

CHAPTER II



HISTORICAL

1. Distribution of the Anthraquinone Compounds in Natural Sources

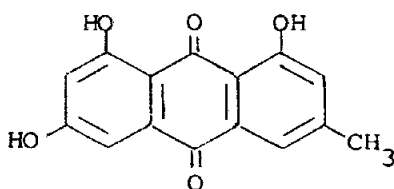
Anthraquinone compounds are the largest group of natural quinones. They are widely distributed in the plant kingdom and some animals such as in insects (Coccidae only) and in feather star (*Crinoidea*) (Shibata, 1967). They are frequently found in lichens such as *Caloplaca* which are remarkable by the yellow to red colour because of its anthraquinone pigments, parietinic acid and emodin (Bohman, 1969). It was reported that the first appearance of anthraquinones was in bacteria (Thomson, 1971). In moulds, anthraquinones are mostly found in *Aspergillus* and *Penicillium*. More than twenty anthraquinones and related compounds have been isolated from *Penicillium isalindicum* Sopp. (Shibata, 1967). The distribution of anthraquinones in higher plants are mostly in the Rubiaceae, they account for half the total number. They are located chiefly in heartwood, bark and roots, sometimes they are in stem, seeds and fruits (Thomson, 1971).

Anthraquinones can be found among the following genera of the Rubiaceae :- *Morinda*, *Galium*, *Coprosma*, *Damnacanthus*, *Hymenodictyon*, *Hedyotis* (Gibbs, 1974), *Prismatomeris* (Lee, 1969) and *Coelospermum* (Thomson, 1971). There was also a report of the anthraquinones in callus cultures of *Cinchona ledgeriana* Moens (Rubiaceae) (Wijnsma *et al.*, 1984). The bark of *Coprosma australis* Robinson contains 17% of its dry weight of anthraquinones. The root of *Rubia tinctorum* Linn. (madder)

has been known to contain about twenty anthraquinones (Thomson, 1971). The other families also containing anthraquinone pigments are Rhamnaceae [*Rhamnus* (Gibbs, 1974), *Maesopsis* (Cumming, 1970), *Ventilago* (Cooke and Johnson, 1963)], Polygonaceae [particularly *Rumex*, *Rheum* and *Polygonum* (Gibbs, 1974)], Leguminosae, subfamily Caesalpinaceae [*Cassia* (Takido, 1958)], subfamily Papilionaceae [*Abrus* (Gibbs, 1974)], Bignoniaceae [*Tabebuia avellanedae* Lor. ex Griseb. (Burnett and Thomson, 1967)], Verbenaceae [*Tectona grandis* Linn. (Ahluwalia and Seshadri, 1957)] and Scrophulariaceae [*Digitalis* species (Gibbs, 1974)]. Anthraquinones are also found among monocotyledons especially in the Liliaceae and Xyridaceae. *Aloe* (Rheede, 1963), *Asphodeline*, *Asphodelus*, *Bulbine*, *Enemerus* (Rheede, 1964) and *Polygonatum* (Gibbs, 1974) represented the genera of the Liliaceae which contained anthraquinones, as well as *Xyris indica* Linn. (Ruangrungsri and Tantivatana, 1980) and *X. semifuscata* Baker (Fournier *et al.*, 1975) of the Xyridaceae. Some anthraquinones are found in Anacardiaceae, Apocynaceae, Asclepiadaceae, Caryophyllaceae, Compositae, Ericaceae, Euphorbiaceae, Lythraceae, Rhizophoraceae, Saxifragaceae (Gibbs, 1974) and Solanaceae (Knapp, 1972).

The distribution of anthraquinone may be overlapping with the distribution of naphthaquinone but this is only of significance in the Bignoniaceae and Verbenaceae where related naphthaquinone and anthraquinones co-exist in the same species.

Emodin (1) is probably, the most widely distributed anthraquinone, being found in moulds, higher fungi, lichens, flowering plants (at least six families) and insects. The structural formula of emodin (1) is shown below.

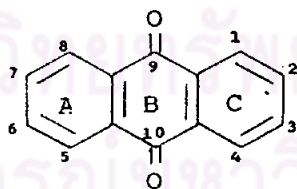


Emodin (1)

2. Chemical Nature of Anthraquinone Compounds

Anthraquinone itself has been obtained from several natural sources but was probably an artefact in all cases (Thomson, 1971). Most of the anthraquinones are red yellow or orange yellow colouring matter, varying from yellow to brown. The anthraquinone crystals are high-melting point compounds, they are soluble in organic solvents. Anthraquinone composes of three benzene rings having the quinoid doublebond. Most of the naturally occurring anthraquinones are hydroxylated at the C-1 position.

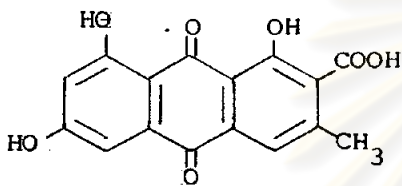
The fundamental structure (2) and the numbering system of anthraquinones is shown below.



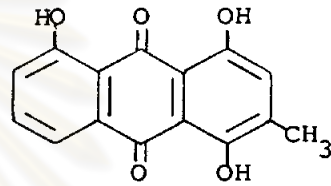
(2)

The anthraquinones can be classified into two groups according to their biosynthetic pathway. The first group consists of anthraquinones with substitution in ring A and C. They are found in fungi and higher plants. In fungi, emoidn (1), endocrocin (3) and islandicin (4) were found. In higher plants anthraquinones are distributed in

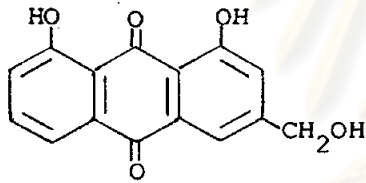
Leguminoceae (subfamily Caesalpinaceae and Papilionaceae), Polygonaceae and Rhamnaceae e.g. emodin (1), aloe-emodin (5) and chrysophanol (6). The other group consists of anthraquinone with substitution only in the ring C. They are found mainly in Bignoniaceae, Rubiaceae, Scrophulariaceae and Verbenaceae e.g. alizarin (7), lucidin (8) and rubiadin (9) (Robinson, 1967).



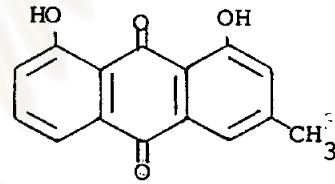
Endocrocin (3)



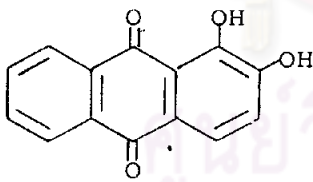
Islandicin (4)



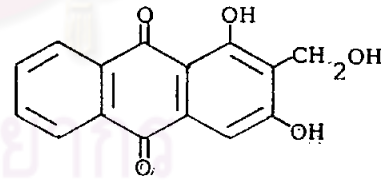
Aloe-emodin (5)



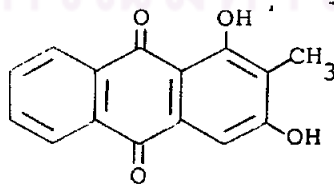
Chrysophanol (6)



Alizarin (7)

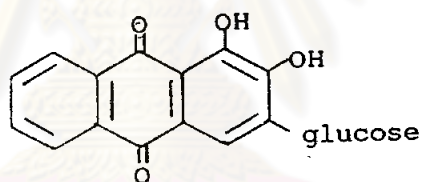


Lucidin (8)

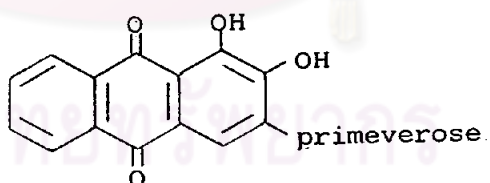


Rubiadin (9)

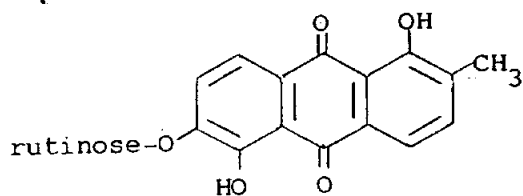
The anthraquinones actually existed in plants are apparently in several forms. They are often found as anthraquinone glycosides rather than hydroxylated anthraquinones or aglycones. Reports of the appearance of free anthraquinones must be regarded cautiously. Many anthraquinones occur as glycosides with the sugar residue linked through one of the phenolic hydroxyl groups. Several different sugars are found in such glycosides. For example, alizarin (7) occurs as a 3-glucoside (10) in *Rubia tinctorum* Linn. (madder) and as a 3-primeveroside (11) in *Galium* species; and morindone occurs as a 6-rutinoside (12) in *Coprosma australis* Robinson and as a 6-primeveroside in *Morinda persicaefolia* Ham. (Robinson, 1967).



Alizarin-3-glucoside (10)

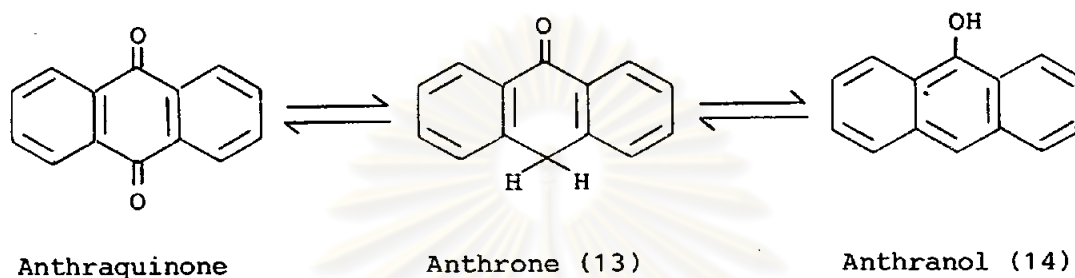


Alizarin-3-primeveroside (11)



Morindone-6-rutinoside

The aglycones of several anthraquinone glycosides may exist naturally in reduced forms, one of which is anthrone (13). The sugar in these reduced glycosides may be linked as usual through phenolic oxygens in the outside ring or they may be attached at C-9 of the enol form of anthrone, anthranol (14) (Robinson, 1967).



Enzymatic or chemical hydrolysis followed by oxidation of anthranol glycoside would give anthrone and anthraquinone. If the sugar is linked at some other positions except C-9 and C-10, anthranol glycosides may be directly oxidised to anthraquinone glycosides. Thus, the anthraquinone glycosides are divided in two types. One is O-glycoside, the sugar links at the phenolic oxygen. The other is C-glycoside with sugar attached through carbon-carbon bond (C-1 to C-8).

A group of glycosides is known as the sennosides. They are the glycosides of sennidin dianthrone. Sennosides A and B are the 8,8'-diglucosides of rhein-rhein dianthrone, and sennosides C and D are the corresponding diglucosides of aloë-emodin-rhein dianthrone. The dianthrone carbon-carbon bond is liable to oxidative cleavage far more easily than the normal carbon-carbon bond. This is due to the stability of the resulting anthrone radical. Oxidation is carried out by using a free radical oxidising agent such as ferric chloride. (Fig. 2, p. 14)

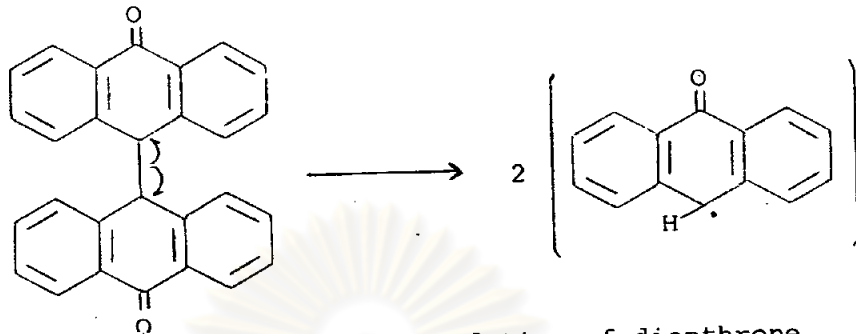
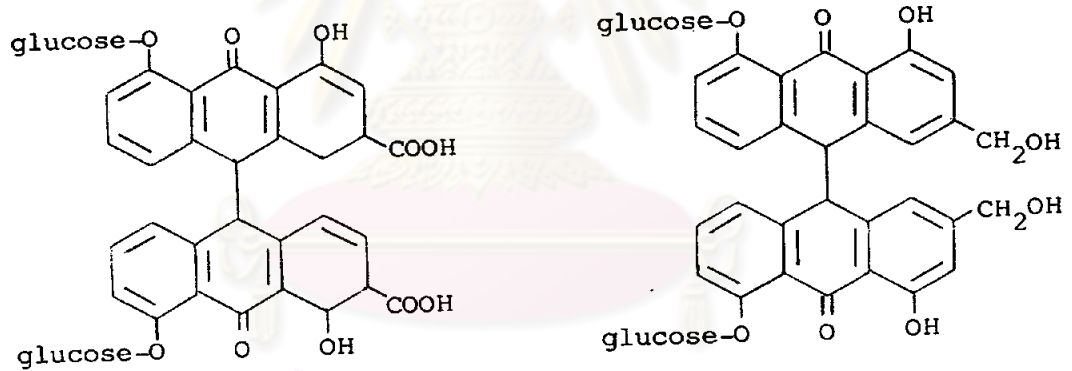


Fig. 2 Free-radical oxidation of dianthrone

The difference between the aglycone of sennosides A and B (15) is one of optical activity and the same relationship can be applied for the aglycones of sennoside C and D (16).



Sennosides A and B
(15)

Sennosides C and D
(16)

Antraquinones can be detected by the Bornträger test in which an organic solution containing the test material is shaken with an aqueous base. In bases, the anthraquinone can form phenolate-type ions which are coloured. The visible result of the test in which the basic layer goes a cherry red and the intensity of the colour can be used as a measure of the amount of anthraquinone in the original material. Only free anthraquinone give a positive Bornträger test and this fact can be used to distinguish between the O- and C-glycosides, the O-glycosides being hydrolyzed to free anthraquinones by reflux

with diluted hydrochloric acid while the C-glycosides release the free anthraquinones only after oxidative cleavage (Ross and Brain, 1977). The colours given with alcoholic magnesium acetate solution are characteristic of different hydroxylation patterns (Robinson, 1967). Compounds containing, two hydroxyl groups in the ortho position e.g. alizarin, exhibit a violet colour, those with two in the meta position e.g. emodin, give an orange-red or pink colour, and those with two in para position e.g. quinizarin, produce a purple. These colour reactions are specific and stable.

The most important group of laxative drugs used today is the group of the plant products derived from materials that contain anthracene derivatives. The active constituents are anthraquinone compounds containing phenolic groups, either free, as methyl ethers, or as glycosides. The free anthraquinones or glycosides are ineffective, and the pharmacologically important compounds are free anthranols. Anthraquinones are active as cathartics only because they are reduced to anthranols by intestinal bacteria (Robinson, 1967).

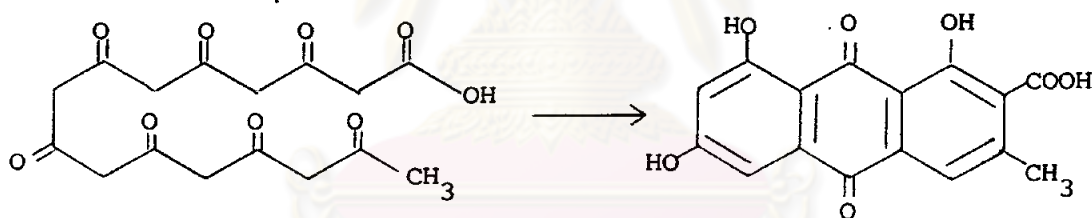
3. Biogenesis of Anthraquinone Compounds

Anthraquinones are derived from a few key intermediate, principally acetate, shikimate and mevalonate by a series of reactions which lead to the formation of benzenoid compounds, they arise by at least two biosynthetic routes. Fungal anthraquinones such as emodin (1) and chrysophanol (6) have structures in accord with the acetate hypothesis, while the anthraquinones found in higher plants, for example alizarin (7), have biosynthetic route *via* shikimate-mevalonate pathway.

The two possible biogenetic pathways of the anthraquinone compounds are as follows :

3.1 Acetate-Malonate Pathway (Thomson, 1971)

The majority of the anthraquinones which are assumed to be elaborated by the acetate-malonate pathway is confined to emodin pattern. They are arised by suitable folding and condensation of a polyketide chain derived from eight acetate units. (Fig. 3, p. 17) Numerous variations of this basic structure exist, resulting from O-methylation, side-chain oxidation, chlorination, dimerisation and the introduction or omission of nuclear hydroxyl groups, while in endocrocin (3) the terminal carboxyl group is retained.



Poly- β -ketomethylene compound

Endocrocin (3)

Emodin can be found in higher plants, it lacks only the carboxyl residue of endocrocin, a component of the lichen *Nephromopsis endocrocea* Asahina (Asahina and Fujikawa, 1935), but bears the same pattern of oxidation.

Bacterial and fungal anthraquinones have their formation *via* the acetate-malonate pathway. As such typical fungal anthraquinones as emodin and chrysophanol are also found in higher plants. Leistner and Zenk (Leistner and Zenk, 1969) have proved that chrysophanol is produced in *Rumex alpinus* Linn. and *Rhamnus frangula* Linn. *via* the

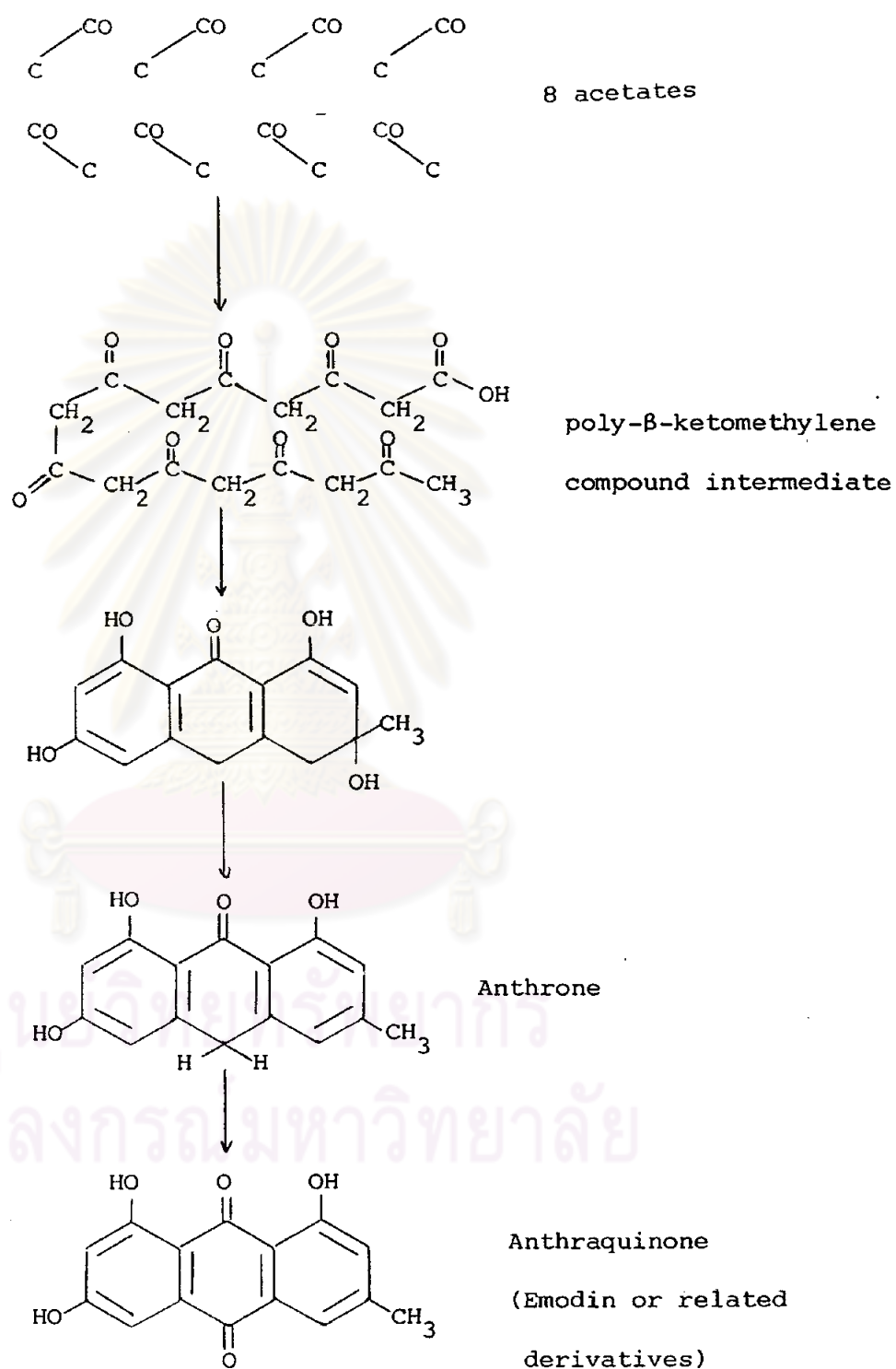
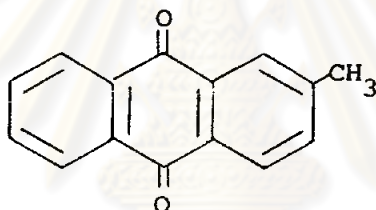


Fig. 3 Acetate-malonate pathway of anthraquinones

acetate-malonate route.

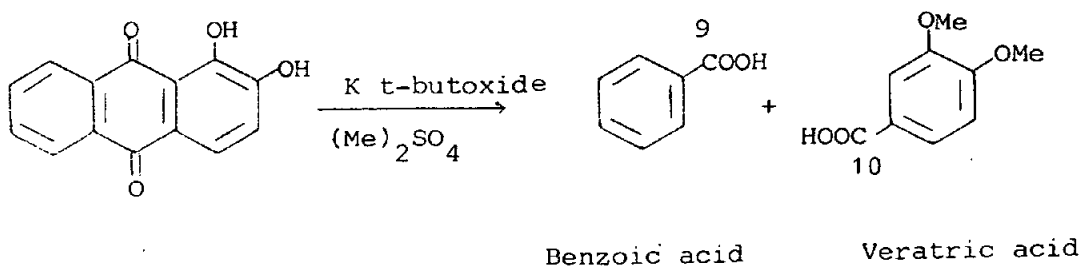
3.2 Shikimate-Mevalonate Pathway

Anthraquinones which are found in some higher plants especially in the order Tubiflorales have shikimate-mevalonate biosynthetic route. They are substituted in only one benzenoid ring (ring C) and may be totally devoid of a carbon side chain or hydroxyl groups in ring A e.g. alizarin (7) and tectoquinone (17). The majority of these anthraquinones occur in the Rubiaceae subfamily Rubioideae and, to a lesser extent, in the Bignoniaceae and Verbenaceae (Thomson, 1971).



Tectoquinone (17)

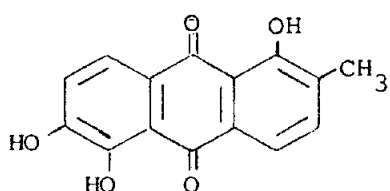
Labelled precursors, carboxyl -¹⁴C-D-shikimic acid, in *Rubia tinctorum* Linn. roots, led to labelled alizarin. The distribution of radioactivity in the alizarin molecule was determined by degradation of the alizarin dimethylether which yielded benzoic acid (ring A plus C-atom 9) and veratric acid (ring C plus C-atom 10). The result of this degradation showed that the carboxyl group of shikimic acid is exclusively incorporated into C-atom 9 of alizarin.



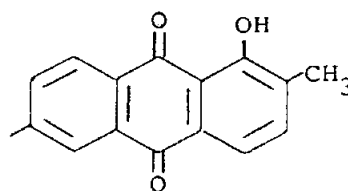
After ^{14}C -2-glutamic acid feeding, it seemed that C-2 of glutamic acid give rise specifically to C-10 of naphthalene or alizarin anthraquinone. This naphthalene could be 1,4-dihydroxy-2-naphthoic acid which is linked to γ,γ -dimethylallyl pyrophosphate derived in turn from mevalonic acid, in the meta position to C-9 of alizarin. The latter observation emerges from the fact that activity from C-5 mevalonic acid is specially incorporated into C-4 of alizarin so suggesting that C-1 to C-4 are derived from mevalonic acid by way of γ,γ -dimethylallyl pyrophosphate. Decarboxylation and ring C closure would lead to anthraquinone such as alizarin (Leistner, 1973).

Leistner (Leistner, 1973) has shown the biosynthesis of alizarin in *Rubia tinctorum* Linn. by using tracer technique. Specific incorporation of labels from carboxyl ^{-14}C -D-shikimic acid, 2 ^{-14}C -D-glutamic acid and 5 ^{-14}C -DL-mevalonic acid suggests that these compounds provide the skeleton of alizarin. Experimental data indicate that β -keto-glutaric acid or derivative combines with shikimic acid, chorismic acid, or prephenic acid to give o-succinylbenzoic acid which is then transformed to a nonsymmetrical 1,4-naphthaquinone intermediate, and γ,γ -dimethylallyl pyrophosphate is then attached. Ring closure and further modification lead to alizarin.

Morindone (18) and soranjidiol (19) anthraquinones of *Morinda citrifolia* Linn. are hydroxylated in both ring A and ring C.

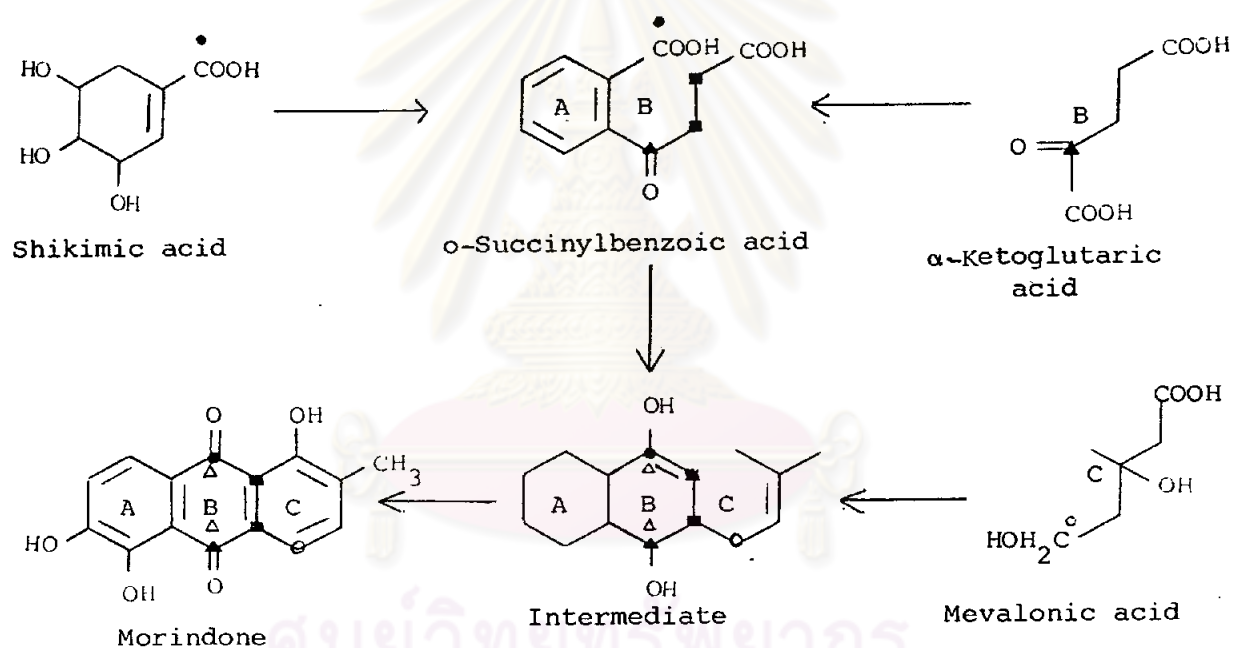


Morindone (18)



Soranjidiol (19)

Experiments carried out by Leistner (Leistner, 1973) showed that anthraquinones in morindone are derived from shikimic acid *via* *o*-succinylbenzoic acid as the same biosynthetic pathway as alizarin. The hydroxyl groups attached to ring A are introduced at the latter stage of biosynthesis and are not derived from hydroxyl groups of shikimic acid.



- ▲ ^{14}C from α -Ketoglutaric acid ○ ^{14}C from Mevalonic acid
- ^{14}C from *o*-Succinylbenzoic acid △ ^{14}C from 2-(γ,γ -Dimethylallyl)-
- ^{14}C from Shikimic acid naphthaquinone

Fig. 4 Migration of radioactivity from different precursors to morindone (Leistner, 1973)

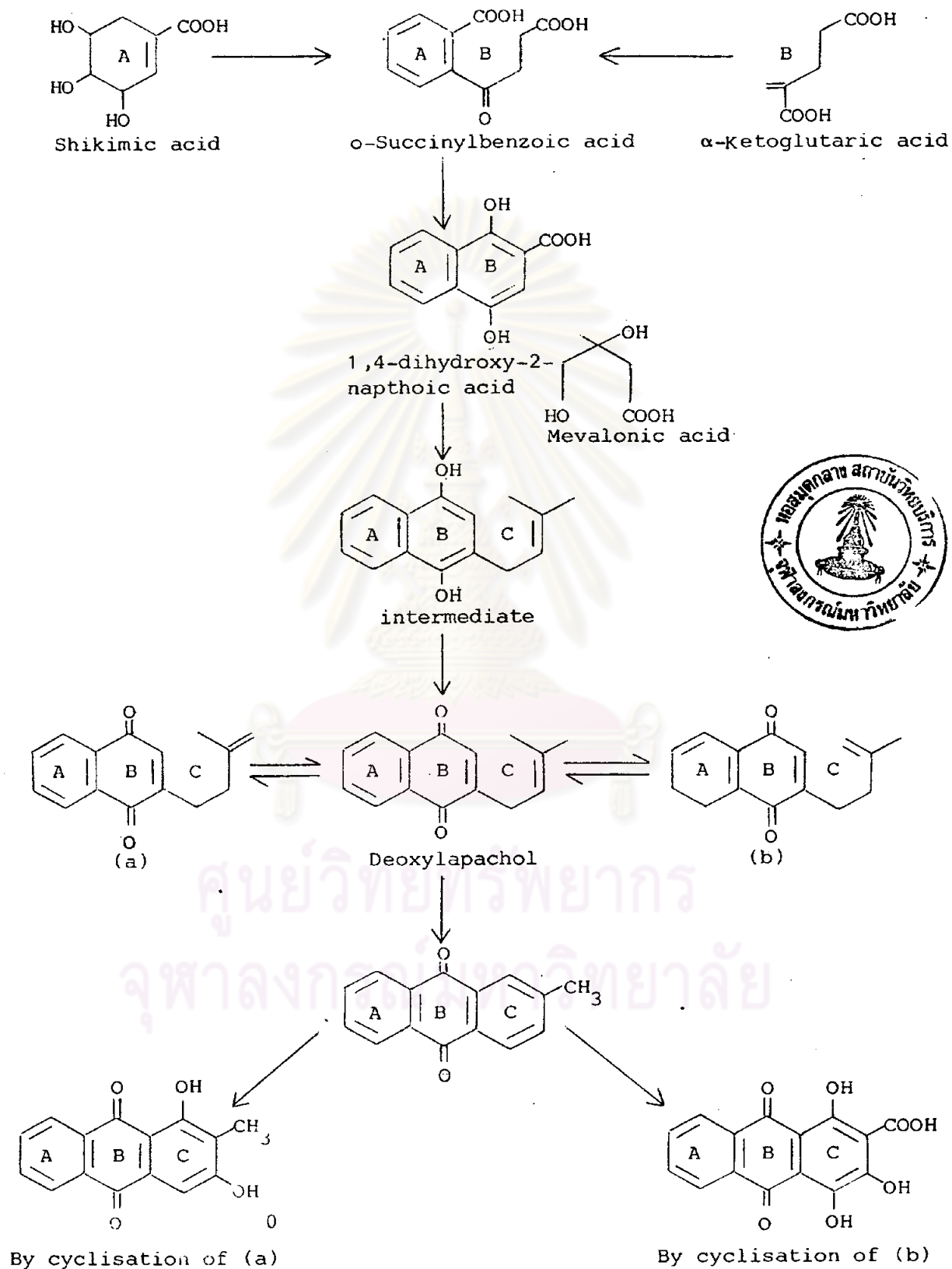


Fig. 5 Shikimate-Mevalonate pathway of anthraquinones

4. Occurrence of the Anthraquinone Compounds in *Morinda* species*Morinda angustifolia* Roxb.

Plant part	Anthraquinone	Reference
	Rhein Aloe-emodin Morindone Morindone	Rao <i>et al.</i> , 1978

Morinda citrifolia Linn.

Plant part	Anthraquinone	Reference
Cell suspension	Alizarin Morindone	Leistner, 1973
	Alizarin Lucidin Lucidin ω -ethyl ether Morindone Nordamnacanthal Rubiadin	Leistner, 1975
	2-Methyl-3,5,6-trihydroxy anthraquinone 3-Hydroxymorindone 5,6-Dihydroxylucidin Lucidin-3- β -primeveroside Morindone-6- β -primeveroside	Inoue <i>et al.</i> , 1981

Morinda citrifolia Linn. (Continued)

Plant part	Anthraquinone	Reference
Cell suspension	3-Hydroxymorindone-6- β - primeveroside 5,6-Dihydroxylucidin-6- β - primeveroside 2-Methyl-3,5,6-trihydroxy anthraquinone-6- β -prime- veroside	Inoue <i>et al.</i> , 1981
Flowers	5,7-acacetin-7-O- β -D(+)- glucopyranoside 5,7-Dimethyl-apigenin-4-O- β - D(+)-galactopyranoside	Tiwari, 1976
Heartwood	Alizarin Anthragallol 2,3-dimethyl ether Damnacanthal Morindone Nordamnacanthal Rubiadin 1-methyl ether	Balakrishma <i>et al.</i> , 1961
Root bark	Morindone Morindonin	Balakrishma <i>et al.</i> , 1960

Morinda lucida Benth. (Continued)

Plant part	Anthraquinone	Reference
Stem	Lucidin-3-O- β -primeveroside Munjistin methyl ester Soranjidiol-6-O- β -prime- veroside	Demagos <i>et al.</i> , 1981

Morinda persicaefolia Buch.-Ham.

Plant part	Anthraquinone	Reference
Stem	Morindin Morindone	Paris and Nguyen Ba Tuoc, 1954

Morinda parvifolia Bartl.

Plant part	Anthraquinone	Reference
Root	Alizarin 1-methyl ether Morindaparvin A	Chang <i>et al.</i> , 1982

Morinda tinctoria Roxb.

Plant part	Anthraquinone	Reference
Heartwood	Damnacanthal Morindone	Eswaran <i>et al.</i> , 1979

Morinda tinctoria Roxb. (Continued)

Plant part	Anthraquinone	Reference
Heartwood	Nordamnacanthal Tinctomorone	Eswaran <i>et al.</i> , 1979
Root	Damnacanthal Morindone Nordamnacanthal	Mishra and Gupta, 1982
Root bark	Morindone Morindonin	Murti <i>et al.</i> , 1959

Morinda tomentosa Heyne ex Roth.

Plant part	Anthraquinone	Reference
Root bark	Anthragallol-2,3-di-methyl ether Alizarin 1-methyl ether Ibericin Rubiadin Soranjidiol	Rao and Rao, 1983
Stem bark	Morindone Morindone-6-primeveroside	Rao and Veera Reddy, 1976

Morinda umbellata Linn.

Plant part	Anthraquinone	Reference
Root	Alizarin Alizarin 1-methyl ether Alizarin 2-methyl ether 1-Hydroxy-2-methyl anthraquinone 2-Hydroxyanthraquinone Lucidin 2-Methylanthraquinone 2-Methoxyanthraquinone 1-Methoxy-2-methyl anthraquinone Munjistin Rubiadin Rubiadin 1-methyl ether Glycoside of rubiadin 1-methyl ether Xanthopurpurin	Burnett and Thomson, 1968
Stem	Alizarin Alizarin 1-methyl ether Alizarin 2-methyl ether 1-Hydroxy-2-methyl anthraquinone 2-Hydroxyanthraquinone 1-Methoxy-2-methyl anthraquinone	

Morinda umbellata Linn. (Continued)

Plant part	Anthraquinone	Reference
Stem	2-Methoxyanthraquinone Rubiadin Rubiadin 1-methyl ether Xanthopurpurin	Burnett and Thomson, 1968
Stem	Tectoquinone	Hui and Yee, 1967



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