CHAPTER II

HISTORICAL

CHEMISTRY OF MAGNOLIACEAE

Plants belonging to Magnoliaceae family have been extensive researched over the last decade. *Magnolia kobus* was the first species to be reported for its chemical constituents (81,82). Since then, there were 157 reports of constituents of Magnoliaceous plants till 1986. They were found to contain a wide range of chemical components while the major categories were alkaloids, sesquiterpene lactones and lignans.

List of components found in various species of Magnoliaceae are shown in Table I.

Table I Chemical investigation of Magnoliaceae.

Botanical Origin	Plant part	Chemical Substance	Reference
Elmerrillia papuana (Shltr.) Dandy	bark	Liriodenine	11
		Norushinsunine	11
		N-methylushinsunine iodide	11
		Elmerrillicine	11
Liriodendron tulipifera	root bark	Costunolide	15
		Epitulipinolide	16
		Lipiferolide	17
		γ -Liriodenolide	17
		Tulipinolide	15,16
	cortex	Esculetin dimethyl ether	18
	discolored-sapwood	(+)-Lirioferine	19
	heartwood	(+)-Liriotulipiferine	19
		(-)-*N*-Acetylanonaine	20
		(-)-*N*-Acetylasimilobine	20

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Liriodendron tulipifera	heartwood	(-)-*N*-Acetylnornuciferine	20,21
		Asimilobine	21,22
		Dehydroglucine	21
		d-Glaucine-HCl	18,23
		Glucine	21
		Liriodenine	18,21,22,24
		O-Methylatheroline	21
		Norushisunine	21
		Syringaldehyde	21
		(+)-Syringaresinol	21
		(+)-Syringaresinol dimethyl ether	21
		(-)-Tuliferoline	20
	inner bark	Liriodendrin	26

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Liriodendron tulipifera	bark	$BzNHCH_2CH(OH)Ph$	25
		Lirionol	25
		O-Methyl-*N*-norlirinine	25
		(+)-Pinoresinol	25
		(+)-Syringaresinol	25
		Syringic acid Me ester	25
	leaf	Anonaine	27
		Armepavine	22
		Asimilobine	22
		Elemanolide (Epitulipdienolide)	28
		Epitulipinolide diepoxide	28
		γ -Liriodenolide	28
		d-Glaucine-HCl	23
		d-Isomerine	29
		Lipiferolide	28

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Liriodendron tulipifera	leaf	Lirinidine	27
		Lirinine	31
		Lirinine *N*-oxide	32
		Lirinine Me ether	32
		Liriodendritol	33
		Liriodenine	18,22
		Magnocurarine	22
		Magnoflorine	22
		N-Norarmepavine	22
		Nornuciferine	22
		d-Nornuciferine	29
		Remerine	27
		Remerine-*N*-oxide	27
		Roemerine	22
		Taxiphyllin	34
		Triglochinin	34
	seed	Octadecadienoic acid	35
		Sitosterol	35

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Liriodendron tulipifera	not specify	*d*-Caaverine	12
		Dehydroremerine	13
		Liridinine	14
		Liriodenine	12
Magnolia acuminata	leaf	Armepavine	22
		Asimilobine	22
		Liriodenine	22
		Magnocurarine	22
		Magnoflorine	22
		N-norarmepavine	22
		Nornuciferine	22
		Roemerine	22
	fruit	Cyanidin	36
		Peonidin	36
Magnolia ashei	leaf	Cyanidin	30

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia biloba	root bark	β -Eudesmol	37
		γ -Eudesmol	37
		Honokiol	37
		Magnolol	37
Magnolia coco	stem	Anolobine	38,39
		Magnoflorine	43
		Salicifoline	43
		Stepharine	38,39
	Not specify	Magnococline	40
		Oxoushinunine	43
Magnolia cordata	fruit	Cyanidin	36
Magnolia denudata Desr.	root	Magnoflorine	46
		Magnoflorine styphnate	46
	bark	1,8-Cineole	48
		Salicifoline chloride	45,47
		(+)-Terpinen-4-ol	48
		(-)- α -Terpineol	48

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia denudata Desr.	branchet	1,8-Cineol	48
		(+)-Terpinen-4-ol	48
		(-)- α -Terpineol	48
	leaf	Armepavine	22
		Asimilobine	22
		Burchellin	49
		β -Caryophyllene	48
		Cyanidin	30
		Denudatin A	49
		Denndatin B	49
		Denudatone	49
		Futoenone	49
		Liriodenine	22
		Magnocurarine	22,45
		Magnoflorine	22,46
		(+)-Nerolidol	48
		N-norarmepavine	22

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia denudata Desr.	leaf	Nornuciferine	22
		Roemerine	22
		Veraguensin	49
	flower	1,8-Cineol	48
	flower bud	1,8-Cineol	48
	fruit	Peonidin	36
Magnolia fraseri	stigma	Rutin	50
	Anther	Rutin	50
Magnolia fuscata	leaf	Magnolamine	51,53,54, 56,57,60, 61
		Magnoline	51,52,53
Magnolia grandiflora L.	root	Candicine	45,62
		Salicifoline	62
	root bark	Costunolide	71
		Reynosin	71
		Santamarine	71

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia grandiflora L.	wood	Anolobine	64
		Anonaine	64
		Liriodenine	64
		Minisperine(Chakrarine)	65
		N-nornuciferine	64
	bark	Magnocurarine	63
		Magnoflorine	45,63,66
		Magnolenin	67
		Magnolidin	67,68
		Magnosidin	67
		Mono-*O*-methyltetra hydrohonokiol	69
		Salicifoline	45,63
	leaf	Anonaine	64
		Bovolide	70
		Costunolide	71
		Costunolide diepoxide	71,72

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia grandiflora L.	leaf	Dihydrobovolide	70
		Liriodenine	64
		Magnograndiolide	72
		Melampomagnolide A	73
		Melampomagnolide B	73
		Pathenolide	71,72
	filament	Cyanidin	36
Magnolia grandiflora variety lanceolata	bark	Magnoflorine	74
Magnolia kachirachirai	wood	*D*-(+)-*N*-Norarmepavine	76
	leaf	Eupomatenoid 7	78
		(±)-Galbacin	78
		Kachirachirol A	79
		Kachirachirol B	79
		(±)-Licarin A	78
		Licarin B	78

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia kachirachirai	duramen	*d*-Glaucine	80
		Magnoflorine	80
		D-N-Norarmepavine	80
		d-N-Norglaucine	80
	not specify	Acuminatin	75
		Glaucine	43
		Magnoflorine	43
Magnolia kobus D.C.	inner bark	Syringin	86
	bark	Coniferin	86,87
		Magnoflorine	59
		Salicifoline chloride	59,88,89
		Syringin	86,87
	leaf	Armepavine	22
		Asimilobine	22
		Liriodenine	22
		Magnocurarine	22

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia kobus D.C.	leaf	Magnoflorine	22
		N-Norarmepavine	22
		Nornuciferine	22
		Roemerine	22
	petal	Rutin	90,91,92
	fruit	Cyanidin	36
		Peonidin	36
	not specify	Anethole	81,82
		I-Camphor	83
		1,8-Cineole	83
		Citral	81,82
		p-Cymene	83
		d-Limonene	83
		d-Nerolidol	83
		Salicifoline	45

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia kobus D.C.	bark	Magnofoline	59
variety borealis		Salicifoline	59
Koidz.	not specify	(-)-Camphor	85
		p-Cymene	85
		α-Terpineol	85
Magnolia liliflora	root	Salicifoline	95
	trunk	Salicifoline	95
	bark	Magnocurarine	95
		Salicifoline	95
	leaf	Liliflodione	96
		Liliflol A	96
		Liliflol B	96
		Liliflone	96
		Salicifoline	95
	flower	Ascorbic acid oxidase	97

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia liliflora	not specify	D-Coclaurine	93
		Futoenone	94
		(-)-Maglifloenone	94
		Magnocurarine	45
		Salicifolin	45
		β-Sitosterol	94
		Taspine	94
		(+)-Veraguensin	94
Magnolia	flower	Rutoside	98
macrophylla Michx.	leaf	Cyanidin	30
	dry bark	Magnolioside	99
Magnolia obovata	root	Anonaine	101
Thunb.		Asimilobine	101
		Glaucine	101
		Liriodenine	101
		Magnocurarine	101

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia obovata Thunb.	root	Obovanine	101
		Reticuline	101
	bark	Cryptomeridiol	102
		Eudesmols	102
		Honokiol	102,103, 105
		Machilol	106
		Magnocurarine	55
		Magnolol	103,105
	leaf	Anonaine	101
		Armepavine	22
		Asimilobine	22,101
		Glaucine	101
		Liriodenine	22,101
		Magnocurarine	22,101
		Magnoflorine	22
		N-Norarmepavine	22

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia obovata Thunb.	leaf	Nornuciferine	22
		Obavanine	101
		Obovatal	107
		Obovatol	107
		Reticuline	101
		Roemerine	22
		Rutin	108
	filament	Peonidin 3,5-diglucoside	36
		Rutoside	98
	not specify	Coniferin	87
		Magnocurarine	45,100
		Syringin	87
Magnolia officinalis Rehd. et Wils.	bark	Bornylmagnolol	109
		8,9-Dihydroxydihydro-honokiol	110
		8,9-Dihydroxydihydro-magnolol	110

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia officinalis Rehd. et Wils.	bark	8,9-Dihydroxy -7-methoxydihydrohonokiol	110
		Eudesmol	111
		Honokiol	93,103, 111-117
		Magnolol	93,103, 111-118
Magnolia parviflora	bark	Magnocurarine	58
		Magnoflorine	58
Magnolia pterocarpa	leaf	(-)-Eudesmin	119
		(+)-Fargesin	119
		Imperatorin	119
		di-Me terephthalate	119
		(+)-Sesamin	119
		β-Sitosterol	119

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia rostrata	bark	Honokiol	113
WW Smith		Magnolol	113
Magnolia	shoot	Methylchavicol	120
salicifolia Maxim.	twig	Citral	121
		Trans-anethole	121
	branchet	1,8-Cineole	120
		Citral-a	120
		Citral-b	120
	bark	Magnocurarine	22,45, 122
		Salicifoline	45,123
		Salicifoline chloride	122
	leaf	Armepavine	22
		Asimilobine	22
		Citral	121
		Liriodenine	22
		Magnocurarine	22

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia salicifolia Maxim.	leaf	Magnoflorine	22
		Methylchavicol	120
		N-Norarmepavine	22
		Nornuciferine	22
		Roemerine	22
		Trans-anethol	121
	bud	*Trans*-Asarone	104
		Asarylaldehyde	104
		(+)-Coclaurine	104
		D-(-)-*N*-methylcoclaurine	104
		Euasarone	104
		Eugenol	104
		cis-*O*-methyleugenol	104
		trans-*O*-methyleugenol	104
		Fenchone	104
		Magnosalicin	126

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia salicifolia Maxim.	bud	Magnosalin	104, 124, 125
		Magnoshinin	124
		β -Pinene	104
		d-Pinoresinol	104
		L-(+)-Reticuline	104
		Safrol	104
		Vanillic acid	104
		Veratric acid	104
		Yuzarine	104
	flower bud	Citral-a	120
		Citral-b	120
		d-Coclaurine	127
		Methylchavicol	120
		l-*N*-Methylcoclaurine	127
		Methyleugenol	120

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia salicifolia Maxim.	flower bud	*d*-Reticuline	127
		Safrol	120
		Yuzirine	127
	flower	Citral-a	120
		Citral-b	120
		Methylchavicol	120
		Methyleugenol	120
		Safrol	120
	fruit	Peonidin	36
Magnolia sieboldi	filament, fruit	Peonidin 3-rhamnoglucosido-5-glucoside	36
	not specify	15-acetoxycostunolide	128
Magnolia x soulangiana "lennei"	*leaf*	*Soulangianolide A*	*129*
		Soulangianolide B	129

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia soulangiana Soul.	flower	Rutoside	98
		Peonidin	36
	fruit	Peonidin	36
		Cyanidin	36
Magnolia sprengeri cv Diva	bark	Magnosprengesine	131
	leaf	Hydrocyanic acid	132
		Taxiphyllin	132
	not specify	Magnocurarine	130
		Salicifoline	130
Magnolia stellata	trunk, flower bud, branchet	Camphor	134
		1,8-Cineole	134
		P-Cymene	134
		Fenchone	134
		β -Myrcene	134
		β -Pinene	134
		γ -Terpinene	134
		α -Terpineol	134
		Terpinen-4-ol	134

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia stellata	bark	Salicifoline chloride	88
	leaf	δ -Cadinene	134
		α -Cadinol	134
		β -Caryophyllene	134
		Elemol	134
		Cyanidin	30
		Trans- α -farnesene	134
		Trans,trans-farnesol	134
		(+)-Trans-nerolidol	134
	not specify	Eudesmin	133
		Kobusin	133
		Magnostellin A	133
		Magnostellin B	133
		(+)-Piperitol	133
		Salicifoline	45
		Sesamin	133
		Vomifoliol	133

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia thompsoniana Hort.	flower	Rutoside	98
Magnolia tripetala	leaf	Armepavine	22
		Asimilobine	22
		Cyanidin	30
		Liriodenine	22
		Magnocurarine	22
		Magnoflorine	22
		N-Norarmepavine	22
		Nornuciferine	22
		Roemerine	22
	flower	Cyanidin	36
	fruit	Peonidin	36
Magnolia virginiana	fruit	Peonidin	36
		Cyanidin	36
Magnolia watsonii	not specify	Watsonol A	135
		Watsonol B	135

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Magnolia wilsonii Rehd.	bark	(+) δ -cadinol	136
		Magnocurarine	136
Magnolia yulan Desr.	petal	Rutoside	98
Magnolia yulan variety Soulangiana	petal	Quercetin	137
Manglictia chinglii	bark	Magnocurarine	138
Manglietia insignin	bark	Eudesmol	139
		Honokiol	139
		Magnocurarine	139
		Magnolol	139
Manglietia yuyuanensis Law.	bark	Anisaldehyde	140
		Borneol	140
		Camphor	140
		Citral	140
		β -Eudesmol	140
		Honokiol	140

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Manglietia yuyuanensis Law.	bark	Linalool	140
		Magnocurarine	140
		Magnolol	140
Michelia alba DC.	flower	Acetaldehyde	142
		Agarol	142
		Allocimene	142
		Bu pentanoate	142
		Camphor	141
		Et α-methylbutyrate	142
		EtOH	142,144
		Et 3-methylbutyrate	144
		Et propionate	142
		Limonene	141,142
		Linalool	141,143
		Me acetate	142
		Me anthranilate	141
		Me benzoate	142

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Michelia alba DC.	flower	Me butyrate	142,144
		Me isobutyrate	142,144
		Me lexanoate	142
		Me α-methylbutyrate	142-144
		Me 2-pentenoate	142
		Me propionate	142
		Myrcene	142
		Nerol	141
		Ocimene	142
		β-phellandrene	142
		α-pinene	141
		β-pinene	142
		Terpinolene	142
		Undecane	142
	not specify	Michelavine	42
		Oxoushinsuine	42
		Salicifoline	42
		Ushinsunine	42

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Michelia cathcartii	trunk bark	Lanuginosine	145
		Liriodenine	145
		Sitosterol	145
Michelia champaca	bark	Magnoflorine	77
		Oxoushinsunine	77
		Ushinsunine	77
	trunk bark	Liriodenine (Micheline B, Oxoushinsunine)	146-148
	root	Parthenolide	148
Michelia compressa	root bark	Compressanolide (Formosanolide)	141
		Costunolide	149
		Dihydroparthenolide	149
		Dihydroreynosin	149
		Lanuginolide	149
		Liriodenine	149

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Michelia compressa	root bark	Michelenolide	149
		Micheliolide	149
		Parthenolide	149
		Reynosin	149
		Santamarin	149
	heart wood	Michepressine	150
		Oxoushinsunine	44
		Ushinsunine	44,151
	bark	Magnoflorine iodide	152
		Michepressine	150
		Oxoushinsunine	44
		Oxyacanthine	152
		Tetrahydroberberine	152
		Tetrahydrojatrorrhizine	152
		Ushinsunine	44,151

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Michelia compressa variety Formosana	heart wood	Liriodenine (Micheline B, Oxoushinsunine)	41,153
		Magnoflorine	41
		Micheline A	153
		Ushinsunine	41
Michelia doltsopa	fruit	11,13-Dehydrolanuginolide	154
		Dihydroparthenolide	154
		Lanuginolide	154
Michelia excelsa	trunk bark	Liriodenine	145
		Sitosterol	145
	root bark	Liriodenine	145
		Sitosterol	145
Michelia figo Spreng	leaf	Magnolamine	155
		iso-Bu acatate	156
		(8Z,11Z,14Z)-8,11,14-heptadecatrien-2-one	156

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Michelia fuscata	bark	Magnocurarine	158
		Magnolamine	158
	leaf	Magnocurarine	158
		Magnoflorine	158
		Magnolamine	158,159
	seed	Albumins	160
		Globulins	160
		Glutelins	160
	not specify	Deacetyllanuginolide	157
		Dehydrolanuginolide	157
		Lipiferolide	157
		Michefuscalide	157
		Syringaresinol	157
Michelia hedyosperma Law.	not specify	(+)-Limonene	161
		Linalool	161
		Methyleugenol	161
		Safrole	161

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Michelia lanuginosa	root bark	Lanuginosine	145
		Liriodenine	145
	bark	11,13-Dehydrolanugi-nolide	163,164
		Dihydroparthenolide	164,165
		Lanuginolide	164,165
		Lanuginosine	145,162, 166
		Liriodenine	145
		Michelanugine	145
		Parthenolide	163,164
		Sinapaldehyde	164
	leaf	Lunuginosine	145
		Liriodenine	145
		Sitosterol	145
	not specify	Liriodenine	162
		Michelanugine	162

Table I Chemical investigation of Magnoliaceae. (continued)

Botanical Origin	Plant part	Chemical Substance	Reference
Talauma abovata Korth.	bark	Anolobine	145
		Asimilobine	167
		Lanuginosine	167
		Xylopine	167

BIOSYNTHESIS OF SESQUITERPENE LACTONES

Terpenes are defined as natural products whose structures may be devided into isoprene units (169). Normally, isoprene units arise from acetate *via* mevalonic acid and are branched chain, 5-carbon units containing 2 unsaturated bonds (168). The preliminary stages in the biosynthesis of isoprenoid compounds are shown in Figure 2.1.

Terpenes are formed by linkage, head to tail, of isoprene units most of which are classified by number of isoprene units as the following (171).

1. Hemiterpenes C_5H_8
2. Monoterpenes $C_{10}H_{16}$
3. Sesquiterpenes $C_{15}H_{24}$
4. Diterpenes $C_{20}H_{32}$
5. Sesterterpenes $C_{25}H_{40}$
6. Triterpenes $C_{30}H_{48}$
7. Tetraterpenes $C_{40}H_{64}$
8. Polyterpenes $(C_5H_8)n$

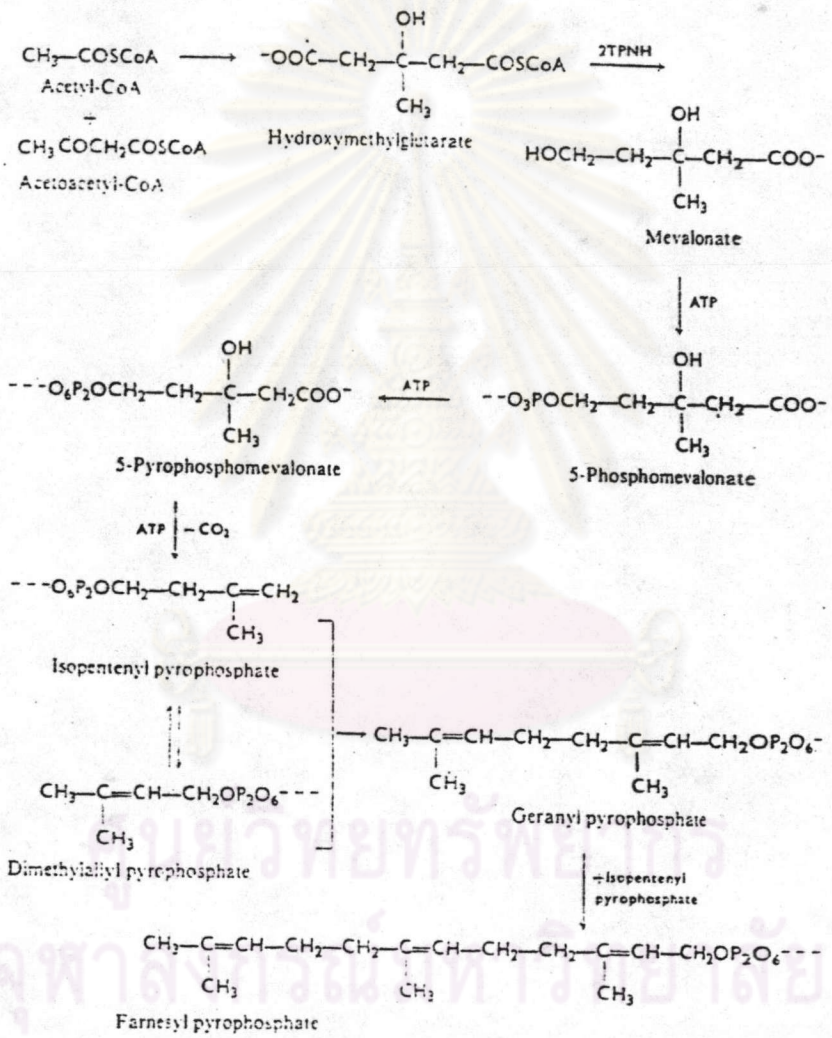


Figure 2.1 Preliminary stages in the biosynthesis of isoprenoid compounds (170).

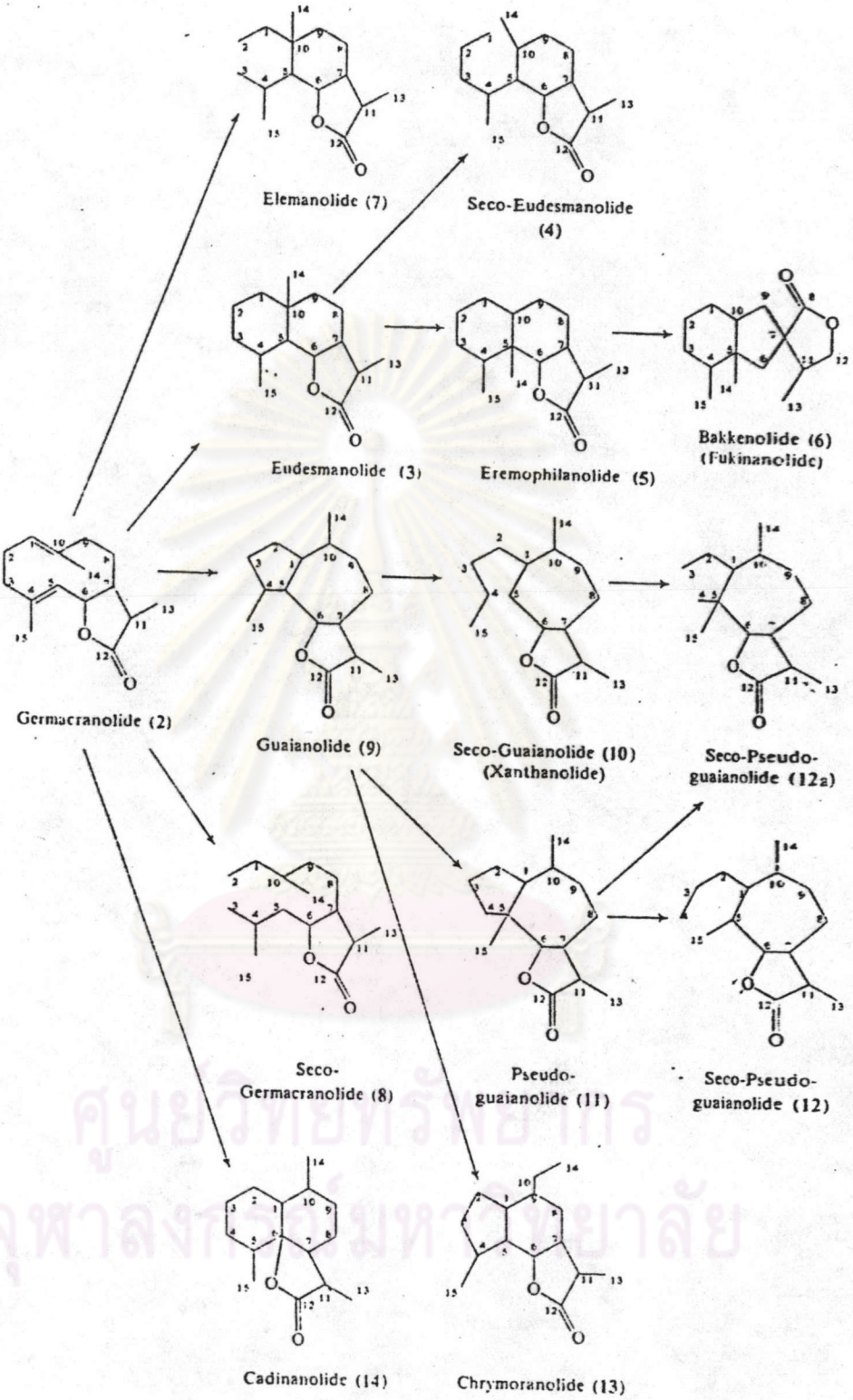
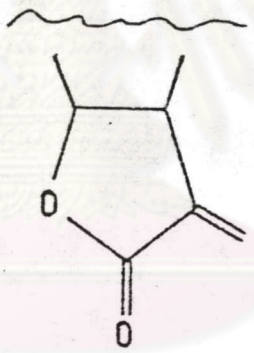


Figure 2.2 Types and biogenetic relationships of germacranolide-derived sesquiterpene lactones (173).

1. Definition of Sesquiterpene Lactones.

Sesquiterpene lactones are colorless, bitter, relatively stable, lyophilic constituents. They formed by oxidation of the " head " methyl group of farnesol the lactonic function commonly represents an α-methylene γ-lactone moiety [1]. The classification of sesquiterpene lactones is based on this carboxylic skeleton in which suffix "olide" refers to the lactonic function (172).



[1]

The majority of sesquiterpene lactones belong to this category which can be considered biogenitic derivatives of the largest class, the germacranolide [2]. The structural classes and names of various carboxylic ring systems are shown in figure 2.2 and the presumed biogenetic relationships are indicated by arrows.

2. Biogenesis of the Germacranolide Skeleton (173)

Cyclization of *trans*, *trans*-farnesyl pyrophosphate [15] result in the *trans*, *trans*-germacradiene intermediate [16] which by enzymatic oxidative modifications provides the germacranolides represented by its simplest member, costunolide [17]. From the germacradiene the different other skeletal types of sesquiterpene lactones shown in figure 2.3 can be derived.

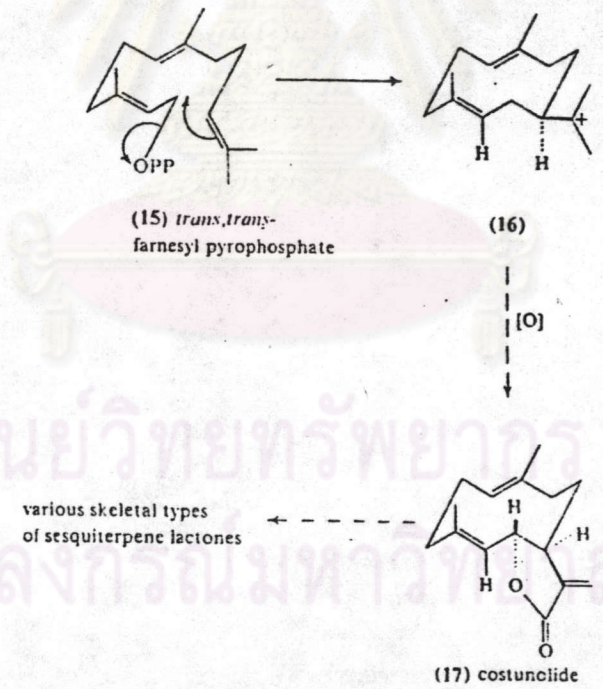


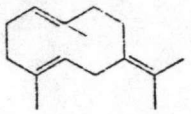
Figure 2.3 Biogenesis of the Germacranolide skeleton (173)

3. Biogenesis of the Lactone Ring (173).

Two possible biogenetic routes have been suggested for the formation of the lactone ring of these sesquiterprenoids.

One hypothetical intermediate *en* route from cation [16] to the lactones [1] and [24] is germacrene A [18] a naturally occurring hydrocarbon in which all non-olefinic carbons are allyllically activated for hydroxylation except C-8. Introduction of an oxygen function at C-12 in [18] to give alcohol [21] could either proceed *via* epoxide intermediate [19] or could involve the hydroperoxide [20], the latter being formed by an enzymatically-mediated reaction mimicking the reaction of singlet oxygen with olefins. In either case the process involves migration of a double bond from what was originally C-11, C-13 to C-11, c-12. Furthur oxidative modifications of [21] *via* aldehyde [22], acid [23] and hydroxylations at c-6 or c-8 would after lactonization give costunolide [17] or inunolide [24], respectively.

Germacrene B [25] should not be excluded as a possible precursor in lactone biosynthesis since C-8 hydroxylation in a sesquiterpene lactone precursor of type [25] would now be favored due to allylic activation of C-8.



[25]

Furthurmore, C-6 in [25] represents a doubly allylic carbon center favoring hydroxylation at this position over all other allylic carbons. This could possibly be the reason for predominant formation of C-6-oxygenated sesquiterpenoids. Sesquiterpene lactones of type [27] commonly cooccur with and are derived from furanosesquiterpenes [26] by autoxidation, suggesting that the lactones are also biogenetically derived from the furan ring adumbrated in figure 2.5 (173).

4. Biogenesis of Germacranolides (173).

The germacranolides present the largest group of sesquiterpene lactones with nearly 300 known naturally occuring members. The variety is mainly due to the unique configurational and configurational features and the reactivity of the cyclodecadiene skeleton. Configurationally isomeric germacranolides has led to a reclassification into four subgroups which are characterized by a cyclodecadiene skeleton with double bonds in the C-1,10- and c-4,5-position. In figure 2.6 the basic configurational types are shown.

The question remains unanswered whether the biosynthesis or the four cyclodecadiene subgroups follows independent biogenetic routes from the four possible configurationally isomeric farnesols or whether the configurational isomers [29] to [31] are formed from the germacranolide skeleton [28] by double bond isomerizations at a later stage of biosynthesis. Cooccurence of more than one skeletal type in the same plant species or genus and the fact that, at least in the melampolide series, all presently known compounds have an oxidized C-14 such that they are either aldehydes or carboxylic acid derivatives (esters or lactones) could be an indication of interconversion from one type of skeletal system to another during or after oxidation of C-14 and/or C-15. Besides enzymatically controlled process, spontaneous or phytochemical double bond isomerizations could be involved.

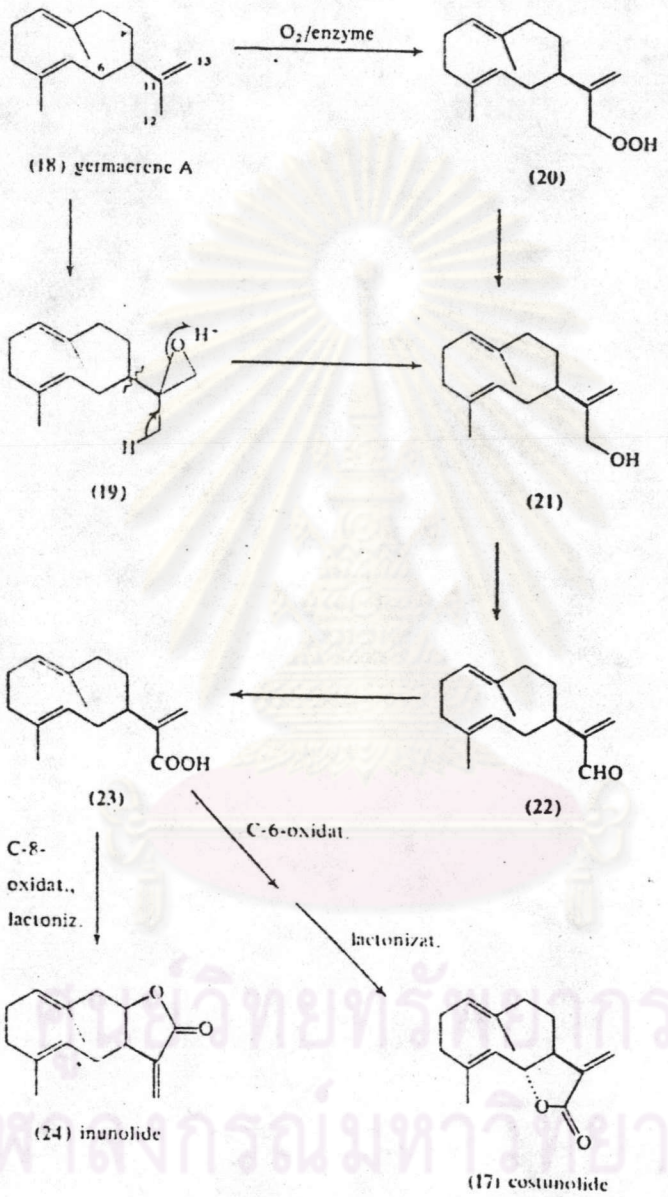


Figure 2.4 Biogenesis of the lactone ring (173).

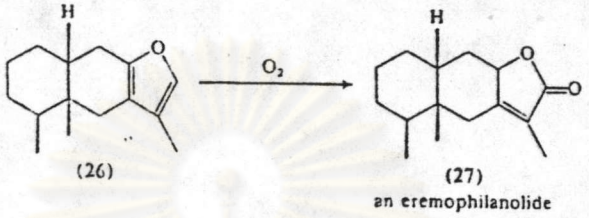


Figure 2.5 Biogenesis of the lactone ring *via* Furanosesquiterpene (173).

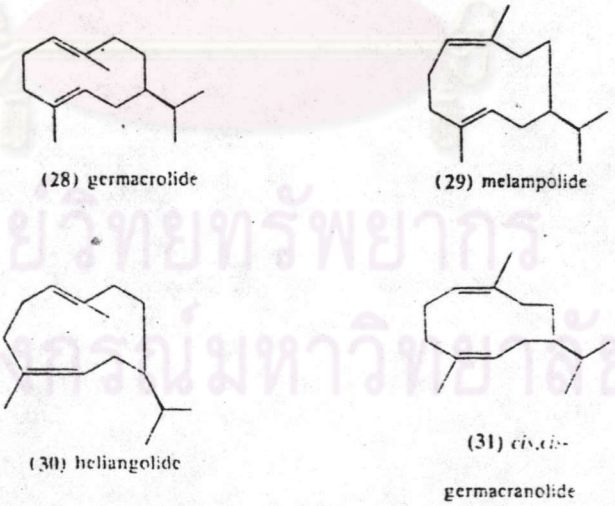


Figure 2.6 Configurational types of germacranolides(173).

BIOLOGICAL ACTIVITIES OF SESQUITERPENE LACTONES

1. Antitumor and Cytotoxic activity.

The earliest record of plants being recommended for what is believed to be cancer is the Ebers papyrus of Egypt dating from about 1550 B.C. Since that time there has been constantly expanding use, both popular and iatric, of plants for the treatment of cancer throughout the ages, until at the present time these is hardly an area of the world where plants in some forms are not administered for this disease. In the field of science, particular medical science, this aspect has been received considerable attention continuously. There are many review articles about anticancer agents from plants (174,198,234-236).

In a review of antineoplastic principles in plants, over 50 sesquiterpene lactones were evaluated for the growth-inhibitory potential against numerous tumor models (236). (see Appendix VII)

Almost 20 years after that time, cancer increases seriousness, over 60 sesquiterpene lactones were evaluated for the antitumor and cytostatic activity. (see Table II)

The structure-activity relationship among sesquiterpene lactones is very interested. Many investigators had studied on this topic. It shown that unsaturated α-exo

-methylene- γ -lactones ring (-O-CO-C=CH_2) conjugated with basic terpene carboxylic skeleton plays the main role for cytotoxicity of sesquiterpene lactones (175-183).

Although this moiety is necessary for cytotoxicity but it is optimal-activity. Many sesquiterpene lactones which have different structures show different degree of activity as the following evidences.

Eighteen novel sesquiterpene lactones were studied by tissue culture methods. It was noted that activity of germacranolide was higher than that of guaianolide (183).

Sesquiterpene lactones which incorporated a cyclopentanone, or α-methylene lactone (in addition to the - β -methylene- γ-lactone) appeared to produce enhanced cytotoxicity. None of the monofunctional sesquiterpene lactone containing only an α- β-unsaturated ester or cyclopentenone displayed significant activity (175,184,185). The sesquiterpene lactones were tested in tissue culture, all having in their molecule an α-methylene γ-lactone group,which had been described as conferring cytotoxic activity. Five of those lactones have a furanic ring in their molecules, whose presence gives them a higher cytotoxic activity (186). In a recent study cytotoxicity of some sesquiterpene lactones from *Eupatorium cannabinum* and related compounds, eupatoriopicrin, eupatoriopicrin acetonides, "substance 1" and hiyodorilactone E showed highest cytotoxicity (ID_{50} 1-2 mcg/ml) following 1 hour incubation. Moieties of the sesquiterpene

lactones (α-methylene- γ-butyrolactone, eupatolide, angelic and tiglic acid) and related compounds (alantolactoneand isoalantolactone) were less or not active at these concentrations. From the results of the experiments it can be concluded that the entire molecule (germacranolide ester) is necessary for optimal cytotoxicity *in vitro* and that cytotoxicity increases with decreasing hydrophilicity (187).

The study of bakkenolides from *Petasites albus*, *P. fragrans* and *P. hybridus* was so surprising (188). It was noted that bakkenolides-A a β-methylene lactone (which does not have an O=C-C=CH_2 system) gave result against cells derived from human carcinoma (H.Ep-2). Similar to this report, in another study comparison of cytotoxic activity of encelin and farinosin (176) disclosed that they are about equally active, suggesting that the principle active center is probably the α,β-unsaturated ketone, in which the methylene grouping is exocyclic, since this can act in the same way as the α-methylene- γ-lactone. This hypothesis shows that the necessity is not for the unsaturated lactone but for the O=C-C=CH_2 system whether it be in the lactone or ketone. Those findings suggest that other structural parameters must be taken into consideration when evaluating the cytotoxic potential of sesquiterpene lactones.

Mode of action of sesquiterpene lactones on growth inhibitory can be concluded that cancer cells are inhibited on DNA, RNA, protein synthesis and thiol-bearing

enzyme by Michael type addition (182,184,189-191,233,235,) (see Appendix IIX) such as the following evidences. Alatolide induces defective changes in HeLa human cervical cancer cells resulting of protien synthesis and RNA synthesis (192). Vernolepin, acting as a glutathione (GSH)-depleting agent, markedly sensitized tumor cells lysis by H_2O_2 (193). The major effect of eupaformasanin as an antineoplastic agent on (mouse) Ehrlich ascites cell metabolism was to inhibit DNA synthesis, specially at DNA polymerase and thymidylate synthetase enzymatic sites. Both pyrimidine and purine systems of Ehrlich ascites were marginally inhibited. RNA synthesis and messenger and ribosomal polymerase activities were suppressed. Cyclic AMP levels were increased significantly, which correlated with the drastic reduction of histone phosphorylation. Epaformasanin suppressed a member of glycolytic and Krebs cycle enzymes an oxidative phosphorylation *in vitro*. All of the inhibited enzymes are known thiol-enzymes that can undergo a Michael-type addition with the α-methylene-γ-lactone moiety of eupaformasanin, as shown with other sesquiterpene lactones.

The plant species *Geigeria*, commonly known as "vomitting shrub", is responsible for vomitting disease in sheep. It has been shown that an ethanol extract of *G. aspera* containing the sesquiterpene lactones dihydrogriesenin, geigerinin and ivalin produced typical vomitting disease symptoms in sheep these lactones irreversibly inhibit the *in*

vitro activity of three key glycolytic emzymes namely phosphofructokinase, hexokinase and glyceraldehyde-3-phosphate dehydrogenase (195). Another 2 sesquiterpene lactones, eupatoriopicrin and hydroxyisonobiline, were studied in phytohemagglutinin stimulated human lymphocytes. Activities of phosphofructokinase, glyceraldehyde-3-phosphate dehydrogenase, pyruvate kinase and lactate dehydrogenase were inhibited by both lactones (196). Vernolepin, a eudesmanolide has also been shown to inhibit phosphofructokinase, an enzyme which has many -SH groups (197).

Table II Sesquiterpene Lactones demonstrated to have antitumor and cytotoxic activity.

Compound	Plant source	Tumor-system assayed*	Reference
Germacranolides			
Alatolide	Not specify	HeLa,	192
		SA/EAC/L1210	199
Chamissonin diacetate	*Ambrosia chamissonis*	KB	175
Deoxyelephantopin	*Elephantopus carolinianus*	WI	200
Elephantopin	*Elephantopus elatus*	KB/OS/WM	175,184
Elephantin	*Elephantopus elatus*	KB/OS/WM	175,184
Epitulipinolide	*Liriodendron tulipifera*	KB	201
Epitulipinolide diepoxide	*Liriodendron tulipifera*	KB	202
Eremantholide A	*Eremanthus elaeagnus*	KB	203,204
Eremantholide B	*Eremanthus elaeagnus*	KB	203,204
Eupacumin	*Eupatorium cuneifolium*	KB	175
Eupaformosanin	*Eupatorium formosanum*	EAC-E4	194
Eupahyssopin	*Eupatorium hyssopifotium*	WA,PS	205

Table II Sesquiterpene Lactones demonstrated to have antitumor and cytotoxic activity. (continued)

Compound	Plant source	Tumor-system assayed*	Reference
Eupatolide	*Eupatorium cannabinum*	KB/HeLa/RK	206
	Eupatorium formosanum	H.EP-2/EAC-E4	207,208
Eupatoriopicrin	Not specify	SA/EAC-E4/ L1210	199
	Eupatorium cannabinum	KB/HeLa/RK /EAC-E4/	206
		FGA	187
Eupatoriopicrin acetonide	*Eupatorium cannabinum*	FGA	187
9- α -Hydroxyparthenolide	*Anvillea garcini*	KB/PS	209
Lanuginolide	*Michelia compressa*	KB	2
Liatrin	*Liatris* sp.	KB	175
Lipiferolide	*Liriodendron tulipifera*	KB	202,210
Molephantin	*Elephantopus millis*	H.EP-2	211
Molephantinin	*Elephantopus millis*	WA	212

Table II Sesquiterpene Lactones demonstrated to have antitumor and cytotoxic activity. (continued)

Compound	Plant source	Tumor-system assayed*	Reference
Parthenolide	*Ambrosia conferiffora*	WI/H.EP-2	176
	Michelia compressa	KB	2
Phantomolin	*Elephantopus millis*	H.EP-2	211
Ridentin	*Artemisia* sp.	WI/H.EP-2	176
Tagitinin F	*Tithonia tagitiflora*	PS	214
Tamaulipin-A	*Ambrosia confertifolia*	WI/H.EP-2	176
Tamaulipin-B	*Ambrosia confertifolia*	WI/H.EP-2	176
Tulipinolide	*Liriodendron tulipifera*	KB	215
Guaianolides			
Arteglasin -A	*Artemisia douglasiana*	WI/H.EP-2/ W-18Va2	176
Canin	*Artemisia cana*	WI/HL.EP-2/ W-18Va2	176
Deacetoxymatricarin	*Achillea lanulosa*	WI/H.EP-2/ W-18Va2	176

Table II Sesquiterpene Lactones demonstrated to have antitumor and cytotoxic activity. (continued)

Compound	Plant source	Tumor-system assayed*	Reference
Eupachlorin acetate	*Eupatorium* sp.	KB	175
Zaluzanin-C	*Zaluzania robinsonia*	PS	217
	Zalyzania parthenoides	PS	232
Pseudoguaianolides			
Ambrosin	*Hymenoclea salsola*	PS	218
Aromaticin	*Helenium aromaticum*	KB	175
Augustibalin	*Balduina angustifolia*	H.EP-2	219
Baileyolin	*Baileya multradiata*	not specify	241
Fastigilin C	*Baileya multiradiata*	PS	216
Helenalin	*Hymenoxys grandiflora*	KB/PS/H.EP-2	221,224
Hymenoflorin	*Hymenoxys grandiflora*	LZ/PS	222
Microlenin	*Helenium microcephalum*	WA	223,234
Mexicanin I	*Helenium mexicanum*	KB	175
Odoratin	*Baileya pauciradiata*	KB/PS	225

Table II Sesquiterpene Lactones demonstrated to have antitumor and cytotoxic activity. (continued)

Compound	Plant source	Tumor-system assayed*	Reference
Parthenin	*Parthenium hysterophorus*	BK/	191
		KLT/MNT/	226
		P815/L1210/	227
		M-1	
Paucin	*Baileya pauciradiata*	KB/PS	225
Plenolin	*Baileya pleniradiata*	H.EP-2	228
Eudesmanolides			
Encelin	*Encelia farinosa*	WI/H.EP-2/	176
		W-18Va2	
Farinosin	*Encelia farinosa*	WI/H.EP-2/	176
		W-18Va2	
Ludovicin	*Artemisia ludovicina*	WI/H.EP-2/	176
		W-18Va2	

Table II Sesquiterpene Lactones demonstrated to have antitumor and cytotoxic activity. (continued)

Compound	Plant source	Tumor-system assayed*	Reference
Reynosin	*Michelia compressa*	KB	2
Santamarine	*Michelia compressa*	KB	2
α-Santomin	*Artemisia sp.*	WI/H.EP-2/ W-18Va2	176
Vernolepin	*Vernonia hymenolepsis*	KB	175
Vulgarin	*Artemisia vulgaris*	WI/H.EP-2/ W-18Va2	176
Bakkenolides			
Bakkenolide-A	*Petasites albus* *P. fragrans* *P. hybridus*	H.EP-2	188

Table II Sesquiterpene Lactones demonstrated to have antitumor and cytotoxic activity. (continued)

Compound	Plant source	Tumor-system assayed*	Reference
Undetermined structure			
$C_{20}H_{26}O_6$	*Anthemis nobilis*	HeLa/KB	229
Hiyodorilactone E	*Eupatorium cannabinnum*	FGA	187
15-Hydroxy-3-dehydrodesoxy fruticin	*Helianthus annuus*	NS-1/EAC	230
Isohelenol	*Helenium microcephalum*	PS	231
Niveusin C	*Helianthus annuus*	NS-1/EAC	217
Ursiniolide A	Not specify	SA/EAC/L1210	199

* Code for tumor systems assayed#

BK	= Bovine Kidney cells
EA	= Ehrlich ascites Mouse
EAC	= Ehrlich ascites breast carcinoma
EAC-E4	= Ehrlich ascites carcinoma cells
FGA	= human small cell Lung carcinoma cell line
K.EP-2	= Human epidermoid carcinoma of larynx
HeLa	= Human tumor cells (Cervical cancer)
HLT	= In vitro human leukocyte test
KB	= Human epidermoid carcinoma of nasopharynx cell culture
L1210	= Mice bearing leukemia
LZ	= Leukemia L-1210 Mouse (subcutaneous)
M-1	= Rhabdomyosarcoma
MNT	= In vivo micronucleus test with Swiss mice
NS-1	= Mouse myeloma cells
OS	= Osteogenic sarcoma He 10734 Mouse
P815	= Mastocytoma
PS	= P-388 Lymphocytic leukemia Mouse
RK	= Human oral carcinoma cells
SA	= Sarcoma 180 mouse
WA	= Walker Carcinosarcoma 256. Ascites Rat
WI	= Walker Carcinosarcoma 256. Rat
WM	= Walker Carcinosarcoma 256.Intramuscular
W-18Va2	= Simian virus 40-transfered cells of human origin

code for tumor system assayed follow by reference papers except BK, HLT, MNT there no code in the papers.

2. Microbial-growth inhibitors(antibiotics).

Some sesquiterpene lactones have been shown to possess anti-bacterial, anti-fungal, anti-helmintic properties (237-240). The pseudoguaianolide, baileyolin from *Bailya multiradiata* has been reported antimicrobial activity (241). The germacranolides, mikanolide and dihydromikanolide from *Makania monagasensis* inhibit the growth in culture, of a bacterium, *Staphylococcus aureus* and also yeast, *Candida albicans* (242). Helenin was shown to exhibit activity against the human pathogenic fungi, *Trichophyton mentagrophytes, T. acriminatum* and *Epidermophyton* sp.(243). Parthenin from *Parthenium hyterphorus* was reported to inhibit sporangial germination and zoospore mobitity in *Sclerospora graminicola*; whereas it had no effect on growth and sporulation of *Aspergillus flavus* (241). Two sesquiterpene lactones which have the same formula, $C_{15}H_{20}O_3$, and have the structure of an γ-lactone with an exocyclic methylene group in addition to a ketonic group in a cyclopentanone ring, were isolated from the extract of *Varthemia candicans*. On testing the antibacterial activity, it was revealed that both are active against *Bacillus subtilis, Staphylococcus aureus, Esherichia coli, Pseudomonas aeruginosa* and *Mycobacterium phlei* (245). Antimicrobial screening tests were also done on the extracts on 11 Iraq plants (246).

The mechanism of action is used to propose by studying activities a sesquiterpene lactone from *Helianthus annuus*, 15-hydroxy-3-dehydrodesoxyfruticin. Its influence on DNA, RNA and protein synthesis can inhibit *Bacillus brevis* and the fungus *Eremothecium ashbyi* (247). However from antifungal activity screening test on forty-five sesquiterpene lactones (238) and antibacterial activity screening test on fifty-seven sesquiterpene lactones (239), their results can be concluded that the eudesmanolides being the most active but cannot be explained simply by the presence or absence of the exocyclic methylene or, in pseudoguaianolides, by presence (absence) of a β-unsubstituted cyclopentanone ring. This implies that other moieties must play an important role, presumably by enhancing or reducing the activities.

3. Anti-Inflammatory.

Certain sesquiterpene lactone producing plants, such as *Eupatorium formosanum*, have been used as anti-inflammatory herbal remedies as well as antipyretic drugs (248). Thus, it was decided to test these agents for anti-inflammatory activity. Ten sesquiterpene lactones; helenalin from *Balduina angustifolia*, tunulin and aromaticin from *Helenium amarum*, eupatoside from *Eupatorium formasanum*, deoxyelephantopin from *Elephantopus carolinianus*, eupahyssopin from *Eupatorium hyssopifolium* and eupaformosanin,

phantomolin, molephantinin from *Elephantopus mollis*, and some related compounds were studied. In the edema-induced carrageenan inflammation screen, the α-methylene-γ-lactone moiety of the sesquiterpene lactone was required for anti-inflammatory activity. The 6-hydroxy group of helenalin also was required for potency. In the tenulin series, the 2,3-epoxy derivatives were marginally active. The same structure was required for inhibition of the writhing reflex. In the chronic adjuvant arthritic screen, compounds containing the α-methylene-γ-lactone moiety, the β-unsubstituted cyclopentanone ring, and the α-epoxy cyclopentanone system afforded significant inhibition at 2.5 mg/kg/day. The sesquiterpene lactones were marginally effective against induced pleurisy. The delayed hypersensitivity was suppresed by these agents whereas immunoglobulin synthesis was slightly stimulated. No deleterious side effects were observed with these agents from the limited tests performed (248).

Mode of action was studied on the same group of sesquiterpene lactones. It appeared to be at multiple sites; for example, at 5×10^{-4} M, the sesquiterpene lactones effectively uncoupled the oxidative phosphorylation of human polymorphonuclear neutrophils and elevated the cyclic adenosine monophosphate levels of rat neutrophils and rat and mouse liver cells. Free and total lysosomal enzymatic activity was inhibited by these agents at 5×10^{-4} M in both rat

and mouse liver and rat and human neutrophils. Furthermore, the structure-activity relationships for the stabilization of lysosomal membrane for rat liver cathepsin activity followed the same structure requirement necessary for anti-inflammatory activity (249).

4. Antihyperlipidermic activity.

Pseudoguaianolides; helenalin from *Balduina angustifolia*, tunulin and aromaticin from *Helenium amarum*, and germacranolides, deoxyelephantopin from *Elephantopus carolinianus* and eupahyssapin from *Eupatorium hyssopifolium* as well as synthetic related compounds were observed to be antihyperlipidermic agents in mice. Several of these compounds at a dose of 20 mg/kg/day resulted in lowering of serum cholesterol by - 30% and of serum triglycerides by - 25%. Thiol-bearing enzymes of lipid synthesis, i.e., acetyl -CoA, citrate-lyase, acetyl-CoA synthetase, and βhydroxy - β-methylglutaryl CoA reductase, were inhibited by these agents *in vitro*, supporting the premise that these agents alkylate thiol nucleophiles by a Michael-type addition. The α-methylene γ-lactone moiety, the β-unsubstituted cyclopentanone ring, and the α-epoxycyclopentanone system of these compounds appeared to be responsible for the lowering for serum lipids (250).

5. Chemoprophylaxis by lactones in Schistosomiases.

Extracts of several Compositae contain lactones that inhibit skin penetration by cercariae of the trematode, *Schistosoma mansoni*. Analysis of the wood oils indicated that the sesquiterpene lactones, eremanthine, costunolide, and β-cyclocostunolide were the active principles. Dihydro-α-cyclocostunolide which lacks an exocyclic methylene groups on the lactone ring was found to be inactive (251). Recently, a novel germacranolide, goyazenolide, isolated from *Eremanthus goyazensis* was also shown to have schistomicidal properties (252).

6. Anti-secretory activity.

Extracts of feverfew, *Tanacetum parthenium*, inhibit secretion of granular contents from platelets and neutrophils and this may be relevant to the therapeutic value of feverfew in migraine and other conditions. Five active compounds were identified as parthenolide, 3-β-hydroxyparthenolide, secotanaparhenolide A, canin and artecanin. All of which are sesquiterpene lactones that contain an α-methylene butyrolactone unit. It is very likely that these and other sesquiterpene lactones that contain and α-methylene butyrolactone unit are responsible for the anti-secretory activity in extracts of feverfew (253).

7. Ant-repellant activity.

The leafcutter ants of the tropical Americas are considered polyphagous, but nontheless they seldom or never attack many of the plants species available to them in nature. Investigations conducted on the foraging behavior of Costa Rican colonies of *Atta cephalotes* have shown that *Eupatorium quadrangulare* is one of the tree species seldom attacked. Five sesquiterpene lactones were isolated from the leaves of *E. quadrangulare* two of which, seco-eudesmanolide and 4-desoxy-8-epi-ivangulin, showed significant ant-repellency (254).

8. Insecticide.

Eupatolide from *Helianthus argophyllus* was tested for insecticidal activity. It showed weak activity against tobacco cutworm larvae (*Spodoptera litura*, 63 % kill after 48 hrs, 2000 ppm in an artificial diet) and mosquito larvae (*Culex pipens*, 60 % kill after 24 hrs in 10 ppm solution). Eupatolide could thus be involved in the natural resistance of *H. argophyllus* to insect predation (255). Six sesquiterpene lactones, alantolactone and isoalantolactone were separated from a commercial mixture of these two compounds, "Helenin" (Sigma chemical Co.), coronopilin and parthenin from *Parthenium hysterophorus* and hymenolin and bipinnatin from *Hymenoclea salsola*, exhibited chronic toxicity effects to the mosquito *Aedes artropalpus* (256).

9. Insect feeding deterrents.

Laboratory experimentals evidence that sesquiterpene lactone provide resistance to insect feeding has been shown by a study of 6 species of *Vernonia*. Larval feeding experiments were conducted on *Spodoptera eridania*, *S. frigiperda*, *S. ornithogalli*; *Diacrisia virginia*, and *Trichoplusia ni*. It resulted in greatly reduced larval feeding; feeding was inversely proportional to the concentration of glaucolide-A, the sesquiterpene lactone in the tested plants , in the medium (257). The later field experiment, 2 years observation, resulted contrary to the pattern observed in the laboratory feeding preference tests, *Vernonia flaccidifolia*, the species lacking sesquiterpene lactones, was consistently fed upon less by insects than were *V. gigantea* and *V. glauca*. Even though glaucolide-A appears to adequately protect some Vernonias against herbivores especially mammals, *V. flaccidifolia* has lost this compound. Apparently *V. flaccidifolia* has evolved an alternative defensive mechanism which is more effective against insects but less effective against mammalian herbivores (258).

The other antifeeding experiments of sesquiterpene latones are the following. Tulirinol from *Liriodendron tulipifera* resulted positive antifeedant test on gypsy moth, *Lymantria dispar* (259). The crude syrup of *Melampodium americanum* L. and *M. leucanthum* Torr. and Gray and their principal sesquiterpene lactones, melampodin A and melampodinin

A, significantly inhibit growth and deter feeding of the fall armyworm, *S. frugiperda*. Melampodin A and melampodinin A effected 2.3x and 5.5x greater ,respectively, than glaucolide A (260). The extracts of *Parthenium schottii* and *P. tomentosum* foliage appear to inhibit larval growth of both *Spodoptera exigua* and *Heliothis zea*. But confertin, the principal sesquiterpene lactone of *P. schottii* foliage and inflorescences, had no effect on feeding or growth of 3rd-instar larvae of *S. exigua*, even at a concentration which strongly inhibits growth of neonate *H. zea* larvae (261).

The mechanism of action was proposed by the experiment of helenin as the tundra redback vole, *Clethrionomys rutilus*. It was concluded that helanin's antifeedant property and subsequent starvation of animals as well as its interferenee with digestive process (262).

The structure activity relationship was interesting also. Nine sesquiterpene lactones were investigated and showed deterrent activity differing according to their structure (263). Morever, four sesquiterpene lactones isolated from *Eupatorium cannabinum*, *Homogyne alpina* and *Petasites albus* (all Compositae), together with 2 of their adducts, as well as 1 lignan lactone of dibenzylbutanolide type from *Libocedrus yateensis* (Cupressaceae), were tested for their feeding deterent activity against the adults of *Sitophilus granarius* and *Tribolium confusum*, as well as against the larvae of *T. confusum* and *Trogoderma granarium*.

The strongest deterrent activity against all species was exhibited by the lignan lactone yatein and the sesquiterpene spirolactone bakkenolide A. Both may be included to the class of very good insect feeding deterrents. The comparison presented here showed that the conjugated α-exomethylene moiety of the lactone ring was not the decisive factor for the deterrent activity in the investigated test model (264).

10. Mulluscicidal activity.

Snails of the genus *Biomphalaria* are the hosts in the life cycle of the blood fluke (genus *Schistosoma*, Schistosomatidae), which is responsible for human schistosomias is (bilharzia), a disease affecting more than 200 million people in many tropical countries (265). There are many sesquiterpene lactones which show molluscidal activity. A sesquiterpene lactone from *Podachaenium eminens*, 7α-hydroxy-3-desoxyzaluzanin C, killed *Biomphalaria glabrata* snails at the 1.0 ppm level within 24 hours(266). On this same snail, damsin and ambrosin from *Ambrosia maritima*, and confertiflorin and allodesacetylconfertiflorin from *Ambrosia confertiflora* show positive activity also (267). Eveven sesquiterpene lactones were examined as potential molluscicides against the planorbid snail *Biomphalaria havanensis*. The most potent compound was helenalin followed by pyrethrosin (268).

11. Allergic contact dermatitis.

Many plants from species of the Compositae, Lauraceae, Magnoliaceae, Umbelliferae and from the liverwort *Frullania*, all of them contain sesquiterpene lactones, have been shown to be a major class of allergens causing allergic contact dermatitis in humans (269-276).

High incidence of these cases are caused by the sesqiterpene lactones; alantolactone, parthenin, arbusculin A, arbusculin C, ambrosin, damsin, 8-deoxycumambrin, rothin A, psilostachynin(273,277,278). Certain persons may be cross-reacted by skin contact allergy to such diverse ptants and plant products as rayweed (Ambrosia), liverwort (*Frullania*), horticultural plants (*Chrysanthemum*), perfumes (*Saussurea*), weeds (*Parthenium* etc.) and vegetables (*Cichorium* etc.) (279-282). The interesting investigation is shown the structures relate to cross-sentivity. They testd 38 sesquiterpene lactones of five different classes with 13 costus sensitive patients by patch testing over a two-year period. Cross-reacting agents fell into two chemical catagories; A. those that resembled the primary sensitizer, and B. those belong to different skeletal classes. An exocyclic methylene groups conjugated to γ-lactone was present in both chemicals that cross-reacted and those that did not. The difference between these two groups is that cross-reacting chemicals are not highly substituted, tending to be lipophlic,

while those giving negative responses all are highly substituted at the c-8/c-6 position. This functional group may hinder binding of exocyclic methylene with skin protein or the actual antigenic site with an immune receptor cell(280).

The known allergenic sesquiterpene lactones contain an exocylic α-methyl function which may conjugate with sulphydryl groups of proteins in cell by a Michael-type addition to form complete antigens capable of producing cell-mediated contact allergic reactions (283-285). But the recent investigation showed that 4 sesquiterpene lactones lacking the exocyclic α-methylene at the ring, brevelin A, arnicolide D, tenulin, 16 α -(1-methyl-1,2-epoxypropyl)-erere-mantholanolide, but possessing further unsaturated centers such as a cyclopentenone ring or an epoxy group, were proved to be sensitizers in guinea pigs. Lack of substitution in the cyclopentenone ring and unsaturation in the side chains appear to be necessary perequisites for nucleophilic attack (286). So the further investigations are necessary for making the clear conclusion.

12. Vertebrate poisoning.

Livestock-poisoning from foraging a bitter tasting plants of Compositae in well documented in agricultural literature (287,288). For example, *Hymenoxys odorata* (bitterweed) is an important livestock toxicant that affects

primarily sheep and goats (287). Chemical studies on *H. odorata* have shown that hymenovin is the toxin involved in the death of sheep (287). Similar poisoning (vomiting disease in sheep) has been noted among sheep grazing on South African species of *Geigeria* which contain the compound vermeerin (289). It was suggested that the sesquiterpene lactone toxicant may alter the microbial composition of the rumen and thus affect vital metabolic function (287).

The structure poisonous activity relationship was studied on seven sesquiterpene lactones from three plants which two of them are poisonous to livestock (287, 288). The sesquiterpene lactones were administered to rats by intra parenteral route, LD_{50} were determined (in mg/kg); mexicanin E [from *Helenium microcepharum*](3.08$\pm$0.10), helanalin [also from H. microcepharum] (9.86$\pm$0.08), hymenoxon [from *Hymenoxys odorata*](16.24$\pm$0.10), psilotropin (112.25$\pm$0.17), hymenoxon dimethyl ether(141.42$\pm$0), tenulin [from *Helenium amarum*] (184.65$\pm$0.06), and hymenolane (also from *H. odorata*] (above 200). The toxicity of a sesquiterpene lactone depends on the mumber of alkylating centers such as cyclopentanone, α-methylene-γ-lactone, or hemiacetal moieties in the molecule (290).

13. Plant-growth inhibitors (phytotoxin).

A variety of sesquiterpene lactones of different skeletal types has been reported to show plant growth regulatory activity, for example, the sesquiterpene heliangine isolated from *Helianthus tuberosa* L., Heliangine inhibits the elongation of *Avena* coleoptile sections and promotes adventitious root formation of *Phaseolus* cuttings. Promotion is almost completely reduced by suppling cysteine which reacts *via* the SH-groups with the exomethylene group of heliangine suggesting that the exomethylene group conjugated to the lactonic carbonyl may therefore be responsible for the growth inhibiting activity (291).

Parthenin, a sesquiterpene lactone, isolated from *Parthenium hysterophorus* Linn. was tested for its root inhibitory property. It checked germination as well as seedling growth of *Crotalaria mucronata* Linn., *Cassia tora* Linn., *Ocimum basilicum* Linn., *O. americanum* Linn. and barley cv. `Ratna' (292).

The sesquiterpene lactones, arbusculin-A, achillin, deacetoxymatricarin, viscidulin B, and viscidulin C from sagebrush inhibited the growth and stimulated the respiration of *Cucumis sativus* seedlings (253).

Argophyllin-A and -B, germacranolide sesquiterpene lactones from *Helianthus argophyllus*, are closely related to heliangine from *H. tuberosus*. So the inhibitory ef-

fect of argophyllin -A and -B on the IAA-induced elongation of Azuki (*Azukia angularis*) hypocotyl sections was examined. They were found to show anti-auxin effects (294).

The characteristic pungency of the European liverwort, *Chiloscyphus polyanthus*, is due to a mixture of four sesquiterpenes, *ent* -5 β -hydroxydiplophyllin, *ent*-3-oxodiplophyllin, diplopyhllin and diplophyllolide. The last two sesquiterpene lactones have been also isolated from *Diplophyllus albicans*. They, four sesquiterpene lactones, showed inhibitory activity towards the germination and root elongation of rice husks (295).

Inunal, a sesquiterpene lactone from *Inula racemosa*, displays considerable biological activity as a plant growth regulator (296). The structure of inunal correlates with alantolactone. The alantolactone was shown to be a potent inhibitor of seed germination, seedling growth rate of respiration, and degradation of starch and proteins in mung bean (*Phaseolus mungo*) (297).

14. Mutagenic activity.

Hymenovin, the major toxic constituent of the range plant *Hymenoxys odorata*, which has caused of poisoning in grazing animals, was mutagenic in the Ames *Samonella typhimurium* test for mutagens and potential carcinogens. An *in vitro* rat-liver metabolizing system did not alter the mutagenic activity significantly (298).