *Ajuga reptans* var. *atropurpurea* contains clerosterol, 22-dehydroclerosterol and cholesterol as its sterol constituents. Feeding of [26-, 27-¹³C₂]desmosterol to this culture and ¹³C NMR analysis of the resulting biosynthesized sterols showed that the substrate was efficiently incorporated into clerosterol and codisterol. Feeding of [26-¹³C] and [27-¹³C]desmosterols revealed that the C-24 alkylation takes place in a highly specific manner wherein the 26- and 27-methyl groups of the substrate becomes C-26 (vinyl methyl) and C-27 (exomethylene carbon), respectively, of the two Δ^{25} -sterols. Further, feeding of [24-²H]desmosterol and ²H NMR analysis of the products showed that H-24 of clerosterol and codisterol is derived from H-24 of desmosterol. Finally, [28-¹³C]ergosta-5,24(28)-dien-3 β -ol was shown to be converted into clerosterol and 22-dehydroclerosterol, but not into codisterol. On the basis of these data, possible biosynthetic mechanism of 24 β -alkyl- Δ^{25} -sterols in this plant is proposed.

Yance D.R. and Sky S.E. (2018) Ecdysterone-rich botanicals that promote anabolic metabolism and metabolic homeostasis. *Natura Health Products*, www.NaturaEDU.com, pp8.

No Abstract.

Yang L., Yan M-L., Xing X-D., Guo X-Y., Jiang H., Yang B-Y., Chan K., Wang Q-H. and Kuang H-X. (2019) Investigation of Radix Achyranthis Bidentatae phytochemistry and pharmacology. World Journal of Traditional Chinese Medicine 5(1), 50-60.

Abstract: Radix achyranthis bidentatae (RAB), a member of the Amaranthaceae family, has been widely used in Traditional Chinese Medicine for 1000s of years. Increasing interest in RAB-derived medicinal has led to the discovery of additional triterpenoid saponins, phytoecdysones, polysaccharides, and many other compounds, as well as investigations into their pharmacology. A large number of pharmacological studies have shown RAB and its active components possess a range of pharmacological activities, including anti-tumor, anti-fertility, anti-senile, and anti-inflammatory effects. This review is an up-to-date summary and synthesis of the uses of RAB from phytochemical and pharmacological perspectives.

Yen K-y., Yang L-l., Tom W-m. and Tam W-s. (1971) Studies on physiologically active phytohormones of ferns in Taiwan. I. Preliminary tests on 100 kinds of ferns. Journal of the Taiwan Pharmaceutical association 23 (1), 83-92. No Abstract.

Yordanova Z.P., Zhiponova M.K., Iakimova E.T., Dimitrova M.A. and Kapchina-Toteva V.M. (2014) Revealing the reviving secret of the white dead nettle (*Lamium album* L.). Phytochemical Reviews 13, 375-389.

Abstract: *Lamium album*, commonly known as white dead nettle or non-stinging nettle is a flowering herbaceous plant, native throughout Europe, Western Asia and North Africa. From ancient times this plant has been endowed with revival, curative and culinary virtues. In the past, in the traditional and folk medicine white dead nettle has been used mainly for its anti-inflammatory, astringent and anti-septic activity. Nowadays significant amount of knowledge on the efficacy of extracts and raw material of *L. album* is accumulated and a number of health-related beneficial activities have been scientifically proven. In vitro analyses conducted in various model systems have demonstrated antiviral, antimicrobial, antioxidant, anticancer, cytoprotective, wound healing and other important pharmacological effects. The present review summarizes the recent information on the phytochemical features of this pharmacologically important species. The findings on the chemical composition, biological activities and the pharmacological properties underlying the revival secret of white dead nettle are described and discussed in the view of potential applications for treatment of human diseases. Trends for further research are outlined.

Youssif K.A., Elshamy A., Rabeh M.A., Gabr N.M. and Haggag E.G. (2019) A phytochemical and biological review on plants of the Family Aizoaceae. Journal of Advanced Pharmacy Research 3(4), 158-181.

Abstract:

Objectives: This study is aimed to be a comprehensive review of the phytochemical constituents and biological activities of Aizoaceae family plants (Mesembryanthemaceae). **Methods:** This study is covering articles between 1969 and 2018, reviewed from internationally accepted databases and scientific data from scientific Journals. **Results:** Phytochemically studied plants of family Aizoaceae have shown the presence of various classes of compounds including; alkaloids, triterpenes, sterols, lignans, phenolic compounds, betacyanins, and essential oils. Biological studies on plants of family Aizoaceae have indicated various bioactive potentials including antioxidant, antidiabetic, antimicrobial, antitumor, hepatoprotective, anti-inflammatory and other effects. The reported medicinal plants of family Aizoaceae were selected and summarized on the basis of their; phytochemical constituents and biological activities. **Conclusion:** The results of this study may inspire further ethno-botanical and ethno-pharmacological research and investigations toward drug discovery.

Zalavadiya V.I., Shah V.K., Santani D.D., Patel M.S., Fosi J.M. and Chaudhary A.K. (2013) *Achyranthes aspera* – plant with high medicinal importance. Research Journal of Pharmacology and Pharmacodynamics 5(4), ISSN online 2321-5836.

Abstract: *Achyranthes aspera* is a common plant found throughout India. Before the introduction of modern medicines, disease treatment was entirely managed by herbal remedies. It is estimated that about 80% of the world population residing in the vast rural areas of the developing and under developed countries still depends mainly on medicinal plants. It is quite obvious that the plant is widely used in traditional medicinal system of India and has been reported to possess hepatoprotective, anti-inflammatory, antitussive, antifungal and also used to check wounds healing and antibacterial properties. *Achyranthes aspera* is a very important plant for its large number of medicinal properties as well as medicinally important chemicals like ecdysterone, achyranthine, betaine, pentatriaontane, 6-pentatriacontanone, hexatriacontane and tritriacontane. The plant shows many pharmacological activities like, anti-allergic, cardiovascular, nephroprotective, antiparasitic, hypoglycemic, analgesic and antipyretic. The present article gives an account of updated information on its phytochemical and pharmacological properties. The review reveals that wide numbers of phytochemical constituents have been isolated from the plant which possesses activities like antiperiodic, diuretic, purgative, laxative, antiasthmatic, hepatoprotective, anti-allergic and various other important

medicinal properties. For the last few decades or so, extensive research work has been done to prove its biological activities and pharmacology of its extracts.

Zhou N., Xu L., Park S-M., Ma M-G., Choi S-E. and Si C. (2021) Genetic diversity, chemical components, and property of biomass *Paris polyphylla* var. *yunnanensis*. *Frontiers in Bioengineering and Biotechnology* **9**, article 713860 (<https://doi.org/10.3389/fbioe.2021.713860>).

Abstract: *Paris polyphylla* var. *yunnanensis* is a kind of biomass resource, which has important medicinal and economical values with a huge market. This review article aims to summarize the recent development of biomass *P. polyphylla* var. *yunnanensis*. The genetic diversity and chemical components of biomass *P. polyphylla* var. *yunnanensis* were reviewed based on the literature. Both the genetic diversity and genetic structure of biomass *P. polyphylla* var. *yunnanensis* were compared by using molecular marker technologies. All the extraction processes, harvest time, and drying methods on the chemical components were summarized in detail. The differences of arbuscular mycorrhizal fungi on the infection rate, diosgenin content, microorganisms, enzyme activities, rhizospheric environment, and endogenous hormones were discussed. This review article is beneficial for the applications of biomass *P. polyphylla* var. *yunnanensis* as a biomass resource in the biomedical field.

Zibareva L.N. (2009) Phytoecdysteroids of Caryophyllaceae Juss. *Contemporary Problems of Ecology* **2**(5), 476-488 [in English]/*Sibirski Ecologicheskii Zhurnal* **16**(5), 753-764 [in Russian].

Abstract: Distribution of phytoecdysteroids is analyzed in the family Caryophyllaceae, which includes a large number of ecdysteroid-containing species. Species of this family synthesize a wide variety of steroid compounds, insect molting and metamorphosis hormones, many of which are not found in other plants. Most of the species of Caryophyllaceae are characterized by high levels of phytoecdysteroids. The flowers of the perennial *Silene frivaldszkyana* accumulate up to 7% 20-hydroxyecdysone.

2. ECDYSTEROID(-RELATED) COMPOUNDS IN ALGAE

Báthori M., Blunden G. and Kalász H. (2000) Two-dimensional thin-layer chromatography of plant ecdysteroids. *Chromatographia* **52**(11/12), 815-817.

Abstract: Two-dimensional thin-layer chromatography has been used to screen plant samples for ecdysteroids. Satisfactory separations were obtained even in the presence of substantial impurities and for mixtures containing many ecdysteroids. Normal-phase silica and octadecyl silica plates were used to differentiate apolar and polar ecdysteroids, respectively. The major advantages of thin-layer chromatography are multiple detection, which enables specific detection of the ecdysteroids. Use of a small amount of water in the mobile phase in the first dimension, the water-free mobile phase in the second dimension, can afford favorable separations.

Dziwornu G.A., Caira M.R., de la Mare J-A., Edkins A.L., Bolton J.J., Beukes D.R. and Sunassee S.N. (2017) Isolation, characterization and antiproliferative activity of new metabolites from the South African endemic red algal species *Laurencia alfredensis*. *Molecules* **22**, 513 (doi: 10.3390/molecules22040513).

Abstract: The marine red algae of the genus *Laurencia* have been widely studied for their structurally diverse and biologically active secondary metabolites. We report here the natural product investigation of the organic extract of a newly identified South African endemic species, *Laurencia alfredensis*. A sequence of column chromatography, preparative TLC and normal phase HPLC resulted in the isolation of eleven compounds comprising three labdane-type diterpenes (**1-3**), four polyether triterpenes (**4-7**), three cholestane-type ecdysteroids (**8-10**) and a glycolipid (**11**). Compounds **1-3**, **5-8** and **10** have not previously been reported, while compound **9** is reported here for the first time from a natural source and the known compound **11** isolated for the first time from the genus *Laurencia*. The structural elucidation and the relative configuration assignments of the compounds were accomplished by extensive use of 1D- and 2D-NMR, HR-ESI-MS, UV and IR spectroscopic techniques, while the absolute configuration of compound **1** was determined by single-crystal X-ray diffraction analysis. All compounds were evaluated against the MDA-MB-231 breast and HeLa cervical cancer cell lines. Compound **2** exhibited low micromolar antiproliferative activity ($IC_{50} = 9.3 \mu M$) against the triple negative breast carcinoma and compound **7** was similarly active ($IC_{50} = 8.8 \mu M$) against the cervical cancer cell line.

Fukuzawa, A., Kumagai, Y., Masamune, T., Furusaki, A., Katayama, C. and Matsumoto, T. (1981) Acetylpinnasterol and pinnasterol, ecdysone-like metabolites from the marine red alga *Laurencia pinnata* Yamada. *Tetrahedron Letters* **22**(41), 4085-4086.

Abstract: The structures of two sterols, isolated from the title alga and designated as acetylpinnasterol and pinnasterol, were determined on the basis of the X-ray crystallography. These metabolites are the first marine phytosterols with ecdysone-like structures and biological activity as moulting hormones.

Fukuzawa A., Miyamoto M, Kumagai Y and Masamune T. (1986) Ecdysone-like metabolites, 14 α -hydroxypinnasterols from the red alga *Laurencia pinnata*. *Phytochemistry* **25**(6), 1305-1307.

Abstract: Four steroids with moulting hormone activity were isolated from *Laurencia pinnata*. These steroids are related structurally to β -ecdysone.

Kalász H., Báthori M. and Csermely T. (2000) Planar versus microcolumn chromatography. *American Laboratory* **32**(19), 28-31.

3. ECDYSTEROID(-RELATED) COMPOUNDS IN FUNGI AND ASSOCIATED REFERENCES

Canonica L., Danieli B., Palmisano G., Rainoldi G. and Ranzi B.M. (1974) The microbiological oxidation of insect moulting hormones. *Journal of the Chemical Society, Chemical Communications* 656-657.

Abstract: Crustecdysone (I) is degraded by a lysed mycelium of the micro-organisms *Rhizopus arrhizus*, *Rhizopus nigricans* and *Curvularia lunata* to give poststerone (VI) which is then transformed into rubrosterone (VIII); makisterone A (II) behaves similarly, muristerone A (III) gives a new product, identified as 5 β ,11 α -dihydroxy-poststerone (VII), and ecdysone (IV) is not affected by these micro-organisms.

Danieli B., Lesma G. and Luisetti M. (1997) *Candida antarctica* lipase B catalyzes the regioselective esterification of ecdysteroids at the C-2 OH. *Tetrahedron* **53**(16), 5855-5862.

Immobilized *Candida antarctica* lipase B (Novozym 435) catalyzes the regioselective acylation of 20R-hydroxyecdysone (**1a**) and its congeners **2a**, **3a** and **4a** at the C-2 OH in high yield and purity.

A number of C-2 OH ester derivatives of 20R-hydroxyecdysone (**1**) and of some analogues have been obtained in high yield and purity by transesterification in organic solvent catalyzed by lipase B from *Candida antarctica*.

Hirotsu M., Asaka I., Ino C., Furuya T. and Shiro M. (1987) Ganoderic acid derivatives and ergosta-4,7,22-triene-3,6-dione from *Ganoderma lucidum*. *Phytochemistry* **26**(10), 2797-2803.

Abstract: Seven new triterpenoids, ganoderic acid T, ganoderic acid S, ganoderic acid R, ganoderic acid P, ganoderic acid Q, ganoderic acid O, 7-O-methyl-ganoderic acid O and a known ergosterol derivative, ergosta-4,7,22-triene-3,6-dione were isolated from the cultured mycelium of *Ganoderma lucidum*. The structure of the first compound was determined using spectroscopic and X-ray analysis, and the structures of the other compounds were elucidated by spectroscopic data.

Ishizuka T., Yaoita Y. and Kikuchi M. (1997) Sterol constituents from the fruit bodies of *Grifola frondosa* (Fr.) S.F. Gray. *Chemical and Pharmaceutical Bulletin* **45**(11), 1756-1760.

Abstract: Four new sterols, 5 α , 6 α -epoxy-(22E, 24R)-ergosta-8(14), 22-diene-3 β , 7 β -diol (1), (22E, 24R)-ergosta-8, 22-diene-3 β , 5 α , 6 β , 7 α -tetrol (2), (22E, 24R)-ergosta-7, 9(11), 22-triene-3 β , 5 α , 6 β -triol (3) and 3 β , 5 α , 6 β -trihydroxy-(22E, 24R)-ergost-22-en-7-one (4), have been isolated from the fruit bodies of *Grifola frondosa* (FR.) S. F. GRAY (Polyporaceae) together with fourteen known ones (5-18), of which two (5 and 6) are reported for the first time from a natural source. The structures of these compounds were elucidated on the basis of spectral data.

Kawahara N., Sekita S. and Satake M. (1995) Two steroids from *Calvatia cyathiformis*. *Phytochemistry* **38**(4), 947-950.

Abstract: Cyathisterone (ergosta-7,22-diene-3,6-dione) and cyathisterol (8 β -hydroxyergosta-4,6,22-trien-3-one), two new sterols, have been isolated from *Calvatia cyathiformis* along with two known ergosterol derivatives, ergosta-4,7,22-triene-3,6-dione and ergosta-4,6,8(14),22-tetraen-3-one. Their molecular structures were defined by spectroscopic means and chemical correlations.

Kovganko N.V. (1999) Ecdysteroids and related compounds in fungi. *Chemistry of Natural Compounds* **35**(6) 597-611 [in English]/*Khimiya Prirodnykh Soedinenii* (6), 691-712 [in Russian].

Abstract: The structure and properties of ecdysteroids and related polyhydroxysterols isolated from fungi are reviewed.

Ohsawa T., Yukawa M., Takao C., Murayama M. and Bando H. (1992) Studies on constituents of fruit body of *Polyporus umbellatus* and their cytotoxic activity. *Chemical and Pharmaceutical Bulletin* **40**(1), 143-147.

Abstract: From the crude drug Chorei, the fruit body of *Polyporus umbellatus*, seven new components named polyporusterone A, B, C, D, E, F and G, were isolated and their structures were determined on the basis of the spectral data. These compounds showed cytotoxic action on leukemia 1210 cell proliferation.

Sun Y. and Yasukawa K. (2008) New anti-inflammatory ergostane-type ecdysteroids from the sclerotium of *Polyporus umbellatus*. *Bioorganic & Medicinal Chemistry Letters* doi: 10.1016/j.bmcl.2008.04.008.

Abstract: Bioassay-guided fractionation of the ethyl acetate extract from the sclerotium of *Polyporus umbellatus* resulted in the isolation of three new ergostane-type ecdysteroids, named polyporoid A (1), B (2), and C (3), together with five known ecdysteroids. The structures of the new compounds were determined on the basis of extensive spectroscopic data (IR, MS, ¹H and ¹³C NMR, and 2D NMR) analyses. All compounds (1–8) exhibited potent anti-inflammatory activity in the test of TPA-induced inflammation (1 µg/ear) in mice, with ID₅₀ values in the range of 0.117–0.682 µM/ear.

Šutara J. (1992) The genera *Paxillus* and *Tapinella* in Central Europe. *Česká Mykologie* 46(1/2), 50-56.

A brief survey of the genera *Paxillus* Fr. and *Tapinella* Gilb. is given with a key to the determination of Central European species of this group. Differences between the above mentioned genera are summarized into seven points. The species *Agaricus atrotomentosus* Batsch: Fr., often treated as a *Paxillus*, is here transferred to the genus *Tapinella*. The following new combinations are proposed: *Tapinella atrotomentosa* (Batsch: Fr.) comb. nov. and *Tapinella panuoides* (Ft.: Fr.) Gilb. f. *ionipes* (Quél.) comb. nov.

Takaishi Y., Adachi R., Murakami Y., Ohashi T., Nakano K. and Tomimatsu T. (1992) A polyoxygenated steroid from *Lasiosphaera nipponica*. *Phytochemistry* 31(1), 243-246.

Takemoto T., Ogawa S., Nishimoto N., Arihara S. and Bue K. (1967) Insect moulting activity of crude drugs and plants (1). *Yakugaku Zasshi* 87, 1414-1418 [in Japanese, with an English abstract].

Abstract: Ethanolic extracts of crude drugs and plants were examined for a possible metamorphosis activity in insects, by observing the state of pupation, using the free abdomen of flies. The results are summarised in Tables I to V. Among the crude drugs, bezoar as well as the leaves of taxum and mulberry showed such activity. Among the plants, no marked was found in mushrooms and sea-weeds. Among the ferns some activity was found in *Blechnum nipponicum*, *Athyrium nipponicum*, *A. squamigerum*, *Lastrea japonica*, *Rumohra miqueliana* and *Dryopteris chinensis*. In higher plants, *Podocarpus nakaii* and *Achyranthes* spp. showed the activity, and so did also *Taxus cuspidata* var. *nana*, *Podocarpus nagi*, *Torreya nucifera*, *Akebia quinata*, *Amaranthus spinosus*, *A. magnostanus* and *A. viridis*.

Tom W.-M., Abul-Hajj Y.J. and Koreeda M. (1975) Microbial oxidation of ecdysones. A convenient preparation of rubrosterone. *Journal of the Chemical Society, Chemical Communications* 24-25.

Abstract: Ponasterone A(I) is converted by the microorganism *Fusarium lini* ATCC 9593 into rubrosterone (IV) in 15% yield.

Valisolalao, J., Luu, B. and Ourisson G. (1983) Steroides cytotoxiques de *Polyporus versicolor*. *Tetrahedron* 39(17), 2779-2785 [in French, with and English abstract].

Abstract: The crude extracts of the European wood-rotting fungus *Polyporus versicolor* contain cytotoxic principles. Two of these have been isolated by monitoring the purification by tests on hepatoma cells grown *in vitro*. They are polyoxygenated derivatives of ergosterol, characterized by the partial structure $\Delta^7, 9\alpha$ -OH.

Vokáč K., Buděšinský M. and Harmatha J. (1995) New ecdysteroids from mushrooms. Communication presented at the 16th Conference on Isoprenoids, Prague, Abstract book, pp. 77-78.

Vokáč K., Buděšinský M., Harmatha J. and Piš J. (1998) New ergostane type ecdysteroids from fungi. Ecdysteroid constituents of mushroom *Paxillus atrotomentosus*. *Tetrahedron* 54, 1657-1666.

Abstract: New ergostane type ecdysteroids from the mushroom species *Paxillus atrotomentosus*: paxillosterone, its 20,22-*p*-hydroxybenzylidene acetal, atrotosterones A, B and C and 25-hydroxy-atrotosterones A and B have been characterized. 20-Hydroxyecdysone as a minor constituent has also been isolated. Configuration at C(24) of paxillosterone was derived from proton 2D-ROESY NMR spectra of its cyclic phenylboronate derivatives. Configuration at C(24) of 25-hydroxyatrotosterone A was assigned by comparison of its NMR spectra with the spectra of both 24-epimers of makisterone A.

Vokáč K., Buděšinský M., Harmatha J. and Kohoutová J. (1998) Ecdysteroid constituents of the mushroom *Tapinella panuoides*. *Phytochemistry* 49(7), 2109-2114.

Abstract: Ecdysteroids, compounds structurally related to the insect moulting hormone ecdysone, were isolated from the mushroom *Tapinella panuoides*. Paxillosterone and two further new ergostane-type ecdysteroids, panuosterone and 25-hydroxypanuosterone, together with the already known cholestane-type ecdysteroids, 20-hydroxyecdysone, ponasterone A, malacosterone and turkesterone were identified by NMR, IR and mass

spectrometric methods. The taxonomic value of ecdysteroids for chemosystematics in fungi and particularly in the genera *Paxillus* and *Tapinella*, is discussed.

Yaoita Y., Amemiya K., Ohnuma H., Furumura K., Masaki A., Matsuki T. and Kikuchi M. (1998) Sterol constituents from five edible mushrooms - Part III. *Chemical and Pharmaceutical Bulletin* **46** (6), 944-950.
Abstract: Eight new sterols, 5 α , 8 α -epidioxy-(22E, 24R)-23-methylergosta-6, 22-dien-3 β -ol (1), 3 β , 5 α , 9 α -trihydroxy-(22E, 24R)-23-methylergosta-7, 22-dien-6-one (2), 3 β , 5 α , 9 α -trihydroxy-(24S)-ergost-7-en-6-one (3), 3 β , 5 α , 9 α , 14 α -tetrahydroxy-(22E, 24R)-ergosta-7, 22-dien-6-one (4), (22E, 24R)-ergosta-7, 22-diene-3 β , 5 α , 6 α , 9 α -tetrol (5), 5 α , 9 α -epidioxy-3 β -hydroxy-(22E, 24R)-ergosta-7, 22-dien-6-one (6), 5 α , 9 α -epidioxy-3 β -hydroxy-(24S)-ergost-7-en-6-one (7) and 5 α , 6 α -epoxy-(22E, 24R)-ergosta-8, 22-diene-3 β , 7 β , 14 α -triol (8), have been isolated from five edible mushrooms, *Lentinus edodes*, *Flammulina velutipes*, *Hypsizigus marmoreus*, *Pleurotus ostreatus* and *Pholiota nameko* together with fifteen known ones (9-23), of which two (16 and 17) are reported for the first time from a fungal source. The structures of these new compounds were elucidated on the basis of their spectral data.

Yaoita Y., Matsuki K., Iijima T., Nakano S., Kakuda R., Machida K. and Kikuchi M. (2001) New sterols and triterpenoids from four edible mushrooms. *Chemical and Pharmaceutical Bulletin* **49**(5), 589-594.
Abstract: Four edible mushrooms, *Panellus serotinus*, *Lepista nuda*, *Tricholoma matsutake* and *Naematoloma sublateritium*, have been investigated chemically. Two new sterols, 5 α , 9 α -epidioxy-(22E)-ergosta-7, 22-diene-3 β , 6 α -diol (1) and 5 α , 9 α -epidioxy-(22E)-ergosta-7, 22-diene-3 β , 6 β -diol (2), have been isolated from *Panellus serotinus*. Compound 2 was also isolated from *Lepista nuda*. A new sterol, 3 β , 5 α , 9 α , 14 β -tetrahydroxy-(22E)-ergosta-7, 22-dien-6-one (3), and compound 2 have been isolated from *Tricholoma matsutake*. Three new triterpenoids, sublateriols A-C (4-6), have been isolated from *Naematoloma sublateritium*. The structures of the new compounds were elucidated on the basis of their spectral data.

Yaoita Y., Yoshihara Y., Kakuda R., Machida K. and Kikuchi M. (2002) New sterols from two edible mushrooms, *Pleurotus eryngii* and *Panellus serotinus*. *Chemical and Pharmaceutical Bulletin* **50** (4), 551-553.
Abstract: Two edible mushrooms, *Pleurotus eryngii* and *Panellus serotinus*, have been investigated chemically. Two new sterols, 5 α , 9 α -epidioxy-8 α , 14 α -epoxy-(22E)-ergosta-6, 22-dien-3 β -ol (1) and 3 β , 5 α , 9 α -dihydroxyergost-7-en-6-one (2), have been isolated from *P. eryngii*, together with six known ones (3-8). Compound 1 was also isolated from *P. serotinus*. The structures of the new compounds were elucidated on the basis of their spectral data.

Yu F-X., Li Z., Chen Y., Yang Y-H., Li G-H. and Zhao P-J. (2017) Four new steroids from the endophytic fungus *Chaetomium* sp. M453 derived of Chinese herbal medicine *Huperzia serrata*. *Fitoterapia* **117**, 41-46.
Abstract: An endophytic fungus, *Chaetomium* sp. M453, was isolated from *Huperzia serrata* (Thunb. ex Murray) Trev. and subjected to phytochemical investigation. Three unusual C25 steroids, neocyclocitrinols E-G (1-3), and 3 β -hydroxy-5,9-epoxy-(22E,24R)-ergosta-7,22-dien-6-one (4) together with three known steroids were isolated from solid fermentation products of the fungus, which were elucidated by extensive spectroscopic analyses, including 1D-, 2D-NMR, and HR-ESI-MS experiments. The absolute configuration of 1 was determined by X-ray crystallographic analysis and CD analyses. The acetylcholinesterase inhibitory activities of compounds 1-4 were tested in vitro. Compound 4 showed weak acetylcholinesterase inhibitory activity.

Zheng S.-H., Yang H.-P., Ma X.-M. and Shen X.-W. (2004) Two new polyporusterones from *Polyporus umbellatus*. *Natural Product Research* **18**(5), 403-407.
Abstract: Two new polyporusterones named as porusterone I and polyprosterone II were isolated from *polyourus umbellatus*. Their structures have been established on the basis of spectroscopic analysis.

Zhou W-W., Lin W-H. and Guo S-X. (2007) Two new polyporusterones isolated from the sclerotia of *Polyporus umbellatus*. *Chemical and Pharmaceutical Bulletin* **55**(8), 1148-1150.
Abstract: In the course of searching for marker components, two new polyporusterones were isolated from the sclerotia of *Polyporus umbellatus*, together with another three known analogs. The structures of the new ones were elucidated as (20S,22R,24R)-16,22-epoxy-3 β ,14 α ,23 β ,25-tetrahydroxyergost-7-en-6-one (1) and (23R,24R,25R)-23,26-epoxy-3 β ,14 α ,21 α ,22 α -tetrahydroxyergost-7-en-6-one (2) by chemical and spectroscopic means, including HR-FAB-MS, 1D- and 2D-NMR.

4. ECDYSTEROID(-RELATED) COMPOUNDS FROM NON-CRUSTACEAN MARINE ORGANISMS

Aiello A., Fattorusso E., Magno S. and Menna M. (1991) Isolation of five new 5α -hydroxy-6-keto- Δ^7 sterols from the marine sponge *Oscarella lobularis*. *Steroids* **56**, 337-340.

Abstract: Five novel sterols isolated from the marine sponge *Oscarella lobularis* have been identified on the basis of spectral arguments: cholest-7-ene-3 β ,5 α -diol-6-one (1), cholesta-7,22E-diene-3 β ,5 α -diol-6-one (2), 24-methylcholesta-7,22E-diene-3 β ,5 α -diol-6-one (3), 24-methylcholesta-7,24(28)-diene-3 β ,5 α -diol-6-one (4), and 24-ethylcholest-7-ene-3 β ,5 α -diol-6-one (5).

Aiello A., Fattorusso E. and Menna M. (1999) Steroids from sponges: recent reports. *Steroids* **64**, 687-714.
No Abstract.

Behrens W. and Bückmann D. (1981) Der Nachweis von Häutungshormonen bei einem Pantopoden, *Pycnogonum littorale* Ström [The detection of moulting hormones in a pycnogonid, *Pycnogonum littorale* Ström] Verhandlungen der Deutschen Zoologischen Gesellschaft 195 [in German].

Behrens W. and Bückmann D. (1983) Ecdysteroids in the pycnogonid *Pycnogonum littorale* (STRÖM) (Arthropoda, Pantopoda). *General and Comparative Endocrinology* **51**, 8-14.

Abstract: Two ecdysteroids were isolated from juveniles and adults as well as from eggs of the pycnogonid *Pycnogonum littorale* Strom and characterized by bioassay, thin layer chromatography, gas liquid chromatography, and high performance liquid chromatography. One of the hormones could be identified as 20-hydroxyecdysone whereas the other behaves in HPLC in a way that is similar to but not identical with ecdysone. This unidentified ecdysone-like hormone exceeded 20-hydroxyecdysone in all developmental stages (1.67: 1 to 3:1). Determination of moulting hormone titers by a radioimmunoassay showed highest amounts of hormones in juvenile females (150 ng/mg tissue dry wt). The results are compared with those obtained from other arthropods and are discussed in view of the evolution of hormonal systems.

Cachet N., Gento-Jouve G., Ivanisevic J., Chevaldonne P., Sinniger F., Culioli G., Perez T. and Thomas O.P. (2015) Metabolomic profiling reveals deep chemical divergence between two morphotypes of the zoanthid *Parazoanthus axinellae*. *Scientific Reports* **5**, article 8282 (doi: 10.1038/srep08282).

Abstract: Metabolomics has recently proven its usefulness as complementary tool to traditional morphological and genetic analyses for the classification of marine invertebrates. Among the metabolite-rich cnidarian order Zoantharia, *Parazoanthus* is a polyphyletic genus whose systematics and phylogeny remain controversial. Within this genus, one of the most studied species, *Parazoanthus axinellae* is prominent in rocky shallow waters of the Mediterranean Sea and the NE Atlantic Ocean. Although different morphotypes can easily be distinguished, only one species is recognized to date. Here, a metabolomic profiling approach has been used to assess the chemical diversity of two main Mediterranean morphotypes, the “slender” and “stocky” forms of *P. axinellae*. Targeted profiling of their major secondary metabolites revealed a significant chemical divergence between the morphotypes. While zoanthoxanthin alkaloids and ecdysteroids are abundant in both morphs, the “slender” morphotype is characterized by the presence of additional and bioactive 3,5-disubstituted hydantoin derivatives named parazoanthines. The absence of these specific compounds in the “stocky” morphotype was confirmed by spatial and temporal monitoring over an annual cycle. Moreover, specimens of the “slender” morphotype are also the only ones found as epibionts of several sponge species, particularly *Cymbaxinella damicornis* thus suggesting a putative ecological link.

Cafieri F., Fattorusso E. and Tagliatalata-Scafati O. (1998) Novel bromopyrrole alkaloids from the sponge *Agelas dispar*. *Journal of Natural Products* **61**(1), 122-125.

Abstract: Further investigation of the Caribbean marine sponge *Agelas dispar* for biologically active constituents has led to the isolation of the novel bromopyrrole alkaloids longamide B (1), and clathramides C (2) and D (3), whose structures have been determined by spectroscopic methods. Isolation of the known keramide (4) and of the ecdysonic sterols ecdysterone (5) and ajugasterone C (6) is also reported. The antimicrobial activities of the isolated bromopyrrole alkaloids is summarized.

Cheng Y-B., Lee J-C., Lo J-W. Chen S-R., Hu H-C., Wu Y-H., Wu Y-C. And Chang F-R. (2016) Ecdysones from *Zoanthus* spp. with inhibitory activity against dengue virus 2. *Bioorganic and Medicinal Chemistry Letters* **26**, 2344-2348.

Abstract: Bioassay-guided fractionation of an ethanolic extract of *Zoanthus* spp. collected in Taiwan has resulted in the isolation of one new ecdysone, zoanthon A (1), along with thirteen known compounds (2-14). The structures of these compounds were determined by spectroscopic methods, especially 2D NMR analyses. The in vitro antiviral activities of all isolated ecdysones (1-14) against dengue virus type 2 (DENV-2) were evaluated using DENV infectious system. New compound (1) exhibited potent antiviral activity (EC₅₀=19.61 ± 2.46 μ M) with a selectivity

index (CC50/EC50) value of 36.7. The structure-activity relationships of isolated ecdysones against DENV-2 were concluded. Molecular docking information of **3** and NS5 polymerase was performed either.

Chludil H.D., Seldes A.M. and Maier M.S. (2002) Antifungal steroidal glycosides from the Patagonian starfish *Anasterias minuta*: structure-activity correlations. *Journal of Natural Products* **65**, 153-157.

Abstract: Two new sulfated steroidal hexaglycosides, anasterosides A (**2**) and B (**3**), along with the known versicoside A (**1**) have been isolated from the Patagonian starfish *Anasterias minuta*. Their structures have been elucidated by spectroscopic analysis (NMR and FABMS) and chemical transformations. Compounds **1** and **2** and the synthetic pentaglycoside **1b** derived from versicoside A showed antifungal activity against the plant pathogenic fungus *Cladosporium cucumerinum*. Desulfation of hexaglycoside **1** rendered a totally inactive saponin.

Costantino V., Dell'Aversano C., Fattorusso E. and Mangoni A. (2000) Ecdysteroids from the Caribbean sponge *Ietrochota birotulata*. *Steroids* **65**(3), 138-142.

Abstract: The sterol composition of the Caribbean sponge *Ietrochota birotulata* was investigated. Structure of a new ecdysteroid 2 β ,3 β ,14 α , 20 β -tetrahydroxy-22 α -(2-hydroxyacetyloxy)-5 β eta-colest-7-en-6-one (**1**) was assigned on the basis of spectroscopic and chemical evidence and molecular mechanics calculations. Isolation of the widespread ecdysteroids 2-5 is also reported.

De Marino S., Palagiano E., Zollo F. and Minale L. (1997) A novel sulphated steroid with a 7-membered 5-oxalactone B-ring from an Antarctic starfish of the family Asteriidae. *Tetrahedron* **53**(25), 8625-8628.

Abstract: Three novel sulphated polyhydroxylated steroids, named asterasterols A-C (**1-3**), have been isolated from an Antarctic starfish of the family *Asteriidae* and their structures elucidated by spectroscopic studies. Steroid **1** has the seven-membered 5-oxalactone B-ring; **2** and **3** are the corresponding 6-oxo steroids.

Three novel sulphated polyhydroxylated steroids, named asterasterols A-C (**1-3**), have been isolated from an Antarctic starfish of the family *Asteriidae* and their structures elucidated by spectroscopic studies. Steroid **1** has the seven-membered 5-oxalactone B-ring, **2** and **3** are the corresponding 6-oxo steroids.

De Marino S., Iorizza M., Zollo F., Minale L., Amsler C.D., Baker B.J. and McClintock J.B. (1997) Isolation, structure elucidation, and biological activity of the steroid oligoglycosides and polyhydroxysteroids from the Antarctic starfish *Acodontaster conspicuus*. *Journal of Natural Products* **60**(10), 959-966.

Abstract: A total of 19 steroids, of which 13 steroidal oligoglycosides (nine new and four known) and six polyhydroxylated steroids (four new and two known), has been isolated from the Antarctic starfish *Acodontaster conspicuus*. The mixture is dominated by glycosides composed of steroidal aglycons having the hydroxyl groups typically disposed on one side of the tetracyclic nucleus, i.e., 3 β ,4 β ,6 α ,8,15 β -, with some having a sulfate at C-6, and differing in the side chains and/or in the disaccharide moieties that are usually attached at C-26, with some at C-28 and C-29. Those compounds are accompanied by minute amounts of glycosides with a $\Delta^{8(14)}$ -double bond in the steroid, which is a structural feature not previously found among polyhydroxysteroids derived from starfish. Small amounts of six related unglycosidated polyhydroxysteroids and three higher-molecular-weight asterosaponins complete the composition of the mixture. The structures of the new compounds were determined by interpretation of their spectral data and by comparison with spectral data of known compounds. Eighteen of these compounds were evaluated for their ability to inhibit growth in Antarctic marine bacteria isolated from either the water column or the surfaces of benthic marine invertebrates. Of these compounds, 50% were active against at least one Antarctic marine bacterium. This suggests that these compounds may play an important role in deterring microbial fouling.

Diop M., Samb A., Costantino V., Fattorusso E. and Mangoni A. (1996) A new iodinated metabolite and a new alkyl sulfate from Senegalese sponge *Ptilocaulis spiculifer*. *Journal of Natural Products* **59**(3), 271-272. [muristerone A also isolated from the sponge].

Abstract: The hydrophilic extract of the Senegalese sponge *Ptilocaulis spiculifer* has been analyzed. It has been shown to contain dakaramine (**1**), a new tyrosine derivative containing iodine, an unusual feature for sponge metabolites. In addition, the new alkyl sulfate **2** (as a counterion of **1**) and the known ecdysonic sterol **3** were isolated from the same extract.

Federov S.N., Stonik V.A. and Elyakov G.B. (1988) Identification of ecdysteroids of hexactinic corals. *Chemistry of Natural Compounds* **24**(4), 517-518 [in English]/ *Khimiya Prirodnikh Soedinenii* (4), 603-604 [in Russian]. No Abstract.

Guerriero A. and Pietra F. (1985) Isolation, in large amounts, of the rare plant ecdysteroid ajugasterone-C from the Mediterranean zoanthid *Gerardia savaglia*. *Comparative Biochemistry and Physiology* **80B**(2), 277-278.

Abstract: Ajugasterone-C, an ecdysteroid so far thought to be an exclusive and rare, terrestrial, plant product, and ecdysone, have been isolated, in relatively large amounts (milligrams per gram dry animal), from both wild and captive Mediterranean zoanthid *Gerardia savaglia*.

Guerrero A., Traldi P. and Pietra F. (1986) Gerardiasterone, a new ecdysteroid with a 20,22,23,25-tetrahydroxylated side chain from the Mediterranean zoanthid *Gerardia savaglia*. *Journal of the Chemical Society, Chemical Communications* 40-41.

Abstract: Homonuclear spin decouplings of the 500 MHz ¹H n.m.r. spectrum, proton shift-correlated 2D (COSY) n.m.r. experiments, selective heteronuclear decouplings of the ¹³C n.m.r. spectrum, and mass spectra of the title compound, as well as ¹H n.m.r. spectra and nuclear Overhauser effect (n.o.e.) experiments with its acetate and acetonide derivatives, suggest the structure 23-hydroxyecdysterone (1a) for the title compound whilst, presumably owing to a flexible 1,3-dioxane ring in the boat conformation, the C-20/23 acetonide (1g) fails to provide configurational information as to the chain.

Guillen P.O., Calabro K., Jaramillo K.B., Dominguez C., Genta-Jouve G., Rodriguez J. and Thomas O.P. (2018) Ecdysonelactones, ecdysteroids from the tropical Eastern Pacific zoantharian *Antipathozoanthus hickmani*. *Marine Drugs* **16**, 58 (doi: 10.3390/md16020058) pp. 10.

Abstract: Despite a large occurrence, especially over the Pacific Ocean, the chemical diversity of marine invertebrates belonging to the order Zoantharia is largely underexplored. For the two species of the genus *Antipathozoanthus* no chemical study has been reported so far. The first chemical investigation of *Antipathozoanthus hickmani* collected at the Marine Protected Area “El Pelado”, Santa Elena, Ecuador, led to the isolation of four new ecdysteroid derivatives named ecdysonelactones. The structures of ecdysonelactones A–D (**1–4**) were determined based on their spectroscopy data, including 1D and 2D NMR and HRMS. The four compounds of this family of ecdysteroids feature an unprecedented γ -lactone fused at the C-2/C-3 position of ring A. These derivatives exhibited neither antimicrobial nor cytotoxic activities.

Guillen P.O., Jaramillo K.B., Genta-Jouve G. and Thomas O.P. (2019) Marine natural products from zoantharians: bioactivity, biosynthesis, systematic, and ecological roles. *Natural Product Reports* (doi: 10.1039/c9np00034g).

Abstract: Zoantharians, also improperly known as zoanthids or colonial anemones, are well known by aquarists because of their ease of use in aquaria but also because of their splendid colours. However, high concentrations of the highly toxic palytoxin found in some species of zoantharians maintained in reef aquaria has raised some issues recently, unveiling at the same time a rather unknown chemical diversity hidden in these marine beauties. Herein, we report the structure of the metabolites described in all species of zoantharians up to the end of 2018 and their associated biological activities. As sessile invertebrates, zoantharians harbour a rich diversity of micro-organisms that can play a role in the biosynthesis of these natural products and we detail the current hypotheses on the metabolic pathways leading to the identified ecdysteroids, zoanthoxanthins, zoanthamines, palytoxins and others. Finally, we assess the possible use of these metabolites in the systematics of such a complex group of marine invertebrates and we discuss their possible ecological roles. Altogether, this review brings some insights into the rich chemical diversity of zoantharians and their potential for marine biodiscovery and marine ecology.

Hansen K.O., Isaksson J., Glomsaker E., Andersen J.H. and Hansen E. (2018) Ponasterone[s] A and F, ecdysteroids from the Arctic bryozoans *Alcyonidium gelatinosum*. *Molecules* **23**, 1481, pp. 9 (doi: 10.3390/molecules23061481).

Abstract: A new ecdysteroid, ponasterone F (**1**) and the previously reported compound ponasterone A (**2**) were isolated from specimens of the Arctic marine bryozoan *Alcyonidium gelatinosum* collected at Hopenbanken, off the coast of Edgeøya, Svalbard. The structure of **1** was elucidated, and the structure of **2** confirmed by spectroscopic methods including 1D and 2D NMR and analysis of HR-MS data. The compounds were evaluated for their ability to affect bacterial survival and cell viability, as well as their agonistic activities towards the estrogen receptors α and β . The compounds were not active in these assays. Compound **2** is an arthropod hormone controlling molting and are known to act as an allelochemical when produced by plants. Even though its structure has been previously reported, this is the first time a ponasterone has been isolated from a bryozoan. *A. gelatinosum* produced **1** and **2** in concentrations surpassing those expected of hormonal molecules, indicating their function as defence molecules against molting predators. This work adds to the chemical diversity reported from marine bryozoans and expanded our knowledge of the chemical modifications of the ponasterones.

Honda T., Takada H., Miki S. and Tsubuki M. (1993) Synthesis and structure elucidation of a novel ecdysteroid, gerardiasterone. *Tetrahedron Letters* **34**(51), 8275-8278.

Abstract: The structure of a novel ecdysteroid, gerardiasterone, is elucidated as **2a** by its synthesis employing a diastereoselective dihydroxylation of the *E*-olefin **22** as a key step.

The structure of gerardiasterone was unambiguously determined by its stereoselective synthesis.

Lee J.-C., Chang F.-R., Chen S.-R., Wu Y.-H., Hu H.-C., Wu Y.-C., Backlund A. and Cheng Y.-B. (2016) Anti-Dengue virus constituents from Formosan zoanthid *Palythoa mutuki*. *Marine Drugs* 14, 151 (DOI: 10.3390/md14080151).

Abstract: A new marine ecdysteroid with an α -hydroxy group attaching at C-4 instead of attaching at C-2 and C-3, named palythone A (**1**), together with eight known compounds (**2–9**) were obtained from the ethanolic extract of the Formosan zoanthid *Palythoa mutuki*. The structures of those compounds were mainly determined by NMR spectroscopic data analyses. The absolute configuration of **1** was further confirmed by comparing experimental and calculated circular dichroism (CD) spectra. Anti-dengue virus 2 activity and cytotoxicity of five isolated compounds were evaluated using virus infectious system and [3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium, inner salt (MTS) assays, respectively. As a result, peridinin (**9**) exhibited strong antiviral activity ($IC_{50} = 4.50 \pm 0.46 \mu\text{g/mL}$), which is better than that of the positive control, 2'CMC. It is the first carotene-like substance possessing anti-dengue virus activity. In addition, the structural diversity and bioactivity of the isolates were compared by using a ChemGPS–NP computational analysis. The ChemGPS–NP data suggested natural products with anti-dengue virus activity locate closely in the chemical space.

Miyata Y., Diyabalanage T., Amsler C.D., McClintock J.B., Valeriote F.A. and Baker B.J. (2007) Ecdysteroids from the Antarctic tunicate *Synoicum adareanum*. *Journal of Natural Products* (on-line 27/11/2007).

Abstract: Five new ecdysteroids, hyousterones A–D (**2–5**) and abeohousterone (**6**), have been isolated from the Antarctic tunicate *Synoicum adareanum* along with the known ecdysteroid dialulsterol B (**1**). Hyousterones B (**3**) and D (**5**) are unusual ecdysteroids in bearing the 14β -hydroxyl group, and abeohousterone incorporates the $13(14\rightarrow 8)$ abeo steroid skeleton, reflecting a rearrangement of the steroid C/D ring system. Abeohousterone has moderate cytotoxicity toward several cancer cell lines. Hyousterones bearing the 14α -hydroxy group (**2** and **4**) were weakly cytotoxic, while the 14β -hydroxy hyousterones (**3** and **5**) were devoid of cytotoxicity. The 14β -hydroxy function may be a thermodynamic pathway to the $13(14\rightarrow 8)$ abeo steroid skeleton. Hyousterones, abeohousterone, and dialulsterol B are the first ecdysteroids reported from tunicates.

Okazaki R.K., Snyder M.J. and Chang E.S. (1988) Ecdysteroids in nemerteans: presence and physiological role. *Hydrobiologia* 156, 153-160.

Abstract: Ecdysteroids are a class of steroidal hormones that are important in molting and reproduction in arthropods. These hormones have been recently detected in non-arthropodan groups, such as soft-bodied worms. To continue our efforts to determine the presence of ecdysteroids in nemerteans, this study further documents the identification of 20-hydroxyecdysone (20E), in *Paranemertes peregrina*, by gas chromatography/mass spectrometry (GC/MS). After C_{18} Sep-Pak extraction, radioimmunoassay of the 40% and 80% methanolic fractions from an extract of 1000 worms indicated 98 and 156 ng of ecdysteroids, respectively. Ecdysteroids of these two samples, as well as the 20E and ecdysone standards, were N-trimethylsilylimidazole (TMSI)-derivatized before GC/MS analysis. The methanolic samples contained a large number of compounds but only small, insignificant peaks in the area where ecdysteroid standards eluted. However, the reconstruction ion current (RIC) chromatograms for m/z 561 indicated the presence of 20E at the correct retention time of 12.48 min in the 40% methanolic fraction. Reanalysis of the samples under selected ion monitoring mode demonstrated the presence of 20E in both the 40% and 80% fractions. These results demonstrate conclusively the presence of an active ecdysteroid in the phylum Nemertea.

Okazaki R.K., Snyder M.J., Grimm C.C. and Chang E.S. (1998) Ecdysteroids in nemerteans: further characterization and identification. *Hydrobiologia* 365, 281-285.

Abstract: Ecdysteroids are a class of steroidal hormones that are important in molting and reproduction in arthropods. These hormones have been recently detected in non-arthropodan groups, such as soft-bodied worms. To continue our efforts to determine the presence of ecdysteroids in nemerteans, this study further documents the identification of 20-hydroxyecdysone (20E), in *Paranemertes peregrina*, by gas chromatography/mass spectrometry (GC/MS). After C_{18} Sep-Pak extraction, radioimmunoassay of the 40% and 80% methanolic fractions from an extract of 1000 worms indicated 98 and 156 ng of ecdysteroids, respectively. Ecdysteroids of these two samples, as well as the 20E and ecdysone standards, were N-trimethylsilylimidazole (TMSI)-derivatized before GC/MS analysis. The methanolic samples contained a large number of compounds but only small, insignificant peaks in the area where ecdysteroid standards eluted. However, the reconstruction ion current (RIC) chromatograms for m/z 561 indicated the presence of 20E at the correct retention time of 12.48 min in the 40% methanolic fraction. Reanalysis of the samples under selected ion monitoring mode demonstrated the presence of 20E in both the 40% and 80% fractions. These results demonstrate conclusively the presence of an active ecdysteroid in the phylum Nemertea.

Parameswaran P.S., Nair C.G., Gonsalves C. and Achuthankutty C.T. (2001) Isolation of 2-deoxyecdysterone, a novel oxytocic agent, from a marine *Zoanthus* sp. *Journal of the Indian Institute of Science* 81, 169-173.

Abstract: The insect-moulting hormone 2-deoxyecdysterone (2-deoxy-20-hydroxyecdysone **1**) has been isolated for the first time and in relatively high yield (0.016%) from a marine *Zoanthus* sp. The compound exhibited promising

oxytocic activity in guinea pig uterus assay. The sup(1)H and sup(13)C NMR assignments of the compound aided by COSY, TOCSY, HMQC and HMBC data are reported here for the first time

Searle P.A. and Molinski T.F. (1995) 4-Dehydroecdysterone, a new ecdysteroid from the zoanthid *Parazoanthus* sp. *Journal of Natural Products* **58**(2), 264-268.

Abstract: A novel ecdysteroid, 4-dehydroecdysterone [3], has been isolated from the zoanthid *Parazoanthus* sp. collected from Port Phillip Bay, Australia. The known ecdysteroids ecdysterone [1] and ajugasterone C [2] were also isolated, together with the known alkaloid paragraine [4].

Shigemori H., Sato Y., Kagata T. and Kobayashi J. (1999) Palythoalones A and B, new ecdysteroids from the marine zoanthid *Palythoa australiae*. *Journal of Natural Products* **62**(2), 372-374.

Abstract: Two new ecdysteroids, palythoalones A (1) and B (2), have been isolated from the marine zoanthid *Palythoa australiae*. The structures have been elucidated on the basis of spectroscopic data and by chemical means.

Snyder M.J., Okazaki R.K. and Chang E.S. (1992) Nemertean ecdysteroids: relationship to reproduction. *Invertebrate Reproduction and Development* **21**(1), 7-13.

Abstract: Ecdysteroids in non-reproductive, reproductive, and post-reproductive specimens of two nemerteans, *Paranemertes peregrina* and *Pantinnemertes californiensis*, were studied using radioimmunoassay (RIA). Ecdysteroid concentrations increased during the reproductive season in females of both species. These data suggest the possibility that ecdysteroids are involved in reproduction. Study of ecdysteroid profiles using reverse phase high-performance liquid chromatography (RP-HPLC) followed by RIA showed that the types and relative amounts of different ecdysteroid metabolites were similar between mature male and female *Pan. californiensis*. In addition, [³H]-ecdysone was injected into mature male and female *Pan. californiensis*. The resulting RP-HPLC profiles indicated that both sexes metabolized [³H]-ecdysone to similar products. Both sexes produced polar and apolar ecdysteroid conjugates as the major metabolites of [³H]-ecdysone, with only traces of [³H]-20-hydroxyecdysone.

Sturaro A., Guerriero A., De Clauser R. and Pietra F. (1982) A new, unexpected marine source of a molting hormone. Isolation of ecdysterone in large amounts from the zoanthid *Gerardia savaglia*. *Experientia* **38**, 1184-1185.

Abstract: Ecdysterone was found in large amounts in the Mediterranean zoanthid *Gerardia savaglia* both immediately after its collection and after it had been kept for 15 months in an aquarium. This is the first time that an ecdysteroid has been found in a marine animal which does not belong to the phylum Arthropoda.

Su J., Peng T., Long K. and Zeng L. (1986) 24ξ-methyl 5α-cholestane-3α,6β,9α,25-tetrol 25 monoacetate, a novel polyhydroxylated steroid from the soft coral *Sarcophyton tortuosum*. *Steroids* **48**, 233-238.

Abstract: A novel polyhydroxylated steroid, named sartortuosterol A, with rare 3 alpha- and 6-hydroxyl groups, was isolated from the South China Sea soft coral *Sarcophyton tortuosum* Tixier-Durivault, and its structure was established as 24xi-methyl 5 alpha-cholestane-3 alpha, 6 beta, 9 alpha,25-tetrol 25-monoacetate from spectroscopic data and chemical conversions.

Suksamrarn A., Jankam A., Tarnchompoo B. and Putchakarn S. (2002) Ecdysteroids from a *Zoanthus* sp. *Journal of Natural Products* **65**, 1194-1197.

Abstract: A new ecdysteroid, zoanthusterone, has been isolated from a marine zoanthid, *Zoanthus* sp. Ten known ecdysteroids, ponasterone A, 20-hydroxyecdysone 2-acetate, viticosterone E, integristerone A 25-acetate, 2-deoxy-20-hydroxyecdysone, ecdysone, ajugasterone C, dacryhainansterone, inokosterone, and 20-hydroxyecdysone, have also been isolated. This is the first report of ecdysteroids in a *Zoanthus* species.

Tomaschko K.-H. (1994a) Defensive secretion of ecdysteroids in *Pycnogonum littorale* (Arthropoda, Pantopoda). *Zeitschrift für Naturforschung* **49c**, 367-371.

Abstract: In response to disturbance, the pycnogonid *Pycnogonum littorale* discharges a mixture of eight ecdysteroids (ES). Repeated intensive molestation causes 99% secretion of the endogenous ES present. The concentration of the total ES in the defensive effluent is $1.0 \times 10^{-3} \text{ mol} \cdot \text{l}^{-1}$. 20-Hydroxyecdysone 22-acetate, the predominant ES, reaches $0.8 \times 10^{-3} \text{ mol} \cdot \text{l}^{-1}$. This is sufficient to deter significantly feeding by the common shore crab *Carcinus maenas*, a generalist predator in the habitat of the pycnogonid. There is evidence that the secreted ES of *P. littorale* contribute to its unpalatability. The present paper describes for the first time defensive secretion in marine arthropods.

Tomaschko K.-H. (1994b) Ecdysteroids from *Pycnogonum littorale* (Arthropoda, Pantopoda) act as chemical defense against *Carcinus maenas* (Crustacea, Decapoda). *Journal of Chemical Ecology* **20**(7), 1445-1455.

Abstract: *Pycnogonum littorale* (Ström) is unpalatable to the common shore crab *Carcinus maenas*, a generalist predator in the pycnogonid's habitat. A feeding bioassay reveals that the crabs are deterred by ecdysteroids that occur in high levels in all developmental stages of *P. littorale*. The total ecdysteroids in the pycnogonids reach 5.9×10^{-4} M. The 20-hydroxyecdysone 22-acetate (20E22A), which is the predominant ecdysteroid in the pycnogonids, and 20-hydroxyecdysone (20E), the arthropod molting hormone, were tested for their antifeedant effect on *C. maenas*. When contained in food pellets in homogeneous concentrations, 20E and 20E22A significantly reduced food consumption at 1.25×10^{-4} and 5.0×10^{-4} molar levels, respectively. The present results demonstrate for the first time chemical defense in arthropods in a marine predator-prey relationship. Furthermore, they provide evidence that ES contained in one animal can act as feeding deterrents on another animal.

Tomaschko K.-H. (1997) Ecdysteroids in Pycnogonids: hormones and interspecific allelochemicals. In: Ecological Studies, 130; Vertical Food Web Interactions, Springer-Verlag, pp.171-188.

Tomaschko K.-H. and Bückmann D. (1993) Excessive abundance and dynamics of unusual ecdysteroids in *Pycnogonum littorale* Ström (Arthropoda, Pantopoda). General and Comparative Endocrinology **90**, 296-305.
Abstract: In embryos, larvae, and juveniles of *Pycnogonum littorale*, unusual ecdysteroids have been found at excessively high levels. Concentrations and dynamics of the following eight ecdysteroids were determined by HPLC: 20-hydroxyecdysone, 20-hydroxyecdysone 22-glycolate, (25R) and (25S) isomers of 20,26-dihydroxyecdysone 22-acetate, 22-deoxy-20,26-dihydroxyecdysone, 20-hydroxyecdysone 22-acetate, 22-deoxy-20-hydroxyecdysone, and ecdysone 22-glycolate. The excessively high content of ecdysteroids is found in all developmental stages. Juvenile pycnogonids contain the highest total ecdysteroid amounts ever found in arthropods: 0.1% of their body dry weight. 20-Hydroxyecdysone 22-acetate is the prominent ecdysteroid and represents at all stages of both sexes, 66.2-85.8% of the total ecdysteroids. This compound, as well as all other ecdysteroids without a free 22-OH group, are presumed to be inactive as molting hormones. In contrast to these compounds, 20-hydroxyecdysone is present only in amounts similar to those in other arthropods. Furthermore, it is the only ecdysteroid with its peak at apolysis. It is regarded as the molt-promoting hormone. The origin and function of the other ecdysteroids in different developmental stages are discussed.

Tursch B., Hootel  C., Kaisin M., Losman D. and Karlsson R. (1976) Chemical studies of marine invertebrates. XVI. Structure and absolute configuration of lobsterol, a novel polyoxygenated sterol from the alcyonacean *Lobophytum pauciflorum* (Coelenterata, Octocorallia). Steroids **27**(1), 137-142.

Abstract: Lobosterol [(24S)-24-methylcholestane-3 β ,4 β ,5 β ,25-tetrol-6-one 25 monoacetate] has been isolated from the Alcyonacean *Lobophytum pauciflorum*. The structural elucidation of this novel polyoxygenated sterol was achieved by chemical and spectroscopic evidence, and by X-ray diffraction analysis.

5. ECDYSTEROID(-RELATED) COMPOUNDS FROM NEMATODES, CESTODES, TREMATODES etc.

5a. General

Gharib B., Baswa id S., Quilici M. and de Reggi M. (1991) Ecdysteroid-like compounds in human urine: they can occur in the absence of any parasitic infection. Clinica Chimica Acta **199**, 159-166.

Abstract: Ecdysteroids (compounds related to 20-hydroxyecdysone, the insect molting hormone) can appear in the blood and urine of man, as a result of an infection with helminths. It has been assumed that the products are released by parasites. However, we found that the phenomenon is not restricted to helminthiasis, but is widely spread among patients suffering from various diseases or injuries: twenty percent of hospital in-patients had urine highly positive in our test. This was due to the appearance of immunoreactive compounds not found in healthy people. Among them, one was remarkable for being largely predominant in some patients. These findings indicate that the origin and significance of ecdysteroids in man should be reconsidered. Since they appear only in association with severe pathological conditions, they could be of potential interest as a clinical marker.

Koolman J. (1990) The occurrence of ecdysteroids in vertebrates infected with helminths. In: Progress in Comparative Endocrinology, Wiley-Liss, pp. 704-709.

No Abstract.

5b. Nematodes

Bottjer K.P., Whisenton L.R. and Weinstein P.P. (1984) Ecdysteroid-like substances in *Nippostrongylus brasiliensis*. Journal of Parasitology **70**(6), 986-987.

No Abstract.

Chitwood D.J. (1987) Inhibition of steroid or hormone metabolism or action in nematodes. In: *Vistas on Nematology* pp. 122-130.

Chitwood D.J. (1999) Biochemistry and function of nematode steroids. *Critical reviews in Biochemistry and Molecular Biology* 34(4), 273-284.

Abstract: Compared to other organisms, nematodes have been the subject of relatively few investigations into their steroid biochemistry. Nutritional experiments have clearly demonstrated a dietary requirement for sterol that results from the inability of nematodes to biosynthesize steroids *de novo*. Although the specificity of the nutritional requirement varies somewhat among nematodes, most 4-desmethysterols can be directly utilized by nematodes or else metabolized to sterols better suited for nematode growth and development. Much knowledge has been obtained about the abilities of microbivorous nematodes to metabolize sterols. Various species can remove ethyl and methyl substituents at C-24, introduce double bonds at C-7, C-23, and C-24(28), reduce Δ^5 -, Δ^{22} -, and Δ^{24} -bonds, isomerize Δ^7 to $\Delta^8(14)$ -bonds, esterify fatty acids at C-3, and directly methylate the sterol nucleus at C-4 by a process unique to nematodes. Moreover, azasteroids and structurally related alkylamines and alkylamides inhibit the activity of nematode Δ^{24} -sterol reductase and also disrupt the life cycles of these species as well as parasitic species. Less is known, however, about sterol metabolism in parasitic nematodes because of the inability to culture them apart from their hosts. Most plant-parasitic nematodes appear to dealkylate plant sterols at C-24 and modify the sterols nucleus, particularly by reduction of Δ^5 -bonds. Sterol metabolism in mammalian parasites has been studied to a very limited extent only. Except for a structural presence of sterols in membranes of nematodes, the function of these compounds in nematodes is poorly understood. Although nematodes do contain steroids that have hormonal functions in other organisms, attempts to demonstrate endogenous biosynthesis of these steroids by nematodes have been unsuccessful and elucidation of the function of these compounds within nematodes warrants immediate investigation. Other areas of nematode steroid biochemistry requiring further research include investigation of the metabolism of sterols by vertebrate-parasitic nematodes, the purification and characterization of enzymes involved in nematode steroid metabolism, evaluation of a broader array of steroid biosynthesis inhibitors upon nematodes, and elucidation of the specific metabolic steps and the function of the 4-methylation pathway. Because *C. elegans* will be the first animal whose entire genome is sequenced, additional questions will be raised by the discovery of genes with sequences similar to those for steroid biosynthesis enzymes or receptors. All of these research fronts could accelerate the development of novel means for controlling parasitic nematodes.

Chitwood D.J. and Feldlaufer M.F. (1990) Ecdysteroids in axenically propagated *Caenorhabditis elegans* and culture medium. *Journal of Nematology* 22(4), 598-607.

Abstract: Ecdysteroids (insect molting hormones) from *Caenorhabditis elegans* were chromatographically purified and quantified by radioimmunoassay. Nematodes from semidefined medium contained the immunoreactive equivalent of 460 pg ecdysone per gram dry weight. Culture medium, however, contained the immunoreactive equivalent of 68 times the quantity within the nematodes. In a defined medium lacking immunoreactivity, *C. elegans* contained 520 pg ecdysone equivalents per gram dry weight but reproduced slowly. Reproduction of *C. elegans* in defined medium was enhanced by formulation in agar. Propagation of *C. elegans* in either agar-based or aqueous defined medium supplemented with [3 H]cholesterol of high specific activity failed to result in production of radiolabeled free ecdysteroids or polar or apolar ecdysteroid conjugates. Failure to demonstrate ecdysteroid biosynthesis in *C. elegans* raises questions about the ecdysteroids identified previously in nematodes being products of endogenous biosynthesis, a necessary condition for these compounds to be nematode hormones.

Chitwood D.J., Lozano R. and Lusby W.R. (1986) Recent developments in nematode steroid biochemistry. *Journal of Nematology* 18(1), 9-17.

Abstract: Current knowledge of steroid nutrition, metabolism, and function in free-living, plant-parasitic and animal-parasitic nematodes is reviewed, with emphasis upon recent investigation of *Caenorhabditis elegans*. A number of 4-desmethylsterols with a trans-A/B ring configuration can satisfy the steroid nutritional requirement in *C. elegans*, but sterols with a cis-A/B ring configuration or trans-A/B sterols with a 4-methyl group cannot. *C. elegans* removes methyl or ethyl substituents at C-24 of the plant sterols sitosterol, campesterol, stigmasterol, stigmasterol, and 24-methylene-cholesterol to produce various sterols with structures partially dependent upon that of the dietary sterol. Additional metabolic steps in *C. elegans* include reduction of $\Delta^2(2)$ - and Δ^4 -bonds, C-7 dehydrogenation, isomerization of a Δ^4 -bond to a Δ^1 -bond, and 4 α -methylation. An azasteroid and several long-chain alkyl amines interfere with the dealkylation pathway in *C. elegans* by inhibiting the $\Delta^2(2)$ -sterol reductase; these compounds also inhibit growth and reproduction in various plant-parasitic and animal-parasitic nematodes. A possible hormonal role for various steroids identified in nematodes is discussed.

Chitwood D.J., McClure M.A., Feldlaufer M.F., Lusby W.R. and Oliver J.E. (1987) Sterol composition and ecdysteroid content of the root-knot nematodes *Meloidogyne incognita* and *M. arenaria*. *Journal of Nematology* 19(3), 352-360.

Abstract: Free and esterified sterols of eggs of the root-knot nematodes *Meloidogyne incognita* races 2 and 3 and *M. arenaria* race 1 were isolated and identified by gas-liquid chromatography-mass spectrometry. The major sterols of eggs of each race were 24-ethylcholesterol (33.4-38.8% of total sterol), 24-ethylcholestanol (18.3-25.3%), 24-methylcholesterol (8.6-11.7%), 24-methylcholestanol (7.7-12.5%), and cholesterol (4.6-11.6%). Consequently, the major metabolic transformation performed by *Meloidogyne* females or eggs upon host sterols appeared to be saturation of the sterol nucleus. The free and esterified sterols of the same race did not differ appreciably, except for a slight enrichment of the steryl esters in cholesterol. Although the sterol composition of *Meloidogyne* eggs differed from that of other life stages of other genera of plant-parasitic nematodes, the three *Meloidogyne* races could not be distinguished from each other by their egg sterols. Ecdysteroids, compounds with hormonal function in insects, were not detected by radioimmunoassay in the *Meloidogyne* eggs either as free ecdysteroids or as polar conjugates.

Cleator M., Delves C.J., Howells R.E. and Rees H.H. (1987) Identity and tissue localization of free and conjugated ecdysteroids in adults of *Dirofilaria immitis* and *Ascaris suum*. *Molecular and Biochemical Parasitology* 25, 93-105.

Abstract: Adult males and females of the dog heartworm, *Dirofilaria immitis*, and of the swine parasite, *Ascaris suum*, were extracted, the free and polar conjugated ecdysteroid fractions separated and the latter hydrolysed enzymically. The ecdysteroids released by hydrolysis of the conjugates and the free hormones were analysed by radioimmunoassay, high-performance liquid chromatography on reversed phase and adsorption columns monitoring fractions by radioimmunoassay, and by gas-liquid chromatography/mass spectrometry (selected ion monitoring). In both species, males and females contained free and polar conjugated ecdysteroids, with evidence for the presence primarily of ecdysone and 20-hydroxyecdysone together with smaller amounts of 20,26-dihydroxyecdysone. Males and females of both species were then dissected into body fluid, reproductive system, gut and remaining body wall compartments, the ecdysteroids extracted, fractionated and analysed by radioimmunoassay and high-performance liquid chromatography monitoring fractions by radioimmunoassay. The results for both sexes in the two species were similar and indicated that ecdysteroids were not detectable in body fluids and that free ecdysteroids occurred in the reproductive system and the body wall, whereas polar conjugated ecdysteroids were detected in the reproductive system and the gut; a minor portion of the free ecdysteroids in *A. suum* was also apparently present in the gut. Further localization of the ecdysteroids in the body wall of *A. suum* females suggested that negligible immunoreactivity was associated with the circumpharyngeal nerve ring. The possible significance of the results is discussed.

Fleming M.W. (1987) Ecdysteroids during embryonation of eggs of *Ascaris suum*. *Comparative Biochemistry and Physiology* 87A(3), 803-805.

Abstract: 1. The optimal temperature for in vitro development of fertilized eggs of *Ascaris suum* was 24 degrees C. 2. Samples (2 X 10⁷) eggs) were obtained from in vitro embryonating cultures every 3 days for 4 weeks; lipids were extracted, partially purified, fractionated with HPLC and analyzed for ecdysteroids by radioimmunoassay. 3. Free ecdysone and 20-hydroxyecdysone (20-HE) were at low levels (less than 20 pg) in freshly excised eggs and rose to maximal values on day 6 of embryonation. 4. Conjugated ecdysone and conjugated 20-HE rose to maximal values on day 9. 5. Both free and conjugated ecdysteroids were undetectable from days 15 to 27 of cultivation. 6. These profiles indicate that ecdysteroids might have a selective role in nematode embryonation and/or tanning of the egg shell.

Fleming M.W. (1993) Ecdysteroids during development in the ovine parasitic nematode, *Haemonchus contortus*. *Comparative Biochemistry and Physiology* 104B(4), 653-655.

Abstract: 1. Samples of embryonating eggs, infective larvae, fourth-stage larvae, young adults, and male and female mature adults of *Haemonchus contortus* were collected for steroids analysis; lipids were extracted, partially purified, fractionated with HPLC, and analyzed for ecdysteroids by radioimmunoassay. 2. Free ecdysone and 20-hydroxyecdysone were detected in all samples; maximal concentrations occurred in the fourth-stage larvae and young adults. 3. Conjugated ecdysone and 20-hydroxyecdysone were detected in all samples but the infected larvae; maximal concentrations occurred in the fourth-stage larvae and young adults. 4. Patterns of ecdysteroid occurrence support regulatory roles in embryogenesis, cuticular deposition, and gonadogenesis. 5. Relative concentrations of ecdysteroids are comparable to those previously reported in eggs and adults of *Ascaris suum*.

Horn D.H.S., Wilkie J.S. and Thomson J.A. (1974) Isolation of β -ecdysone (20-hydroxyecdysone) from the parasitic nematode *Ascaris lumbricoides*. *Experientia* 30(10), 1109-1110.

No Abstract.

Mendis A.H.W., Rose M.E., Rees H.H. and Goodwin T.W. (1983) Ecdysteroids in adults of the nematode, *Dirofilaria immitis*. *Molecular and Biochemical Parasitology* 9, 209-226.

Abstract: Adult males and females of the dog heartworm, *Dirofilaria immitis*, were extracted separately and, following separation of the free and conjugated ecdysteroid fractions, the conjugates were hydrolysed enzymically.

Both the ecdysteroids released by hydrolysis of the conjugates and the free hormones were further purified and analysed by a combination of radioimmunoassay, thin-layer chromatography and high-performance liquid chromatography monitoring fractions by radioimmunoassay, and by gas-liquid chromatography/mass spectrometry (selected ion monitoring). Both males and females contained free and conjugated ecdysteroids. Evidence was obtained for the presence of ecdysone, 20-hydroxyecdysone, 20,26-dihydroxyecdysone and possibly ponasterone A. The possible parallel between ecdysteroid endocrinology in nematodes and insects is discussed.

Mercer J.G., Munn A.E. and Rees H.H. (1988) *Caenorhabditis elegans*: occurrence and metabolism of ecdysteroids in adults and dauer larvae. *Comparative Biochemistry and Physiology* **90B**(2), 261-267.

Abstract: 1. Ecdysteroids were detected in extracts of egg-producing adult *Caenorhabditis elegans*, in dauer larvae and in dietary bacteria. 2. Similar concentrations of free ecdysteroids were recorded in adults and larvae, although the two life cycle stages differed in their ratio of ecdysone: 20-hydroxyecdysone. 3. Patent adults metabolized [³H]ecdysone into apolar products and putative [³H]ecdysone 22-phosphate.

O'Hanlon G.M., Howarth O.W. and Rees H.H. (1987) Identification of ecdysone 25-O-β-D-glucopyranoside as a new metabolite of ecdysone in the nematode *Parascaris equorum*. *Biochemical Journal* **248**, 305-307.

Abstract: A major metabolite of [³H]ecdysone injected into adults of the nematode *Parascaris equorum* maintained in vitro for 48 h was secreted into the culture medium. The compound could be hydrolysed with a crude hydrolase preparation from *Helix pomatia*, yielding ecdysone. Sufficient quantity of this material for identification was produced by administration of ecdysone to the parasites. The resulting compound was purified by h.p.l.c. and identified as ecdysone 25-O-beta-D-glucopyranoside by n.m.r. spectroscopy and by fast atom bombardment mass spectrometry of the conjugate and of the sugar released by enzymic hydrolysis. The significance of formation of the glucoside is uncertain.

O'Hanlon G.M., Cleator M., Mercer J.G., Howells R.E. and Rees H.H. (1991) Metabolism and fate of ecdysteroids in the nematodes *Ascaris suum* and *Parascaris equorum*. *Molecular and Biochemical Parasitology* **47**, 179-188.

Abstract: When injected with [³H]ecdysone and maintained in vitro, the parasitic nematodes, *Ascaris suum* and *Parascaris equorum* each produced a series of polar and relatively apolar metabolites. *A. suum* metabolised the compound into ecdysoneic acid ([³H]EOIC), ecdysone 25-glucoside ([³H]E25gluc), putative ecdysone 22-phosphate ([³H]E22P) and a series of at least six relatively apolar metabolites. All of these, except ecdysoneic acid, were hydrolysed by a crude enzyme preparation from *Helix pomatia*, releasing ecdysone. In a similar study, *P. equorum* produced ecdysone 25-glucoside, putative ecdysone 22-phosphate and a series of relatively apolar compounds all of which were hydrolysed by *H. pomatia* enzymes, releasing ecdysone. [³H]Ecdysone 25-glucoside was the most abundant single metabolite in both species, and in *P. equorum*, at least, was released into the culture medium in relatively large amounts. Apolar metabolites were present in worm samples and were the major, if not the only radiolabelled compounds detected in eggs of both species. Data indicated a metabolic relationship between some of the apolar conjugates found in both nematode species and ecdysone 25-glucoside.

Rogers W.P. (1973) Juvenile and moulting hormones from nematodes. *Parasitology* **67**, 105-113.

Abstract: Juvenile hormone and its analogues inhibited growth of *Panagrellus redivivus* and of the free-living stages of *Haemonchus contortus*. Substances extracted from nematodes had similar actions and also showed the activity of juvenile hormone in insects. Partly purified material from 1 g of freeze-dried juveniles of *H. contortus* contained about 200 *Tenebrio* units.

5c. Cestodes

Mendis A.H.W., Rees H.H. and Goodwin T.W. (1984) The occurrence of ecdysteroids in the cestode, *Moniezia expansa*. *Molecular and Biochemical Parasitology* **10**, 123-138.

Abstract: The occurrence of free ecdysteroids in the sheep cestode, *Moniezia expansa*, was demonstrated. Significant amounts of conjugated ecdysteroids were not detected. Characterization of the free hormones by high-performance liquid chromatography monitoring fractions by radioimmunoassay, and by gas chromatography/mass spectrometry (selected ion monitoring) indicated the presence of ecdysone, 20-hydroxyecdysone and 20,26-dihydroxyecdysone. Analysis of the ecdysteroids by radioimmunoassay in segments along part of the strobila indicated that the anterior parts contained the greatest amount of hormone. GC/MS (SIM) analysis of the hormones in a strobilar segment containing the most mature proglottids suggested the presence of several ecdysteroid metabolites.

Mercer J.G., Munn A.E., Arme C. and Rees H.H. (1987) Analysis of ecdysteroids in different developmental stages of *Hymenolepis diminuta*. *Molecular and Biochemical Parasitology* **25**, 61-71.

Abstract: Prepatent and patent adult *Hymenolepis diminuta* from the intestines of rats, *H. diminuta* eggs recovered from the faeces of rats harbouring patent infections, and infective cysticercoids from the beetle intermediate host

were analysed for free and conjugated ecdysteroids. Adult worms and eggs contained both free ecdysteroids and hydrolysable polar conjugated ecdysteroids, with comparatively large amounts of immunoreactive material also being detected following hydrolysis of the possible apolar conjugated ecdysteroid fraction. Free ecdysteroids were not detected in the cysticercoid sample. The concentration of free ecdysteroids in *H. diminuta* eggs was higher than that detected in the tissues of the adult worms. Ecdysone and 20-hydroxyecdysone were the major identified compounds of the free ecdysteroid fraction, whereas in the hydrolysed polar conjugated ecdysteroid fraction these two compounds were accompanied by 20,26-dihydroxyecdysone. The free ecdysteroid fraction also contained comparatively large amounts of unidentified immunoreactive material.

5d. Trematodes

Basch P.F. (1986a) Immunocytochemical localization of ecdysteroids in the life history stages of *Schistosoma mansoni*. *Comparative Biochemistry and Physiology* **83A**(1) 199-202.

Abstract: 1. Sporocysts, cercariae and adults of *S. mansoni* exhibit focal immunoreactivity against anti-ecdysone serum in an indirect immunofluorescence assay. 2. In adult males immunoreactivity is limited to cell bodies and linear connections in the parenchyma surrounding the intestinal caeca. 3. In both unisexual and paired mature females part of the lining of the ootype is reactive, especially near the entrance of the vitelline duct; this demonstrates that females can make ecdysteroids without male contact. 4. Adult worms cultured completely in vitro show a similar pattern of reactivity. 5. Immunoreactivity is strong in cercariae, but is essentially absent in miracidia.

Basch P.F. (1986b) Internal chemical communication within flatworms. *Journal of Chemical Ecology* **12**(8), 1679-1686.

Foster J.M., Mercer J.G. and Rees H.H. (1992) Analysis of ecdysteroids in the trematodes *Schistosoma mansoni* and *Fasciola hepatica*. *Tropical Medicine and Parasitology* **43**(4), 239-244.

Abstract: Adult *Schistosoma mansoni* from experimentally infected mice and *Fasciola hepatica* recovered from ovine livers post mortem were analyzed for free and conjugated ecdysteroids by radioimmunoassay, high-performance liquid chromatography monitoring fractions by radioimmunoassay, and by capillary gas chromatography/mass spectrometry (selected ion monitoring). Both species contained ecdysone and 20-hydroxyecdysone as free ecdysteroids and as polar conjugates. *F. hepatica* also contained a polar conjugate of 2-deoxyecdysone. Evidence of apolar ecdysteroid conjugates was only obtained for *F. hepatica*. The free ecdysteroid-containing fraction of *S. mansoni* contained small amounts of unidentified immunoreactive material.

Nirde P., Torpier G., de Reggi M.L. and Capron A. (1983) Ecdysone and 20-hydroxyecdysone: new hormones for the human parasite *Schistosoma mansoni*. *FEBS Letters* **151**(2), 223-227.

Abstract: The insect moulting hormones, ecdysone and 20 hydroxyecdysone, were detected by the combined use of radioimmunoassay and high performance liquid chromatography in the human parasite *Schistosoma mansoni*. On day 11 after infection only the ecdysone form is present, but, on day 40 after infection the ratio between ecdysone and 20 hydroxyecdysone changes with anatomic localization of the adult worms in mammalian host. In the eggs, the ratio of these two hormones is identical to the ratio found in sexually mature worms located in mesenteric veins. These data demonstrate for the first time that *S. mansoni* synthesizes the steroid hormones ecdysone and 20 OH ecdysone which are potent molecules in stimulating growth and vitello-genesis of this gonochoric trematode.

Nirde P., de Reggi M.L., Tsoupras G., Torpier G., Fressancourt P. and Capron A. (1984) Excretion of ecdysteroids by schistosomes as a marker of parasite infection. *FEBS Letters* **168**(2), 235-240.

Abstract: Ecdysteroids produced by schistosomes are released in biological fluids of infected hosts. In the sera, the concentration of ecdysteroids correlates with the permissiveness of the host to schistosome infection and its detection is available in the absence of positive parasitological tests. In the urine, ecdysteroid concentration decreases markedly after chemotherapy. 20-Hydroxyecdysone and its epimer were identified in the urine of infected patients using mass spectrometry. These data demonstrate for the first time that ecdysteroids are released by organisms. Moreover, they are potent molecules of parasite infection and can be used for parasite diagnosis.

Schallig H.D.F.H., Young N.J., Magee R.M., de Jong-Brink M. and Rees H.H. (1991) Identification of free and conjugated ecdysteroids in cercariae of the schistosome *Trichobilharzia ocellata*. *Molecular and Biochemical Parasitology* **49**, 169-176.

Abstract: Extracts of cercariae of the avian schistosome *Trichobilharzia ocellata* were analysed for the presence of ecdysteroids by radioimmunoassay, high-performance liquid chromatography monitoring fractions by radioimmunoassay, and gas chromatography/mass spectrometry (selected ion monitoring). Both free ecdysteroids and polar conjugated ecdysteroids were detected in the cercarial extracts. The free ecdysteroid fraction, as well as the hydrolysed polar conjugated ecdysteroid fraction, contained both ecdysone and 20-hydroxyecdysone in approximately equal amounts. The amount of ecdysteroids detected is comparable to those found in other

platyhelminths. A possible role for the ecdysteroids in the development of the parasite and/or the interactions between the parasite and its intermediate host, the freshwater snail *Lymnaea stagnalis*, is discussed.

6. NONSTEROIDAL ECDYSTEROID AGONISTS FROM PLANTS

Dinan L. (1995) A strategy for the identification of ecdysteroid receptor agonists and antagonists from plants. *European Journal of Entomology* **92**, 271-283.

Abstract: A strategy is described for receptor-based phytochemical screening of plant extracts for ecdysteroid agonists and antagonists. Milligram amounts of seed are methanol extracted. Lipids and pigments are removed by hexane partitioning. Agonist and antagonist activities are detected with a microplate-based specific bioassay using the *Drosophila melanogaster* ecdysteroid-responsive BII cell line. Extracts are also screened with ecdysteroid-specific RIAs to identify extracts containing phytoecdysteroids. Over 1,700 species of plant have been screened in this way so far. Extracts are being sought which (i) contain large amounts of phytoecdysteroid, (ii) which contain novel phytoecdysteroids, (iii) which contain non-steroidal agonists and (iv) which contain antagonists. The aim of this paper is to describe the rationale behind the strategy, to describe its operation and to present, as an example, the results obtained with members of the Poaceae and of one genus, *Briza*, in particular. It is hoped that this approach will result in the identification of new sources of phytoecdysteroids, provide new phytoecdysteroid structures for structure/activity relationship studies, throw light on the phylogenetic distribution of phytoecdysteroids in the plant kingdom and provide useful agonists and antagonists for the investigation of ecdysteroid receptor function and as potential lead compounds for new classes of insect control agents.

Dinan L., Whiting P., Bourne P. and Coll J. (2001) 8-O-Acetyl harpagide is not an ecdysteroid agonist. *Insect Biochemistry and Molecular Biology* **31**, 1077-1082.

Abstract: We have reinvestigated the activity of 8-O-acetylharpagide, an iridoid glucoside, as an ecdysteroid agonist. Elbrecht et al. (*Insect Biochem. Mol. Biol.* 26 (1996) 519) isolated a preparation of this compound from *Ajuga reptans* L. and ascribed ecdysteroid agonist activity on the basis of the induction of an ecdysteroid-like response in *Drosophila melanogaster* KcO cells, the displacement of [3H]ponasterone A from the *Drosophila* receptor and the activation of an ecdysteroid-regulated gene in a transactivation assay. We provide evidence that the agonist activity derives from contaminating ecdysteroids; *A. reptans* is a species rich in ecdysteroids. Purified 8-O-acetylharpagide is not active in the *D. melanogaster* B(II) cell bioassay, neither as an agonist nor as an antagonist, nor does it displace [3H]ponasterone A from dipteran or lepidopteran ecdysteroid receptor complexes.

Elbrecht A., Chen Y., Jurgens T., Hensens O.D., Zink D.L., Beck H.T., Balick M.J. and Borris R. (1996) 8-O-Acetylharpagide is a nonsteroidal ecdysteroid agonist. *Insect Biochemistry and Molecular Biology* **26**(6), 519-523.

Abstract: We have identified a novel nonsteroidal ecdysteroid agonist. This compound was isolated from a methanol extract of *Ajuga reptans* L. (Lamiaceae) and the structure was identified by spectroscopic methods as 8-O-acetylharpagide. We have characterised this compound as an ecdysteroid agonist in a transactivation assay using beta-galactosidase as the reporter gene regulated by ecdysteroid response elements. In this assay, 8-O-acetylharpagide has an EC50 of 22 microM. The compound also competes with tritiated-ponasterone A for binding to the *Drosophila* ecdysteroid receptor. Finally, it induces differentiation of *Drosophila* Kc cells as would be expected of an ecdysteroid agonist. This iridoid glycoside is common to several plant species and may play a role in the natural defense mechanisms of plants.

Oberdörster E., Clay M.A., Cottam D.M., Wilmot F.A., McLachlan J.A. and Milner M.J. (2001) Common phytochemicals are ecdysteroid agonists and antagonists: a possible evolutionary link between vertebrate and invertebrate steroid hormones. *Journal of Steroid Biochemistry and Molecular Biology* **77**, 229-238.

Abstract: Many plant compounds are able to modulate growth and reproduction of herbivores by directly interacting with steroid hormone systems. In insects, several classes of phytochemicals, including the phytoestrogens, interfere with molting and reproduction. We investigated whether the anti-ecdysone activity may be due to interaction with the ecdysone receptor (EcR) using a reporter-gene assay and a cell differentiation assay of an ecdysone-responsive cell line, Cl.8+. We tested rutin (delays molt in insects); four flavones: luteolin and quercetin (metabolites of rutin), and apigenin and chrysin; and three non-flavones, coumestrol and genistein (both estrogenic) and tomatine (alters molt in insects). None of the phytochemicals tested were ecdysone agonists in the reporter-gene assay, but the flavones were able to significantly inhibit EcR-dependent gene transcription. In the Cl.8+ cells, quercetin and coumestrol were mixed agonists/antagonists, while genistein, tomatine and apigenin showed a synergistic effect with ecdysteroid in the reduction of cell growth. We suggest that the rutin effects on molting in insects are most likely due to the metabolites, luteolin or quercetin, while tomatine acts via a non-EcR pathway. Flavones not only interact with EcR and estrogen receptor (ER), but also signal nitrogen-fixing bacteria to form root nodules. The NodD protein which regulates this symbiosis has two ligand-binding domains similar to human ERalpha. The evolutionary significance of these findings are discussed.

7. ECDYSTEROID ANTAGONISTS FROM PLANTS

Dinan L. (1995) A strategy for the identification of ecdysteroid receptor agonists and antagonists from plants. *European Journal of Entomology* **92**, 271-283.

Abstract: A strategy is described for receptor-based phytochemical screening of plant extracts for ecdysteroid agonists and antagonists. Milligram amounts of seed are methanol extracted. Lipids and pigments are removed by hexane partitioning. Agonist and antagonist activities are detected with a microplate-based specific bioassay using the *Drosophila melanogaster* ecdysteroid-responsive BII cell line. Extracts are also screened with ecdysteroid-specific RIAs to identify extracts containing phytoecdysteroids. Over 1,700 species of plant have been screened in this way so far. Extracts are being sought which (i) contain large amounts of phytoecdysteroid, (ii) which contain novel phytoecdysteroids, (iii) which contain non-steroidal agonists and (iv) which contain antagonists. The aim of this paper is to describe the rationale behind the strategy, to describe its operation and to present, as an example, the results obtained with members of the Poaceae and of one genus, *Briza*, in particular. It is hoped that this approach will result in the identification of new sources of phytoecdysteroids, provide new phytoecdysteroid structures for structure/activity relationship studies, throw light on the phylogenetic distribution of phytoecdysteroids in the plant kingdom and provide useful agonists and antagonists for the investigation of ecdysteroid receptor function and as potential lead compounds for new classes of insect control agents.

Dinan L., Whiting P., Alfonso D. and Kapetanidis I. (1996) Certain withanolides from *Iochroma gesnerioides* antagonize ecdysteroid action in a *Drosophila melanogaster* cell line. *Entomologia Experimentalis et Applicata* **80**, 415-420.

Abstract: Sixteen withanolides isolated from *Iochroma gesnerioides* (Kunth) Miers (Solanaceae) have been assessed for their activities as ecdysteroid agonists and antagonists. None of the compounds showed any agonistic activity, but several showed significant antagonistic activity. With a 20-hydroxyecdysone concentration of 5 times 10^{-8} M, the ED₅₀ values for 2,3-dihydro-3ξ-methoxywithaferin A, 2,3-dihydro-3ξ-methoxywithacnistine, 2,3-dihydro-3ξ-methoxyiochromolide and 2,3-dihydro-3ξ-hydroxyξwithacnistine are 3.5 times 10^{-5} M, 1 times 10^{-5} M, 5 times 10^{-6} M and 2.5 times 10^{-6} M, respectively.

Dinan L., Whiting P., Sarker S.D., Kasai R. and Yamasaki K. (1997) Cucurbitane-type compounds from *Hemsleya carnosiflora* antagonize ecdysteroid action in the *Drosophila melanogaster* B_{II} cell line. *Cellular and Molecular Life Sciences* **53**, 271-274.

Abstract: The ecdysteroid agonist and antagonist activities of 3 cucurbitanes, 2 cucurbitane glycosides and 2 cucurbitacins isolated from *Hemsleya carnosiflora* (Cucurbitaceae) have been determined in the *Drosophila melanogaster* BII bioassay. Carnosiflogenins A and C and carnosiflosides II and VI possess antagonistic activity. Carnosiflogenin A was also found to induce the formation of spindle-shaped cells with high frequency in both the agonist and antagonist assays. At 10^{-3} M, carnosiflogenins B and C were cytotoxic, 23,24-Dihydrocucurbitacin F and 25-acetoxy-23,24-dihydrocucurbitacin F are also antagonistic at high concentrations. The concentration dependencies of the antagonistic activities of these two cucurbitacins, carnosiflosides II and VI and carnosiflogenin C are presented. The biological and ecological significance of these results are discussed in relationship to the concentrations present in the rhizomes of *H. carnosiflora*.

Dinan L., Whiting P., Girault J.-P., Lafont R., Dhadialla T.S., Cress D.E., Mugat B., Antoniewski C. and Lepasant J.-A. (1997) Cucurbitacins are insect steroid hormone antagonists acting at the ecdysteroid receptor. *Biochemical Journal* **327**, 643-650.

Abstract: Two triterpenoids, cucurbitacins B and D, have been isolated from seeds of *Iberis umbellata* (Cruciferae) and shown to be responsible for the antagonistic activity of a methanolic extract of this species in preventing the 20-hydroxyecdysone (20E)-induced morphological changes in the *Drosophila melanogaster* BII permanent cell line. With a 20E concentration of 50 nM, cucurbitacins B and D give 50% responses at 1.5 and 10 microM respectively. Both cucurbitacins are able to displace specifically bound radiolabelled 25-deoxy-20-hydroxyecdysone (ponasterone A) from a cell-free preparation of the BII cells containing ecdysteroid receptors. The K_d values for cucurbitacins B and D (5 and 50 microM respectively) are similar to the concentrations required to antagonize 20E activity with whole cells. Cucurbitacin B (cucB) prevents stimulation by 20E of an ecdysteroid-responsive reporter gene in a transfection assay. CucB also prevents the formation of the *Drosophila* ecdysteroid receptor/Ultraspiracle/20E complex with the hsp27 ecdysteroid response element as demonstrated by gel-shift assay. This is therefore the first definitive evidence for the existence of antagonists acting at the ecdysteroid receptor. Preliminary structure/activity studies indicate the importance of the Delta23-22-oxo functional grouping in the side chain for antagonistic activity.

Hexanorcucurbitacin D, which lacks carbon atoms C-22 to C-27, is found to be a weak agonist rather than an antagonist. Moreover, the side chain analogue 5-methylhex-3-en-2-one possesses weak antagonistic activity.

Dinan L., Savchenko T., Whiting P. and Sarker S.D. (1999) Plant natural products as insect steroid receptor agonists and antagonists. *Pesticide Science* 55, 331-335.

Abstract: Findings to date on plant secondary compounds which possess ecdysteroid-like or anti-ecdysteroid activities in an efficient and effective bioassay based on an ecdysteroid-responsive insect cell-line are summarised. Several novel antagonists have been identified, among which the cucurbitacins are the best characterised and have been shown to compete with ecdysteroids for the ligand binding site of the insect steroid hormone receptor. Certain withanolides, limonoids and resveratrol derivatives also antagonise 20-hydroxyecdysone action. Additionally, several new phytoecdysteroids have been isolated and identified. In common with all other ecdysteroids, these possess agonistic activity in the B_{II} bioassay. Extensive SAR studies based on the potencies of a large number of purified ecdysteroids have been performed and molecular (CoMFA) modelling used to characterise ecdysteroid binding to the ligand binding site of the receptor complex.

Dinan L., Savchenko T. and Whiting P. (2002a) Chemotaxonomic significance of ecdysteroid agonists and antagonists in the Ranunculaceae: phytoecdysteroids in the genera *Helleborus* and *Hepatica*. *Biochemical Systematics and Ecology* 30, 171-182.

Abstract: We present here a survey of ca. 100 species within 16 genera of the family Ranunculaceae for the presence of ecdysteroid agonist and antagonist activities in methanolic seed extracts. The levels of phytoecdysteroids (agonists) have been quantified by radioimmunoassay and bioassay. A few samples possess weak antagonistic activity. Phytoecdysteroids are most prominently associated with the genus *Helleborus*. In this genus, species fall into two distinct classes: those with low or undetectable ecdysteroid levels and those with high ecdysteroid levels. The relationship between ecdysteroid levels and the biology of the plants in this genus is discussed. Additionally, the extract of *Hepatica triloba* Chaix seeds contains a significant level of phytoecdysteroids. Several other species contain low levels of phytoecdysteroids, as detected by radioimmunoassay. Together with our previous data on the genera *Anemone* and *Pulsatilla*, this survey allows us to present an overview of the distribution of ecdysteroids in this family.

Keckeis K., Sarker S.D. and Dinan L. (2000) Resveratrol-type oligostilbenes from *Iris clarkei* antagonize 20-hydroxyecdysone action in the *Drosophila melanogaster* B_{II} cell line. *Cellular and Molecular Life Sciences* 57, 333-336.

Abstract: Bioassay-guided high-performance liquid chromatography analysis of a MeOH extract of *Iris clarkei* seeds yielded the resveratrol-type oligomeric stilbenes, ampelopsin B and alpha-viniferin, which antagonize the action of 20-hydroxyecdysone; with a 20-hydroxyecdysone concentration of 50 nM, the ED₅₀ values were 33 microM and 10 microM, respectively. The structures of these compounds were determined by spectroscopic analysis, notably ultraviolet, liquid secondary ion mass spectrometry and modern one- and two-dimensional nuclear magnetic resonance techniques.

Koreeda M., Nakanishi K. and Goto M. (1970) Ajugalactone, an insect moulting inhibitor as tested by the *Chilo* dipping method. *Journal of the American Chemical Society* 92(25), 7512-7513.
No Abstract.

Kubo I., Klocke J.A. and Asano S. (1981) Insect ecdysis inhibitors from the East African medicinal plant *Ajuga remota* (Labiatae). *Agricultural and Biological Chemistry* 45, 1925-1927.
No Abstract.

Meng Y., Bourne P.C., Whiting P., Šik V. and Dinan L. (2001) Identification and ecdysteroid antagonist activity of three oligostilbenes from the seeds of *Carex pendula* (Cyperaceae). *Phytochemistry* 57(3), 393-400.

Abstract: Methanolic extracts of seeds of several (*Carex* species) were found to antagonise the action of 20-hydroxyecdysone in the *Drosophila melanogaster* microplate-based B(II) cell bioassay. Bioassay-guided HPLC analysis of seeds of *Carex pendula* (drooping sedge) provided one previously unknown tetrastilbene (cis-miyabenol A) and two known oligostilbenes (kobophenol B and cis-miyabenol C) as the biologically active compounds (EC₅₀ values were 31, 37 and 19 microM, respectively, vs. 5 x 10⁻⁸ M 20-hydroxyecdysone). The structures and relative stereochemistries of these compounds were deduced by comprehensive ID- and 2D-NMR experiments. These compounds are isolated from *Carex pendula* for the first time. In vitro experiments with dipteran and lepidopteran ecdysteroid receptor proteins demonstrate that the oligostilbenes are able to compete with radiolabelled ecdysteroid ([³H]ponasterone A) for occupancy of the ligand binding site. IC₅₀/K_i values are similar to the EC₅₀ values obtained in the B(II) bioassay.

Oberdörster E., Clay M.A., Cottam D.M., Wilmot F.A., McLachlan J.A. and Milner M.J. (2001) Common phytochemicals are ecdysteroid agonists and antagonists: a possible evolutionary link between vertebrate and invertebrate steroid hormones. *Journal of Steroid Biochemistry and Molecular Biology* **77**, 229-238.

Abstract: Many plant compounds are able to modulate growth and reproduction of herbivores by directly interacting with steroid hormone systems. In insects, several classes of phytochemicals, including the phytoestrogens, interfere with molting and reproduction. We investigated whether the anti-ecdysone activity may be due to interaction with the ecdysone receptor (EcR) using a reporter-gene assay and a cell differentiation assay of an ecdysone-responsive cell line, Cl.8+. We tested rutin (delays molt in insects); four flavones: luteolin and quercetin (metabolites of rutin), and apigenin and chrysin; and three non-flavones, coumestrol and genistein (both estrogenic) and tomatine (alters molt in insects). None of the phytochemicals tested were ecdysone agonists in the reporter-gene assay, but the flavones were able to significantly inhibit EcR-dependent gene transcription. In the Cl.8+ cells, quercetin and coumestrol were mixed agonists/antagonists, while genistein, tomatine and apigenin showed a synergistic effect with ecdysteroid in the reduction of cell growth. We suggest that the rutin effects on molting in insects are most likely due to the metabolites, luteolin or quercetin, while tomatine acts via a non-EcR pathway. Flavones not only interact with EcR and estrogen receptor (ER), but also signal nitrogen-fixing bacteria to form root nodules. The NodD protein which regulates this symbiosis has two ligand-binding domains similar to human ER α . The evolutionary significance of these findings are discussed.

Sarker S.D., Whiting P., Lafont R., Girault J.-P. and Dinan L. (1997) Cucurbitacin D from *Cercidiphyllum japonicum*. *Biochemical Systematics and Ecology* **25**(1), 79-80.

Sarker S.D., Savchenko T., Whiting P., Šik V. and Dinan L.N. (1997) Two limonoids from *Turraea obtusifolia* (Meliaceae), prierianin and rohitukin, antagonise 20-hydroxyecdysone action in a *Drosophila* cell line. *Archives of Insect Biochemistry and Physiology* **35**, 211-217.

Abstract: Bioassay-assisted HPLC analyses yielded two prierianin-type limonoids, prierianin and rohitukin, from the seeds of *Turraea obtusifolia*, which act as antagonists of 20-hydroxyecdysone action in the *Drosophila melanogaster* BII cell line. With a 20-hydroxyecdysone concentration of 5×10^{-8} M, the ED₅₀ values for prierianin and rohitukin are 10^{-5} M and 1.25×10^{-4} M, respectively.

Sarker S.D., Lafont R., Šik V. and Dinan L. (1997) Arvenin I (cucurbitacin B 2-O- β -D-glucopyranoside) from *Iberis umbellata*. *Biochemical Systematics and Ecology* **25**, 365-366.

No Abstract.

Sarker S.D., Whiting P., Dinan L., Šik V. and Rees H.H. (1999) Identification and ecdysteroid antagonist activity of three resveratrol trimers (suffruticosols A, B and C) from *Paeonia suffruticosa*. *Tetrahedron* **55**, 513-524.

Abstract: Bioassay-guided HPLC analysis of the seeds of *Paeonia suffruticosa* has afforded three novel resveratrol trimers (suffruticosol A, suffruticosol B and suffruticosol C), together with cis-resveratrol and paeoniflorin. The structures of these new compounds have been elucidated mainly by comprehensive 1D- and 2D-NMR experiments. Resveratrol and its oligomers are active as ecdysteroid antagonists (ED₅₀ values = 10 to 50 μ M vs. 5×10^{-8} M 20-hydroxyecdysone) in the *Drosophila melanogaster* BII bioassay. The activities of other “pseudo-oestrogens” in this bioassay have also been assessed.

Sarker S.D., Whiting P., Šik V. and Dinan L. (1999) Ecdysteroid antagonists (cucurbitacins) from *Physocarpus opulifolius*. *Phytochemistry* **50**(7), 1123-1128.

Abstract: Methanolic extracts of seeds of 4 species in the genus *Physocarpus* antagonise the action of the insect steroid hormone 20-hydroxyecdysone on a *Drosophilamelanogaster* permanent cell line. The active components in the extract of *P. opulifolius* (ninebark) have been identified as cucurbitacin D, cucurbitacin F and 3-epi-isocucurbitacin D. The potencies of the individual cucurbitacins have been determined as 5×10^{-7} , 8×10^{-7} and 7×10^{-6} M, respectively (versus 5×10^{-8} M 20-hydroxyecdysone). The distribution of antagonistic activity in plants of *P. opulifolius* has been assessed and HPLC/bioassay has been used to determine the chromatographic profiles of antagonist activity in parts of the growing plant

Zou C., Liu G., Liu S., Liu S., Song Q., Wang J., Feng Q., Su Y. and Li S. (2017) Cucurbitacin B acts as a potential insect growth regulator by antagonizing 20-hydroxyecdysone. *Pest Management Science* doi: 10.1002/ps.4817.

Abstract:

Background: 20-Hydroxyecdysone (20E), a crucial insect steroid hormone, can bind to its cognate nuclear receptor composed of ecdysone receptor (EcR) and ultraspiracle (USP) to activate expression of 20E-response genes, enabling subsequent metamorphosis. In this study, we tried to find out which steroid-like compounds can block insect metamorphosis effectively and provide useful information for biopesticide study. For this purpose, we

screened 126 steroid-like compounds for possible 20E antagonists using a dual-luciferase reporter assay with *Drosophila melanogaster* Kc and *Bombyx mori* Bm12 cells.

Results: Among 126 steroid-like compounds, three cucurbitacins (CucB, D and E) were identified as 20E antagonists in both Kc and Bm12 cells. Notably, CucB caused significant molting defects and mortality in both *B. mori* and *D. melanogaster* larvae, and dramatically hindered larval growth of *Helicoverpa armigera* by its anti-feeding activity.

Conclusion: In vivo and in vitro experiments demonstrate that CucB acts as a potential insect growth regulator by antagonizing 20E activity and thus blocking molting and metamorphosis induced by 20E signaling.

8. References known about, but not yet obtained

Liu H., Huang Y., Zhang T., Wang Q. and Cheng X-Q. (2006) Studies on chemical constituents of *Paris delavayi* Franch. *Journal of China Pharmaceutical University* 37(5), 409-412.