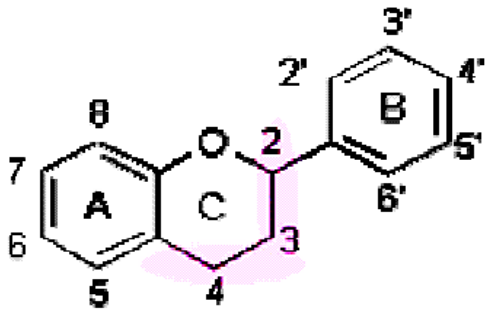
1-Introduction

1.1 Diffention

The term flavonoid (from the Latin word*flavus*meaningyellow. Is generally used to describe a broad collection of natural products possessing 15 carbon atoms, having C_6-C_3-C_6 carbon framework, comprising two benzene rings (A and B) linked through a heterocyclic pyrane ring (C);

- i.e. a chromane moiety bearing a second aromatic ring.(A)

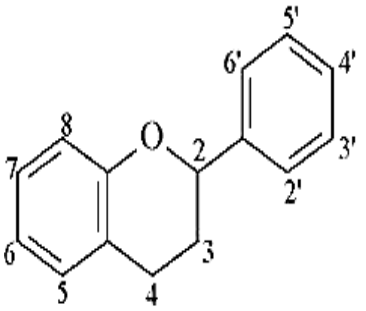


-(A)-

1-2-Classification:

Depending on the position of the linkage of the aromatic ring to the benzopyrano (chromano) moiety, this group of natural products may be divided into three classes:

- Flavonoids (2- phenyl benzopyrans) 1
- Isoflavonoids (3- benzopyrans) **2**
- Neoflavonoids (4- benzpyrans) **3**



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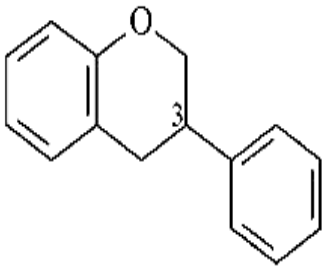

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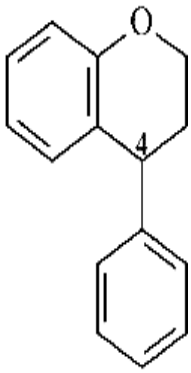

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In addition, flavonoids are important factors in biological interactions between living organisms. This is best illustrated by the last review ‘‘Advances in flavonoid research since 1992’’ focusing on these topics.[1] In contrast, mere distribution studies or chemosystematically oriented compilations are rare (e.g., on Asteraceae).[2] Naturally, the presentlyknown distribution of flavones and flavonols in plants reflects the current scientific interests, and hence the interpretation of their chemodiversity must be made with caution.

The main part of this compilation consists of extensive tables listing the flavonoids and their plant sources, which are commented accordingly.

The data originate primarily from excerpts of current literature, the use of *ChemicalAbstracts* and of *CurrentContents* (LifeSciences and

Agriculture) databases, supported by a review on prenylated flavonoids[3]anddata taken from the *Handbook of NaturalFlavonoids*.[4]

Whenever possible, original literaturewas consulted to verify structures and their sources.

For compilation and arrangement of compounds, earlier reviews and surveys were taken as the basis[5,6].

in comparison to the previously published reviews, the increasing number and complexity of structures observed is striking. Thus it became quite difficult to list all of these structures in a logical sequence, particularly prenylated derivatives with additional cyclized substituents. Substitution patterns used for grouping of the flavone and flavonol derivative are as follows.

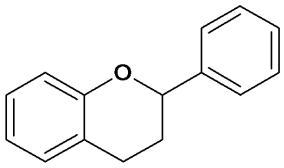
OH-, OMe-groups; *C*-methyl; methylenedioxy groups; *C*-prenylation; *O*-prenylation; (dihydro)furanosubstitution; pyranosubstitution; complex cyclosubstitution; aromatic substitution; Esterification, chlorination. These residues may also occur combined in one flavonoid structur.

In many cases, abbreviation of substitutents could no longer be made without ending up with hardly understandable chemical nomenclature (e.g., complex-*O*-cyclosubstitution).

Thisproblem was already obvious in the publication of Barron and Ibrahim[3] who shifted toillustrations of such complex compounds. Consequently, figures of structures showing characteristicsubstitution patterns will complete the tabulated information provided here.

Based on the degree of oxidation and saturation present in the heterocyclic C-ring, the flavonoids may be divided into different categories:

- Flavan**4**
- Flavanone**5**
- Flavone **6**
- Flavonol**7**
- Dihydroflavol**8**
- Flavan-3-ol **9**
- Flavan-4-ol **10**
- Flavan-3,4-diol **11**



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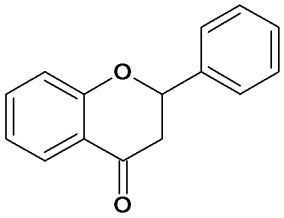

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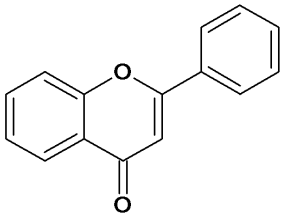

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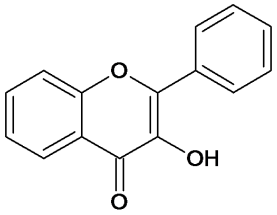

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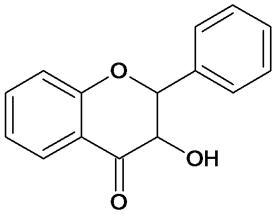

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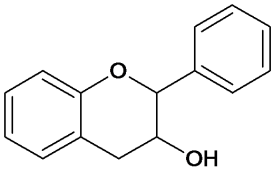

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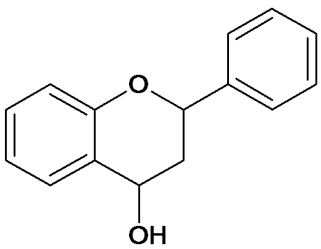

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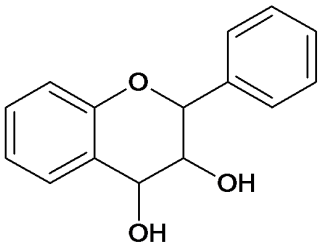

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1.2.1 **Flavones**

In flavones, the C ring bears:

- C = O bond at the 4 position double bond between the 2and 3 positions.
- Flavones mainly occur as 7-*O* glycosides in plants.
- Flavones usually hydroxylated at (C-5 and C-7).
- The most frequently bound sugar is glucose.

Common members of the flavones are:

- apigenin
- luteolin

Apigenin is the aglycone of apiin (apigenin-7-glucoapioside) which is abunant in parsley and celery.

The substitution patterns range from unsubstituted flavone to octa-O-substituted flavones. As expectedThe number of plant species accumulating these structures is growing. Since the last complilation,[6] some eight compounds (scut-6-Me, scut-6,7-diMe, scut- 6,4'-diMe, scut-6,7,4'-triMe; lut-3'-Me, 6-OH-lut-6-Me, 6-OH-lut-6,7-diMe, and 6-OH-lut- 6,3'-diMe) fall in the category of ''widespread'' in addition to apigenin, genkwanin (apigenin- 7-Me), acacetin (ap-4'-Me), and luteolin. Thus, no specific sources have been listed for these compounds:

- Luteolin is the aglycone of Cynaroside (luteolin-7-glucoside).
- It can be found in dandelion coffee(dandelion root)
- The number of newly described structures increased by about 50 entries during the

- reporting period. These include a series of 2'- and 5'-substituted flavones, which have been
- reported from several Asteraceae such as from leaves of *Psiadia punctulata*.[9] The genus
- *Andrographis*(Acanthaceae) yielded several of these more complex substituted flavones,
- isolated from whole plants.[10–14] In contrast to other reports on *Andrographis,*[7] the meaning
- of ''whole plant'' could not be clarified, with root tissue probably included in the analysis as well.

  Since all Andrographis species are annuals, inclusion of root tissue probably has little influence on the flavonoid composition. Capitula of *Leiothrixflavescens (Eriocaulaceae)*

yielded a new flavone with a rare 5,6,7,8,3',4' hexahydroxy substitution . This is remarkable insofar as many of the listed flavones are (poly)methoxyderivatives and other hexahydroxyflavones are known as glycosides only.

Most of the source reports concern equally the families of the Asteraceae and Lamiaceae,followed by Rutaceae. However, it must be taken into consideration that the long list may rather be due to the number of species and not to the number of genera. The large number of results in both families may also be due to the research focus on these groups by the authors.In these families, flavone accumulation is mostly reported in leaves, aerial parts and in exudates.

Some flavone structures have been revised during the reporting period. The structure of 5,8,2'-triOH-6,7-diOMe flavone had been ascribed to a product isolated from Scutellaria*baicalensis.*[23] After synthesis, it needs to be revised to 5,7,2'-triOH-6,8-diOMe flavone Pedunculin, earlier isolated from *Tithonia*species and claimed as 5,8-diOH-6,7,4'-triOMe-flavone

needs to be revised, after synthesis, to 5,7-diOH-6,8,4'-triOMe flavone¼nevadensin. In theprevious review, the compound 5,6,7,4'-tetraOH-3',5'- diOMe had erroneously been cited as a component of *Artemisia assoana*.[6] Data have now been included for the correct structure, 5,7,4'-triOH-6,3',5'-triOMe flavone . A further flavone reported from Ageratum conyzoides as 5,6,8,3',4',5'-hexamethoxyflavone,[27] was revised to 5,6,7,3',4',5'-hexaOMe flavone after synthesis.[28]

Pharmacological:

Apigenin and luteolin have been shown to possess remarkable:

- anti-inflammatory
- antioxidant
- anti-carcinogenic properties.

1.2.2**Flvonols**:

- Their structures differing from flavones only in the presence of a hydroxyl group at the 3 position on the C ring.
- They are generally glycosylated at the C-3 position.
- Onions, berries, cherries, apple, grapefruit and tea are rich sources of flavonols.

These include a series of polymethoxylated derivatives from species of the Asteraceae, where they are reported to occur in aerial parts as well as in leaf exudates. Species from the Rutaceae accumulate highly methoxylatedflavonols in leaves as well as in fruit peels, whereas species of *Fabaceae* were found to accumulate such compounds mainly in the heartwood . A hexamethoxylatedflavonol was isolated from

Distemonanthusbenthamianus (Fabaceae),[29] a species also known for accumulation of complex cycloflavonols . Of the Moraceae, only one report concerns the genus *Ficus*, which produces another hexamethoxylated flavonol
in the aerial parts,[30] . The same applies to accumulation of flavonolaglycones in roots of *Duroiahirsuta*(Rubiaceae) [31] .The number of 2'- and 5'-substituted derivatives appears to be lower than that of the corresponding flavones.

Most of the new source reports concern species from the Asteraceae, with many of the flavonols being isolated from aerial parts, where they are accumulated externally. They range from simple to more complex structures. There appears to be a tendency towards 6-methox-

ylation rather than towards 8-methoxylation, in addition to possible OMe-substitution of other positions of the flavonol molecule. Flavonols with 6,8-di-O-methylation and additional OMe-groups are also found in several genera such as *Senecio*,[32] *Psiadia*,[33] or*Inula*,[34] to cite but a few examples.

Aerial parts, fruits, flowers, and bark tissue of a series of Rutaceae species yielded a number of hexamethoxylatedflavonols. Once more, the complexity of metabolic pathways in this family is demonstrated by the formation of such compounds. The number of entries for this family is the second largest following the Asteraceae, but it must be taken into account that only a few genera of this large family are concerned. The third largest group concerns *Heliotropium* species of the family Boraginaceae, where particularly leaf exudates yielded flavonols.[35,36]

For species of *Alkanna*, flavonols were reported for aerial parts without indicating possible external occurrence.[37]

Interestingly, almost no flavones were reported From*Heliotropium*, and species of the genus *Nonea* were so far found to accumulate flavones only in their exudates.[36]

Further distribution studies will have to confirm the possible chemosystematic value of these accumulation trends.

A number of new listings concern the families of Scrophulariaceae and Solanaceae. In both cases the number of reports concerning external accumulation is also increased. Thus, further research will probably reveal that this phenomenon is more widespread in these

families as is obvious from the present data. In Fabaceae, most reports concern accumulation in heartwood, with a few exceptions such as leaves of *Millettia*racemosa.[38] However, no indication to possible external accumulation is made. Similar to flavone accumulation data, pollen of Myrtaceae were also found to accumulate flavonols.[20]

Very few reports exist on Gymnosperms such as *Cryptomeria* (Taxodiaceae)[39] or*Ephedra*,[40] without indication of external accumulation. So far, no new reports on flavones are known for these taxa.

In contrast to the numerous reports on flavones in Lamiaceae, only very few genera were found to accumulate flavonols in their exudates. The accumulation of 5,6-di-O-methylated derivatives in species of *Salvia*[41] may be of chemosystematic significance, in relation to other species of this genus. Single reports exist for families such as the Saxifragaceae[42] orNycta- ginaceae[43].

New results on Rosaceae and Viscaceae deserve special consideration. A quite complex derivative was isolated from several Rosaceae. By

contrast, rather simple derivatives were found in the leaf wax of *Viscum*spp.[44]

Particularly with the Rosaceae, more results in this direction are to be expected when more material is analyzed. Frond exudates of several ferns proved to be a rich source for various flavonol derivatives, which outnumber the few corresponding flavones.[45]

Several compounds were structurally revised. 6-Hydroxygalangin ,(as reported from *Platanus* buds, [46]was revised to 8-hydroxygalangin after synthesis.)

-Hydroxykaempferol-3,7,4'-triMe had been reported as ''tane-tin'' from *Tanacetumparthenium*.[48]

Its structure was later revised to 6-hydroxykaempferol-3,6,4'-triMe = santin).

The name ''tane-tin''is hence obsolete. 5,4-diOH-3,6,8-triOMe-flavone had been isolated from Tephrosia candida and named ''candiron'' .)

Synthesis revealed that thestructure must be revised to 5,4'-diOH-3,6,7-triOMe- flavone=penduletin).

The name ''candiron'' must not be used, therefore. ''Santoflavone,'' a com-pound isolated from *Achilleasantolina* and claimed to be 7-OH,3,6,3',4'-tetramethoxyflavone, was later revised to 5-hydroxy-6,7,3',4'-tetraOMe flavones.

Bhardwaj et al. had reported ''allopatuletin'' to be a ,3,6,7,3,4-pentahydroxy-5-methoxyflavone , from *Tagetespen-dula*.[54]

After synthesis, revision of this structure to quercetagetin-7-Me is requir.(Zhang et al. reported ‘‘viscidulin III’’ from the roots of *Scutellaria planipes*.[55]

Unfortunately, it remains dubious whether the authors used the name in its initial meaning, that is, as 3,5,7,3’-tetraOH-2’,4’-diOMe flavone or as its revised structure 5,7,3’,6-‘ tetraOH-8,2’-diOMe (for which the name ganhuangenin would apply).[56]

Since the authors did not answer relevant requests, this report has not been included in our tables.

1.2.3 Flavanones and Dihydroflavonols

They are characterized by absence of the double bond between the 2 and 3 positions on the heterocyclic C ring.Indihydroflavonols, the C-3 atom bears a hydroxyl group.

Flavanones (also called dihydroflavones) and dihydroflavonols (also called 3-hydroxyflavanones or flavanonols) lack the double bond between carbons 2 and 3 in the C-ring of the flavonoid skeleton, which is present in flavones and flavonols. Thus, in flavanones, C-2 bears one hydrogen atom in addition to the phenolic B-ring, and C-3 two hydrogen atoms.

Two stereoisomeric forms of each flavanone structure are possible, since C-2 is a center of asymmetry (epimeric center). Consequently, the B-ring can be either in the (2S)- or (2R)- configuration .The great majority of the flavanones isolated from plants are laevorotatory (_)- or (2S)-flavanones, because the enzymatic reaction catalyzing the conversionfchalcones to flavanones is stereospecific. The C-3 atom of dihydroflavonols bears both a hydrogen atom and a hydroxylgroup, and is therefore an additional center of asymmetry. Thus, four stereoisomers are possible for each

dihydroflavonol structure, (2R,3R), (2R,3S), (2S,3R), and (2S,3S). All four configurations have been found in naturally occurring dihydroflavonols, but the (2R,3R)-configuration is by far the most common.

As in all other flavonoids, there is structural variation in flavanones and dihydroflavonols
because of variation in hydroxylation, methoxylation, methylation, prenylation, benzylation, glycosylation, etc. of suitable carbon atoms in the skeleton, i.e., C-5, C-6, C-7, and C-8 of the A-ring, C-2', C-3', C-4', C-5', and C-6' of the B-ring, and C-2 of the C-ring in both flavanone .

- Flavanones exist in citrus fruits such as grapefruit, oranges and lemons.

The most common flavanones are:

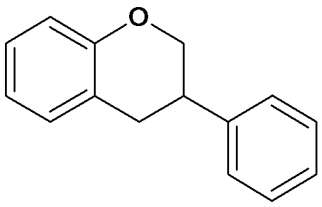
Naringenin and hesperetin are the aglycones of naringin and hesperidin, respectively.

Naringin is a major flavonoid in grapefruit and gives the grapefruit juice its bitter taste. Hesperidin, found abundantly in citrus fruits.

1.2.4 Isoflavonoids (3-benzopyrans)

In isoflavonoids, ring B occupiesposition[3]

- Isoflavan**12**
- Isoflavone**13**
- Isoflavanone **14**



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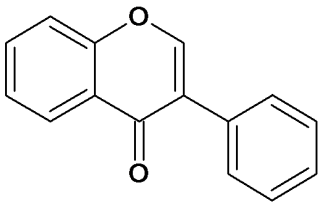

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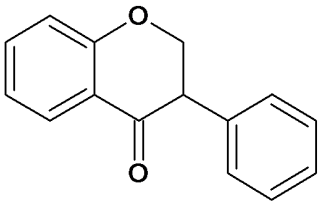

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The best isoflavones are:

- genistein (4\',5,7-triydroxyisoflavone).
- daidzein(4\',7-dihydroxyisoflavone).

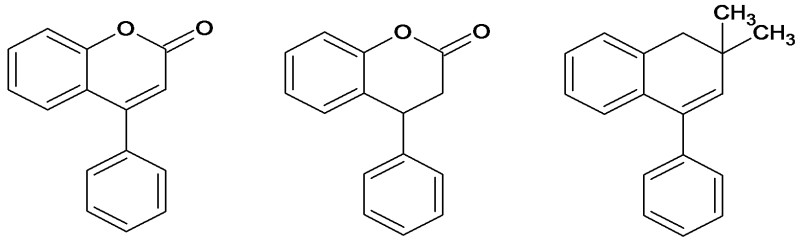
Isoflavonoids abundant in soybeans and consequently in a wide range of soy-derived foods and to a lesser extent in other legumes. Theiso flavonoids are a distinctive subclass of the flavonoids. These compounds possess a 3-phenylchroman skeleton that is biogenetically derived by 1,2-aryl migration in a 2-phenylchroman precursor. Despite their limited distribution in the plant kingdom, isoflavonoids are remarkably diverse as far as structural variations are concerne .

This arises not only from the number and complexity of substituents on the basic 3-phenylchroman system, but also from the different oxidation levels and presence of additional heterocyclic rings.

1.2.5 Neoflavonoids (4-benzopyrans):

The neoflavonoids are structurally and biogenetically closely related to theflavonoids and the isoflavonoids and comprise the 4-arylcoumarins (4-aryl-2*H*-1-benzopyran-2-ones), 3,4-dihydro-4-arylcoumarins, and neoflavenes.

- 4- arylcoumarin 15
- 3,4- dihydro-4-arylcoumarin **16**
- Neoflavene 17



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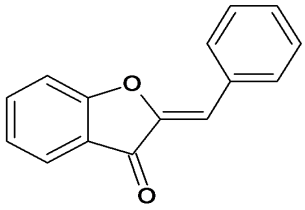
The Neoflavonoids are a group of chromane derivatives with ring B in position 4 (4- phenyl-coumains).

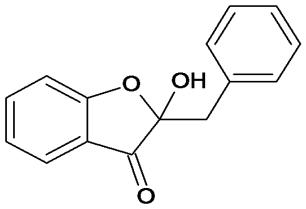
- The isoflavonoids and the neoflavonoids are regarded as abnormal flavonoids.

1.2.6 Minor Flavonoids :

Natural products such as chalcones and aurones also contain a C6-C3-C6 backbone and are considered to be minor flavonoids. These groups of compounds include the 2′-hydroxychalcones, 2′-OH-dihydrochalcones,2′-OH-*retro*-chalcone,aurones(2-benzylidenecoumaranone), and auronols .

- Aurone**18**
- Auronols 19





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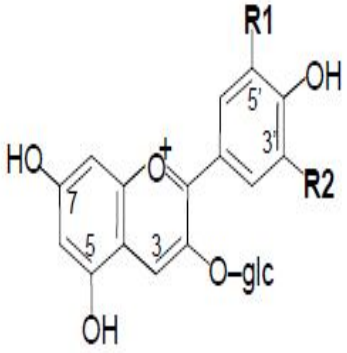
1.2.7 Anthocyanins 22:

The term anthocyanins refer to the glycosylated flavonoids whereas theaglycones are termed anthocyanidins which is structurally based on the flavyliumcation.

Anthocyanins are natural pigments responsible for the blue, purple, red and orange colors of many fruits and vegetables(flavyliumcation).

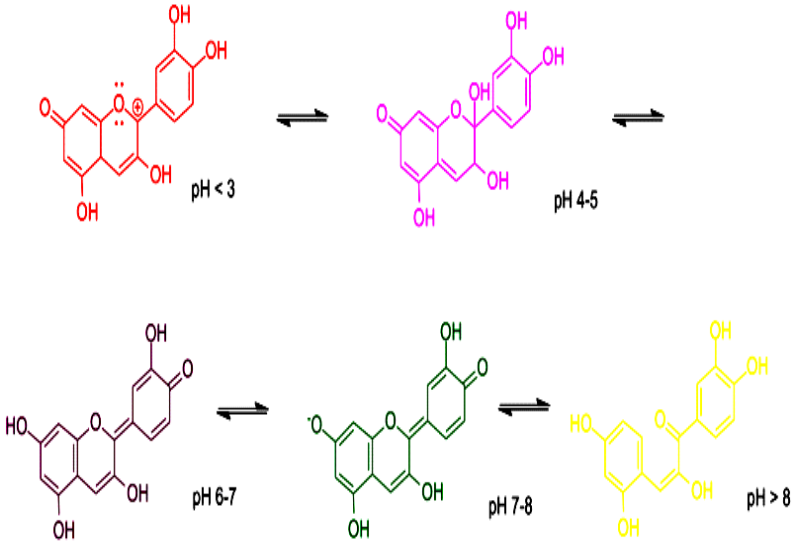
Depending on the number and position of the hydroxyl and methoxyl groups in the B-ring of the flavyliumcation, different anthocyanins have been described, and six of them are commonly found in fruitsand vegetables.fig (1)



R1	R2	Anthocyanidin
H	H	Pelargonidin
OH	H	Cyanidin
OH	OH	Delphinidin
OH	OCH_3	Petunidin
OCH_3	H	Peonidin
OCH_3	OCH_3	Malvidin

Fig(1):

- Anthocyanin colour is pH- dependent.

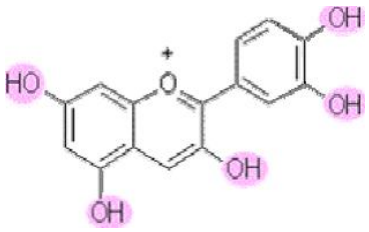


- Most anthocyanins contain two, three, or just one monosaccharide unit. The most common forms are 3-glycosides and 3, 5-diglycoside.

- The most common sugar present was glucose, but rhamnose, xylose and galactose were also encountered.

1- Cyanidin (Anthocyanidins). 20

2- Cyanin (Anthocyanin). 21



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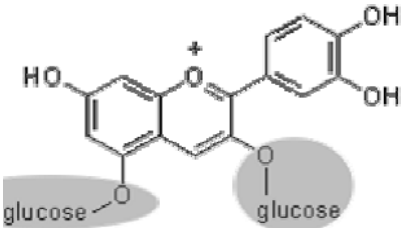

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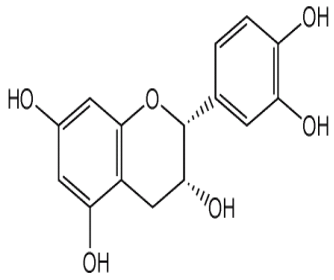
The main dietary sources of anthocyanins are red fruits, like berries and red grapes, cereals and purple corn, as well as some vegetables such as red cabbage.

. Catechins (Flavanols or Flavan-3-ols)

- They are very similar to those of anthocyanidins, but no positive charge on the oxygen atom and no double bonds in the C ring .
- The common flavan-3-ols that occur in many plants are catechin and its isomer, epicatechin. 22



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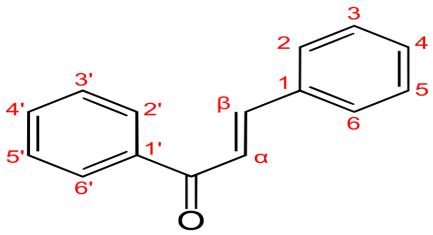
- The monomer catechin, or its isomer (epicatechin) are the building blocks for proanthocyanidins, which may be consisting of up to 50 catechin, or epicatechin subunits.
- Catechin polymers contribute a bitter taste and brown pigments to foods such as tea, cocas and dark chocolate.
- The most abundant monomeric flavanols of black tea
- Flavanols are sometimes conveniently called tea flavonoids. Besides tea, flavanols have also been determined at high level in chocolate, black grape, cherry and other fruits.

Flav-3-ols, such as epicatechin, catechin and epigallocatechin (and procyandins their polymers) are:

- Powerful antioxidants
- Have beneficial effects on cardiac health and immunity.

1.2.8 Chalcones 23:

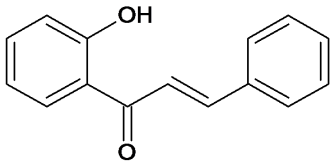
The unique feature that distinguishes chalcones from other flavonoids, the open-chain three-carbon structure linking the A and B rings in place of a heterocyclic C ring.



23

A typical chalcone is butein:

- - 2- OH –chalcone . 24
- 2-OH- dihydrochalcone . 25
- 2-OH- retro-chalcone . 26



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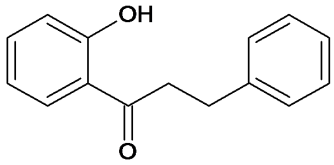

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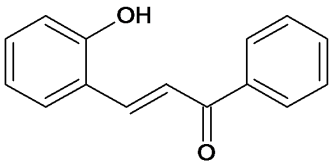
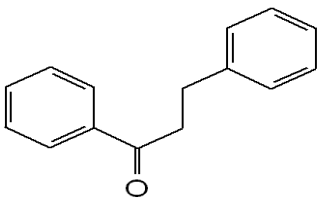


26

Dihydrochalcones 27, are directly related to the chalcones and are derived from them by reduction of the chalconeα-, β-double bond.



27

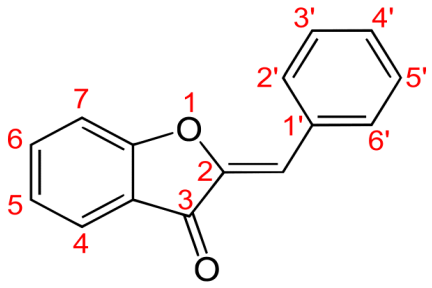
Dihydrochalcones[1] are directly related to the chalcones and are derived from them by reduction of the chalconeα-, β-double bond. The best known dihydrochalcone is phloridzin, which occurs in the skin of apple. A special property of some dihydrochalcones is their extremely sweet taste and there has been much interest in developing them as food sweeteners.

Pharmacological activity:

- anti-peptic
- anti-hepatotoxic activities
- more recently, chalcones have been found with anti-angiogenic effect.

1.2.10-Aurones28:

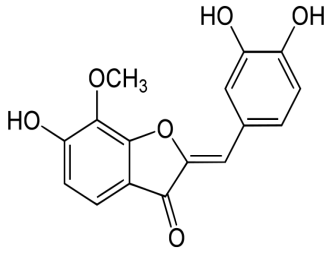
In aurone, the open-chain three-carbon structure linking the A and B rings in chalcone is closed into a 5-membered ring instead of the 6-membered ring more typical of flavonoids.



28

- Aurones are plant flavonoids that provide yellow color to the flowers of some popular ornamental plants.

- Leptosidin . 29 is the first aurone isolated as a yellow pigment from *Coreopsis grandiflora* (tickseed) in 1943.



29

Pharmacological activity:

- Aurones are anti-inflammatory agents.
- Recently, it was discovered that, aurones exhibit interesting antileishmanial properties

Aurones products:

AURONE® FORTE EAR DROPS

- Used to treat inflammatory conditions and infections of the ear.

1.3 EXTRACTION:

Flavonoids (particularly glycosides) can be degraded by enzyme action when collected plant material is fresh or nondried. It is thus advisable to use dry, lyophilized, or frozen samples.

When dry plant material is used, it is generally ground into a powder. For extraction, the solvent is chosen as a function of the type of flavonoid required. Polarity is an important consideration here. Less polar flavonoids (e.g., isoflavones, flavanones, methylated flavones, andflavonols) are extracted with chloroform, dichloromethane, diethyl ether, or ethyl ace-tate, while flavonoid glycosides and more polar aglycones are extracted with alcohols or alcohol–water mixtures. Glycosides have increased water solubility and aqueous alcoholic solutions are suitable. The bulk of extractions of flavonoid-containing material are still performed by simple direct solvent extraction.

Powdered plant material can also be extracted in a Soxhlet apparatus, first with hexane, for example, to remove lipids and then with ethyl acetate or ethanol to obtain phenolics. This approach is not suitable for heat-sensitive compounds.

A convenient and frequently used procedure is sequential solvent extraction. A first step, with dichloromethane, for example, will extract flavonoid aglycones and less polar material.

A subsequent step with an alcohol will extract flavonoid glycosides and polar constituents.

Certain flavanone and chalcone glycosides are difficult to dissolve in methanol, ethanol, or alcohol–water mixtures. Flavanone solubility depends on the pH of water-containing solutions.

Flavan-3-ols (catechins, proanthocyanidins, and condensed tannins) can often be extracted directly with water. However, the composition of the extract does vary with the solvent — whether water, methanol, ethanol, acetone, or ethyl acetate. For example, it is claimed that methanol is the best solvent for catechins and 70% acetone for procyanidins.[8]

Anthocyanins are extracted with cold acidified methanol. The acid employed is usually acetic acid (about 7%) or trifluoroacetic acid (TFA) (about 3%). The use of mineral acid can lead to the loss of attached acyl groups.

Extraction is typically performed with magnetic stirring or shaking but other methods have recently been introduced to increase the efficiency and speed of the extraction procedure.

The first of these is called pressurized liquid extraction (PLE). By this method, extraction is accelerated by using high temperature and high pressure. There is enhanced diffusivity of the solvent and, at the same time, there is the possibility of working under an inert atmosphere and with protection from light. Commercially available instruments have extraction vessels with volumes up to about 100ml. In a study involving medicinal plants, solvent use was Andersen and Markham / Flavonoids: Chemistry, Biochemistry, and Applications #2021_c001 Revise Proof page 2 8.9.2005 9:23pm Flavonoids: Chemistry, Biochemistry, and Applicationsreduced by a factor of two.[9]

The optimization of rutin and isoquercitrin recovery from older)Sambucusnigra, Caprifoliaceae) flowers has been described. Application of PLE gave better results than maceration — and shorter extraction times and smaller amounts of solvent were required.[10]

PLE of grape seeds and skins from winemaking wastes proved to be an efficient procedure for obtaining catechin and epicatechin with little decomposition, provided the temperature was kept below 1308C.[11]

As its name suggests, supercritical fluid extraction (SFE) relies on the solubilizing proper-ties of supercritical fluids. The lower viscosities and higher diffusion rates of supercritical fluids, when compared with those of liquids, make them ideal for the extraction of diffusion-controlled matrices, such as plant tissues. Advantages of the method are lower solvent con-sumption, controllable selectivity, and less thermal or chemical degradation thanmethods such asSoxhlet extraction. Numerous applications in the extraction of natural products have been reported, with supercritical carbon dioxide being the most widely used extraction solvent.[12,13]

However, to allow for the extraction of polar compounds such as flavonoids, polar solvents)like methanol) have to be added as modifiers. There is consequently a substantial reduction in selectivity. This explains why there are relatively few applications to polyphenols in the literature. Even with pressures of up to 689 bar and 20% modifier (usually methanol) in the extraction fluid, yields of polyphenolic compounds remain low, as shown for marigold)*Calendulaofficinalis*, Asteraceae) and chamomile (*Matricariarecutita*, Asteraceae).[14]

Ultrasound-assisted extraction is a rapid technique that can also be used with mixtures of immiscible solvents: hexane with methanol–water (9:1), for example, is a system used for the Brazilian plant *Lychnophoraericoides* (Asteraceae). The hexane phase concentrated less polar sesquiterpene lactones and hydrocarbons, while the aqueous alcohol phase concentrated flavonoids and more polar sesquiterpene lactones.[15]

Microwave-assisted extraction (MAE) has been described for the extraction of various compounds from different matrices.[16]

It is a simple technique that can be completed in a few minutes. Microwave energy is applied to the sample suspended in solvent, either in a closed vessel or in an open cell. The latter allows larger amounts of sample to be extracted. A certain degree of heating is involved.[17]

1.4 Chrozophoraplicata

Genus: chrozophora.

Family: Euphorbiaceae.

Sub family: Acolypholdeae.

Tribe: Chrozophoreae.

Subtribe: chrozophorinae.

Namenumber: 401609.

Placeofpublication: Syst, veg, 3:850, 1826.

Nameverifiedon: 27-Mar-1995 by ARS.

Commonname: giradol(source: comucopia).English.

Economic Importance:

-Materials: Lipids (for soap and pigment fide pi book) .

Mor: viewethootanical data from dukels Phytochemical and Ethnobotanical Data bases.

2- Material and Method

The plant was identified by my project supervisor (Mrs M-abdalkareem).

2-1 Sample Collection:

Theplant chrozophoraplicatawas collected from kurdofan state

2-2 Preparation of the Sample:

The leaves of chrozophoraplicata were air-dried after collection . The dried leaves were ground into fine powder and then weighed.

2-3 Extraction of the Sample:

Solvent-solid extraction was carried out on the weighed, air-dried and leaves of chrozophoraplicataThe weighed sample was soaked with methanol (95%) for two days , with continuose shaking . The separation of the residue from filtrate was done by using filter paper. It was followed by the concentration of the filtrate by using distillation method .

2-4 Chromatography:

The type of chromatographic method was used to separate the constituents that were present in the extract is thin-layer chromatography.

2-4-1 Thin-Layer Chromatography (TLC):

TLC was used to ascertain the number of constituents present in the extract and to determine their purity. TLC was also used to determine the solvent mixture that will affect the separation of the components. The TLC were eluted with butanol :acetic acid : water (4 : 1.5 : 6) .

-Preparation of Silica Gel Plates:

50g of silica gel powder was weighed into a conical flask, 100ml of distilled water was added and the resulting solution was shaken vigorously in order to avoid lumps. The white smooth paste mixture was spread over the glass plate and was allowed to solidify. The coated glass plates were put inside oven for 1-2 hours at 1100C to ensure further solidification.

-Spotting of the Plates:

This is done with aid of capillary tubes to introduce few drops of the dissolved sample extract unto the coated plate, allowing each drop to dry before adding another drop.

-Developing of the Plates:

After the solvent had travelled some distance across the plate, the plate was removed and allowed to dry and then spray a clouration reagent which is vaanalin solution with concentrated sulfuric acid . The separated components appeared as dark braown-red spots.

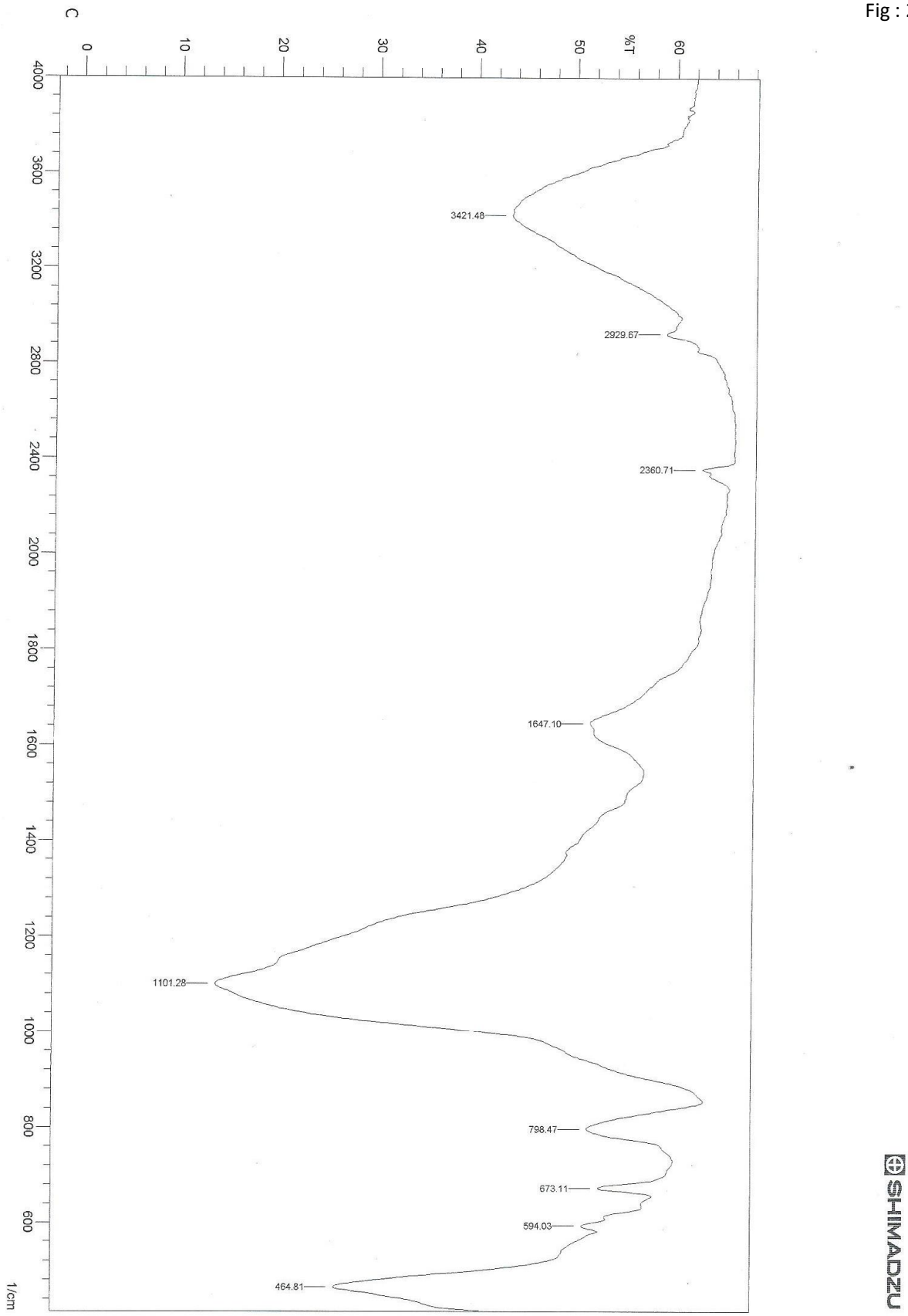
3. Result and discussion

The flavonoids of chrozophoraplicata were extracted by ethanol. After removing the ethanol solvent.Acrud product was fractionated by TLC (silica gel, 50% acetic acid). After the usual work up a pure flavonoid chrozophoraplicata compound was isolated.

In the IR spectrum(fig.2) it gave v(KBr) 3421.48(O- H, H –bond) , 2929.67 (C-H) , 2360.71 (C-N) , 1647.10 (C=C,Ar) , 1101.28 (C-O), 798.47 , 673.11 ,594.03 , 494.81 (are benzene ring) . fig (2).

Since the IR related absence of C=O st.vib then this flavonoids is either an anthocyanin of flavan.

Fig : 2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REFERENCES:

1 . Harborne, J.B. andWilliams, C.A., Advances in flavonoid research since 1992. *Phytochemistry*, 55,184 ,(2000).

2 .Bohm, B.A. and Stuessy, T.F., *Flavonoids of the Sunflower Family* (*Asteraceae*). Springer, Vienna/ New York,(2001).

3 .Barron, D. and Ibrahim, R.K., Isoprenylated flavonoids—a survey, *Phytochemistry*, 43, 921,(1996).

4 .Harborne, J.B. and Baxter, H., *The Handbook of Natural Flavonoids*, vol. 1. Wiley & Sons, Chichester, (1999).

5 .Wollenweber, E. and Dietz, V.H., Occurrence and distribution of free flavonoid aglycones in plants, *Phytochemistry*, 20, 869, (1981).

6 .Wollenweber, E., Flavones and flavonols, in *The Flavonoids — Advances in Research since 1986*, J.B. Harborne, Ed., Chapman and Hall, London, Chap. 7,(1994.)

7 .Chung, H.S. et al., Flavonoid constituents of *Chorizanthe diffusa* with potential cancer chemopre- ventive activity, *J. Agric. Food Chem.*, 47, 36, (1999).

9 .Mossa, J.S. et al., A flavone and diterpene from *Psiadia arabica, Phytochemistry*, 31, 2863, (1992).

20 .Campos, M.G., Webby, R.F., and Markham, K.R., The unique occurrence of the flavone aglycon tricetin in Myrtaceae pollen, *Z. Naturforsch*., 58c, 944,(2002).

21 .Mayer, R., A b-hydroxychalcone from *Leptospermum scoparium, Planta Med*., 59, 269,(1993).

22 .Kraut, L., Klein, R., and Mues, R., Flavonoid diversity in the liverwort genus *Monoclea, Z. Naturforsch*., 47c, 794,(1992).

23 .Takagi, S., Yamaki, M., and Inoue, K., Flavone di-C-glycosides from Scutellaria baicalensis, *Phytochemistry*, 20, 2443,(1981).

24 .Horie, T. et al., Synthesis of 5,8-dihydroxy-6,7-dimethoxyflavones and revised structures for some natural flavones, *Phytochemistry*, 39, 1201,(1995).

25 .La Duke, J.C., Flavonoid chemistry and systematics of *Tithonia, Am. J. Bot*., 69, 784,(1982).

26 .Martinez, V. et al., Phenolic and acetylenic metabolites from *Artemisia assoana, Phytochemistry*,26,2619,(1987)..

27 .Gonza ´ lez, A.G. et al., Methoxyflavones from *Ageratum conyzoides*, *Phytochemistry*, 30,1269,(1991).

28 .Horie, T. et al., Studies of the selective O-alkylation and dealkylation of flavonoids. XIV. A convenient method for synthesizing 5,6,7,8-trihydroxy-3-methoxyflavones from 6hydroxy-3,5- ,trimethoxyflavones, *Bull. Chem. Soc. Jpn,* 66, 877,(1993).

29 .Malan, E., A flavonol with a tetrasubstituted B-ring from *Distemonanthus benthamianus, Phyto-chemistry,* 32, 1631,(1993).

30 .Sharaf, M., Abu-Gabal, N.S., and El-Ausari, M.A., Exudate flavonoids from *Ficus altissima, Biochem. Syst. Ecol.*, 28, 291,(2000).

31 .Aquino, R. et al., New 3-methoxyflavones, an iridoid lactone and a flavonol from *Duroia hirsuta, J. Nat. Prod*., 62, 560,(1999).

32 .Wollenweber, E. et al., Exudate flavonoids in several Asteroideae and Cichorioideae (Asteraceae,(*Z. Naturforsch*., 52c, 137,(1997).

.33. Midiwo, J.O. and Owuor, F.A.O., Epicuticular flavonoids of *Psiadia puntulata and Polygonum senegalense, 5th NAPRECA Symposium on Natural Products, Antananarivo, Madagascar*, p. 81, (1992).

34 .Ahmad, V.U. and Ismail, N., 5-Hydroxy-3,6,7,2',5'-pentamethoxyflavone from *Inula grantioides, Phytochemistry*, 30, 1040, (1991).

35 .Urzua, A. et al., Flavonoids in the resinous exudate of *Chilean Heliotropium* species from *Cochranea* section*, Biochem. Syst. Ecol*., 21, 744, (1993).

36 .Wollenweber, E. et al., On the occurrence of exudate flavonoids in the Borage family (Boragina- ceae), *Z. Naturforsch*., 57c, 445, (2002).

37 .El-Sohly, H.N. et al., Antiviral flavonoids from *Alkanna orientalis*, *Planta Med*., 63, 384, (1997).

38 .Ganapaty, S., Pushpalatha, V., and Naidu, K.C., Flavonoids from *Millettia racemosa* Benth,. *Indian Drugs,* 36, 635, (1999).

39 .Su, W.-C., Fang, J.-M., and Cheng, Y.-S., Flavonoids and lignans from leaves of *Cryptomeria japonica*, *Phytochemistry*, 40, 563, (1995).

.40 Hussein, S.A. et al., Flavonoids from *Ephedra aphylla*, *Phytochemistry*, 45, 1529, (1997).

.41. Wollenweber, E. et al., Exudate flavonoids of some *Salvia* and a *Trichostoma species*, *Z. Natur- forsch*., 47c, 782, (1992).

42 .Urzua, A. et al., Flavonoid aglycones in the resinous exudate of some Chilean plants, *Fitoterapia*,62,385,(1991).

.

43. Wollenweber, E. and Do ¨ rr, M., Exudate flavonoids from aerial parts of *Mirabilis viscosa* (Nycta- ginaceae), *Biochem. Syst. Ecol*., 24, 799, (1996).

44 .Wollenweber, E., Wieland, A., and Haas, K., Epicuticular waxes and flavonol aglyones of the European mistletoe, *Viscum album*, *Z. Naturforsch.,* 55c, 314,(2000).

.45. Wollenweber, E. et al., Flavonoid aglycones and a dihydrostilbene from the frond exudate of *Notholaena nivea, Phytochemistry*, 33, 611, (1993).

46 .Kaouadji, M. et al., 6-Hydroxygalangin and C-prenylated kaempferol derivatives from *Platanus acerifolia* Buds., *Phytochemistry*, 31, 2131, (1992)..

48 .Williams, C.A. et al., A biologically active lipophilic flavonol from *Tanacetum parthenium, Phyto- chemistry*, 38, 267, (1995).

54 .Bhardwaj, D.K. et al., Quercetagetin-5-methyl ether from the petals of *Tagetes patula, Phytochem- istry,* 19, 713, (1980).

55 .Zhang, Y.-Y. et al., Studies on the constituents of roots of *Scutellaria planipes, Planta Med.*, 63,36,(1997).

56 .Wollenweber, E. and Jay, M., Flavones and flavonols, in *The Flavonoids — Advances in Research since 1980*, J.B. Harborne, Ed., Chapman and Hall, London/New York, Chap. 7, (1988).

.8. Hussein, L., Fattah, M.A., and Salem, E., Characterization of pure proanthocyanidins isolated from the hulls of faba beans, *J. Agric. Food Chem*., 38, 95, (1990).

9 .Benthin, B., Danz, H., and Hamburger, M., Pressurized liquid extraction of medicinal plants, *J. Chromatogr.* A, 837, 211,(1999).

10 .Dawidowicz, A.L. et al., Optimization of ASE conditions for the HPLC determination of rutin and isoquercitrin in *Sambucus nigra* L., *J. Liq. Chromatogr. Relat. Technol*., 26, 2381,(2003).

11 .Pineiro, Z., Palma,M., and Barroso, C.G., Determination of catechins by means of extraction with pressurized liquids, *J. Chromatogr. A*, 1026, 19,(2004).

12 .Bevan, C.D. and Marshall, P.S., The use of supercritical fluids in the isolation of natural products, *Nat. Prod. Rep*., 11, 451, (1994).

13 .Jarvis, A.P. and Morgan, E.D., Isolation of plant products by supercritical fluid extraction, *Phytochem. Anal*., 8, 217,(1997).

.14. Hamburger, M., Baumann, D., and Adler, S., Supercritical carbon dioxide extraction of selected medicinal plants — effects of high pressure and added ethanol on yield of extracted substances, *Phytochem. Anal*., 15, 46,(2004).

.15. Sargenti, S.R. and Vichnewski,W., Sonication and liquid chromatography as a rapid technique for extraction and fractionation of plant material, *Phytochem. Anal.*, 11, 69, (2000).

16 .Ganzler, K., Szinai, I., and Salgo, A., Effective sample preparation method for extracting bio- logically active compounds from different matrices by a microwave technique, *J. Chromatogr*., 520,257,(1990).

.17. Kaufmann, B. and Christen, P., Recent extraction techniques for natural products: microwave- assisted extraction and pressurized solvent extraction, *Phytochem. Anal*., 13, 105,(2002).

- Constituent of medicinal plant S.Babad,(1970).Cairo,Eygypt.
- Pharmacosrosy trees flavan,(1989).