FLAVONOIDS

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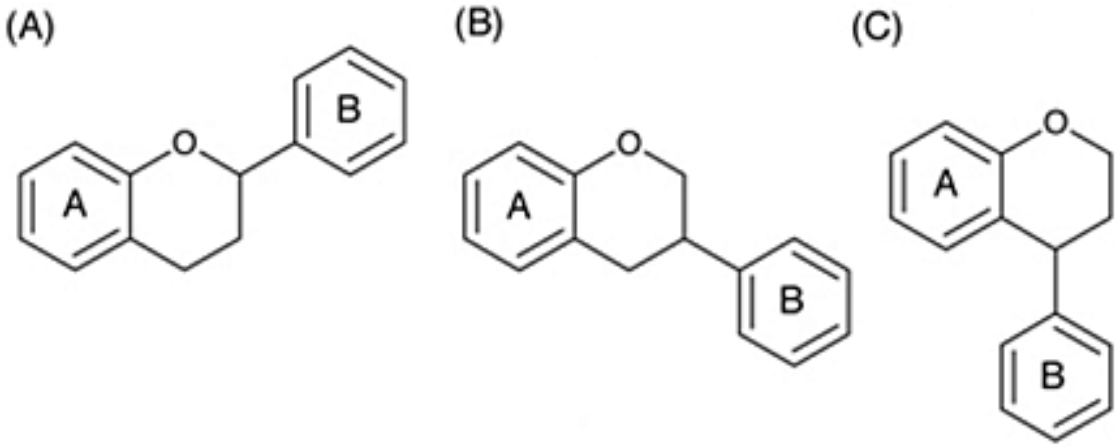
Flavonoids

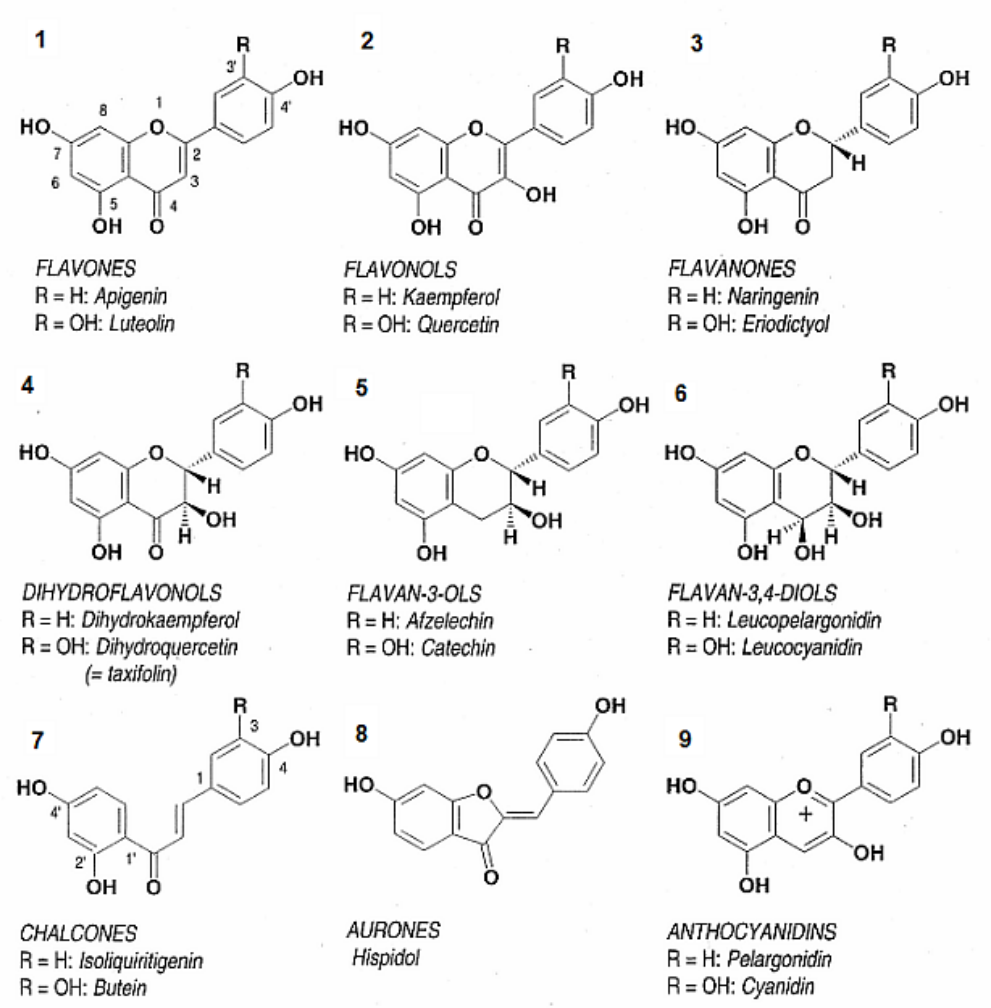
- Flavonoids in the broad sense of the term are virtually **universal plant pigments**.
- Almost always **water-soluble**, they are responsible for the **color** of flowers, fruits, and sometimes leaves,
- Examples are **yellow flavonoids** (chalcones, aurones, and yellow flavonols) and **red, blue, or purple anthocyanins**.
- When they are not directly visible, they contribute to the color by acting as **copigments:** for example, colorless flavone and flavonol copigments protect anthocyanins.

- All flavonoids (~4000) have a *common biosynthetic origin*, and therefore possess *the same basic structural element*: → the 2- (A), or 3- (B) or 4- (C) -**phenylchromane skeleton .**
- **Flavonoids (A), isoflavonoids (B), neoflavonoids (C).**



Flavonoids

- They fall into about a dozen classes depending on the degree of oxidation of the central pyran ring, which can be opened and recyclized into a furan ring (dihydrofuranone):
- 2-phenylbenzopyriliums (9), 2-phenylchromones (1, 2, 3, 4), 2-phenylchromanes (flavans, 5, 6); chalcones and dihydrochalcones (the pyran ring opens); 2-benzylidene coumaranones (= aurones).



Flavones, Flavonols

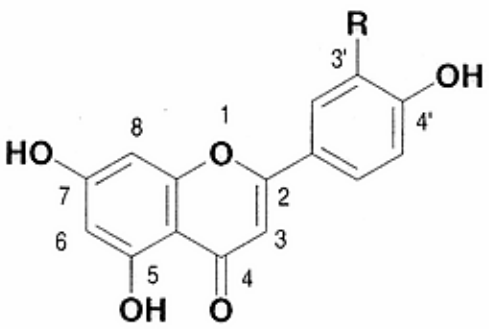
They represent the majority of known flavonoids in the strict sense.

Ring A substituents

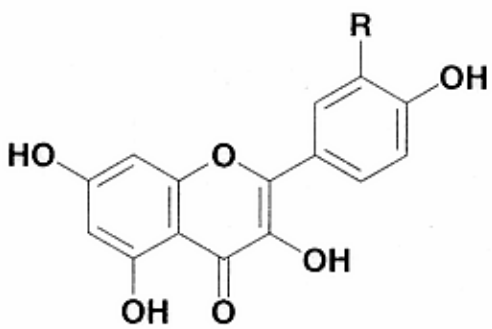
- **Phenolic hydroxyl groups** at C-5 and C-7 , (in over 90% of cases), free or etherified, or one of them engaged in a glycosidic linkage.
- **Other substitutions** :
  - free or etherified -OH groups at C-6 or C-8 or both,
  - isoprenylation or methylation at C-6 or C-8, or
  - involvement of C-6, C-8, or both in a carbon-carbon bond with a saccharide .

B ring

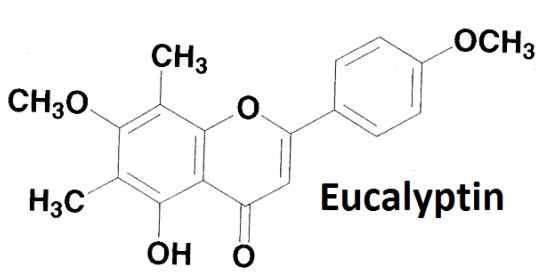
- may be substituted at C-4' , or 3',4'-di-substituted , or 3',4',5'-trisubstituted; by -OH or -OCH_3
- Exceptionally, substituents at 2' and 6'can occure.



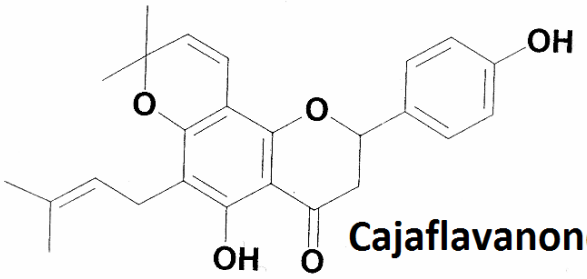
FLAVONES
R = H: *Apigenin*
R = OH: *Luteolin*



FLAVONOLS
R = H: *Kaempferol*
R = OH: *Quercetin*



Eucalyptin



Cajaflavanone

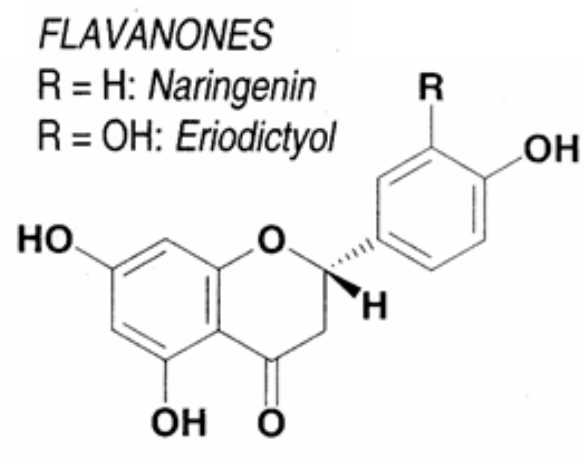
Flavanones and Dihydroflavonols

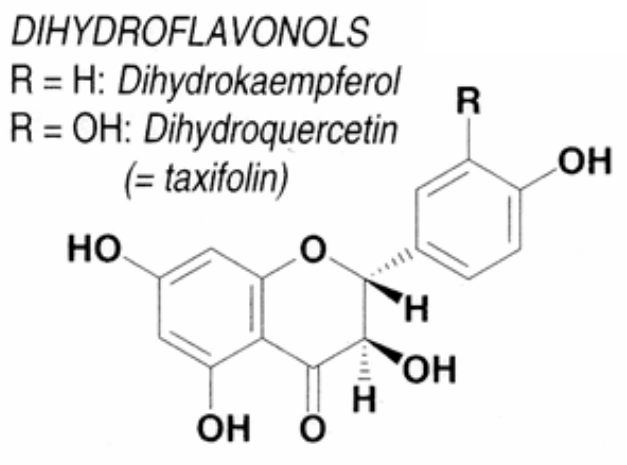
Flavanones, charcteristics:

- **absence of a 2,3-double bond**
- **presence of at least one asymmetric center**,
- **C-2 is normally in the 2*S* configuration (**natural flavanones**)**.

Dihydroflavonols:

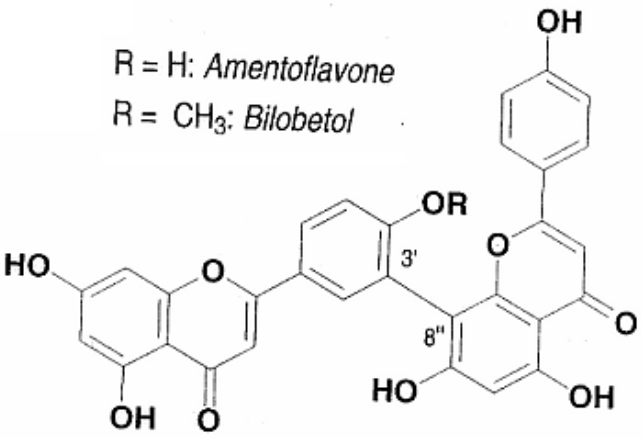
- Mostly **2*R*,3*R* configuration**, with **the phenyl and hydroxyl groups in trans** (4 isomers are possible in theory).
- Structural variations are the same as those described above for flavones and flavonols.
- They are **less common** than their unsaturated homologs
- Some families tend to accumulate their **C-alkylated derivatives** (Asteraceae, Fabaceae).

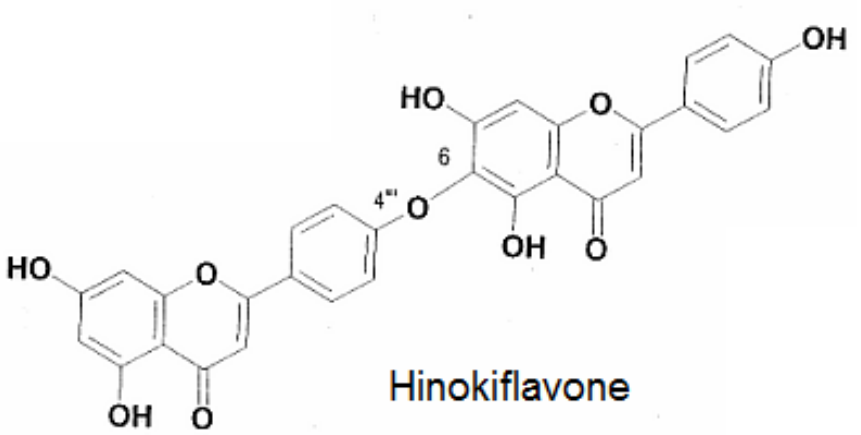




Biflavonoids

- Flavonoids can also bond to one another, particularly through their **very reactive C-6 or C-8**. The result is a dimer known as a biflavonoid.
- Most frequently, biflavonoids are dimers of flavones and flavanones, are generally 5,7,4'-trisubstituted.
- The **interflavanic linkage** can be
  - of the **carbon-carbon-type** (3' ,8", e.g., in amentoflavone; 6,8", e.g., in agathisflavone; 8,8", e.g., in cupressiflavone) or
  - of the **carbon-oxygen-carbon-type** (6-0-4", e.g., in hinokiflavone).
- The two consecutive units of the biflavonoid may or may not be of the same type (biflavone, biflavanone, flavone-flavanone, flavanone-chalcone).
- The OH groups may be free or (frequently) methylated. In this group, few glycosides are known.
- Biflavonoids are **characteristic of the Gymnosperms** (see above), sporadic in the Angiosperms (e.g., Hypericum, Semecarpus, Schinus, Garcinia).



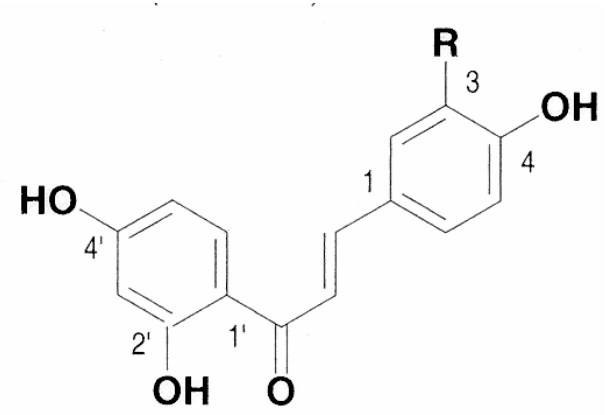


Chalcones, Aurones

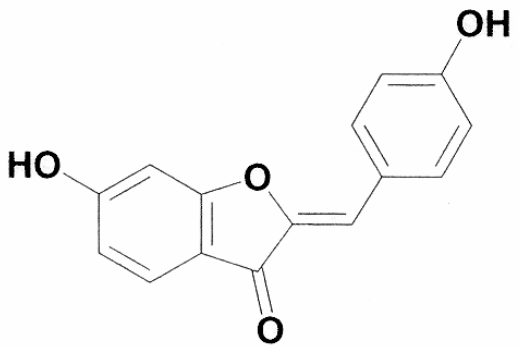
Chalcones are characterized by

- the lack of a central heterocyclic nucleus, and
- a **three-carbon chain with a ketone function** + **α,β-unsaturation**.
- Substitutions on the **A ring** are most often identical to those of other flavonoids (2',4',6'), whereas the **B ring** is fairly often unsubstituted.
- **Isoprenyl-** and **pyranochalcones** seem **rather common**, especially in the **Fabaceae.**

Aurones are characterized by a **2-benzylidene- coumaranone** structure.

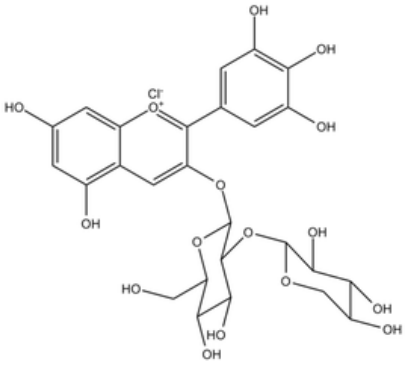


CHALCONES
R = H: *Isoliquiritigenin*
R = OH: *Butein*



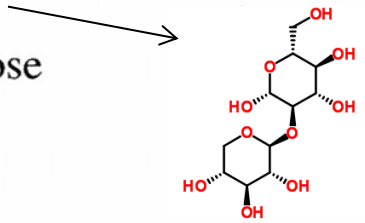
AURONES
Hispidol

Glycosyflavonoids



- The **sugar moiety** may be a mono-, di-, or trisaccharide.
- **Monosaccharides** include
  - D-glucose, D-galactose or D-allose,
  - pentoses (D-apiose, L-arabinose, L-rhamnose, or D-xylose), or
  - D-glucuronic or D-galacturonic acid.
- **Disaccharide** and **trisaccharide moieties** may be linear or branched.

structure	usual name
O-β-D-xylosyl-(1—>2)-glucose	sambubiose
O-α-L-rhamnosyl-(1—>2)-glucose	neohesperidose
O-α-L-rhamnosyl-(1—>6)-glucose	rutinose
O-β-D-glucosyl-(1—>2)-glucose	sophorose
O-β-D-glucosyl-(1—>6)-glucose	gentiobiose
O-β-glucosyl-(1—>2)-*O*-β-glucosyl-(1—>2)-glucose	sophorotriose
O-α-rhamnosyl-(1—>2)-*O*-β-glucosyl-(1—>3)-glucose	2'-rhamnosyl-laminaribiose
O-α-rhamnosyl-(1—>4)-*O*-[α-rhamnosyl-(1—>6)-galactose]	4^{Gal}-rhamnosylrobinobiose



Examples of di- and trisaccharides found in glycosylflavonoids

Glycosylflavonoids
(continued)

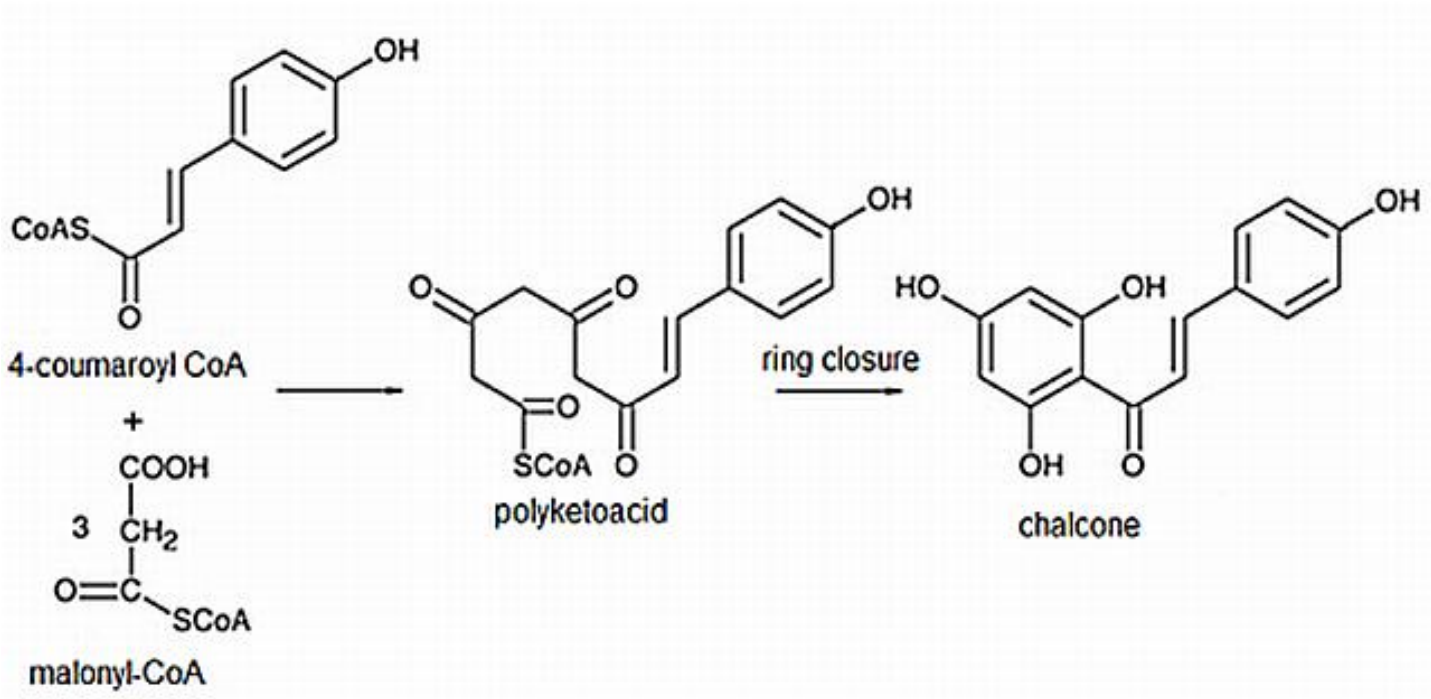
- Advances in analytical technology, especially in the field of **mass spectrormetry** or **MS** (fast atom bombardment-MS or **FAB-MS**, electrospray ionization MS or **ESI-MS**), make possible the characterization of an increasing number of acylated structures.
- **Acylated structures**, in which a **hydroxyl group** of the sugar moiety is **esterified by**
  - **an aliphatic acid** (acetic, malonic, tiglic, and others) or
  - **an aromatic acid** (gallic, benzoic, 4-coumaric, and other cinnamic derivatives).
- **Sulfated flavonoids** (>80 known structure).

Special Case: *C*-Glycosylflavonoids

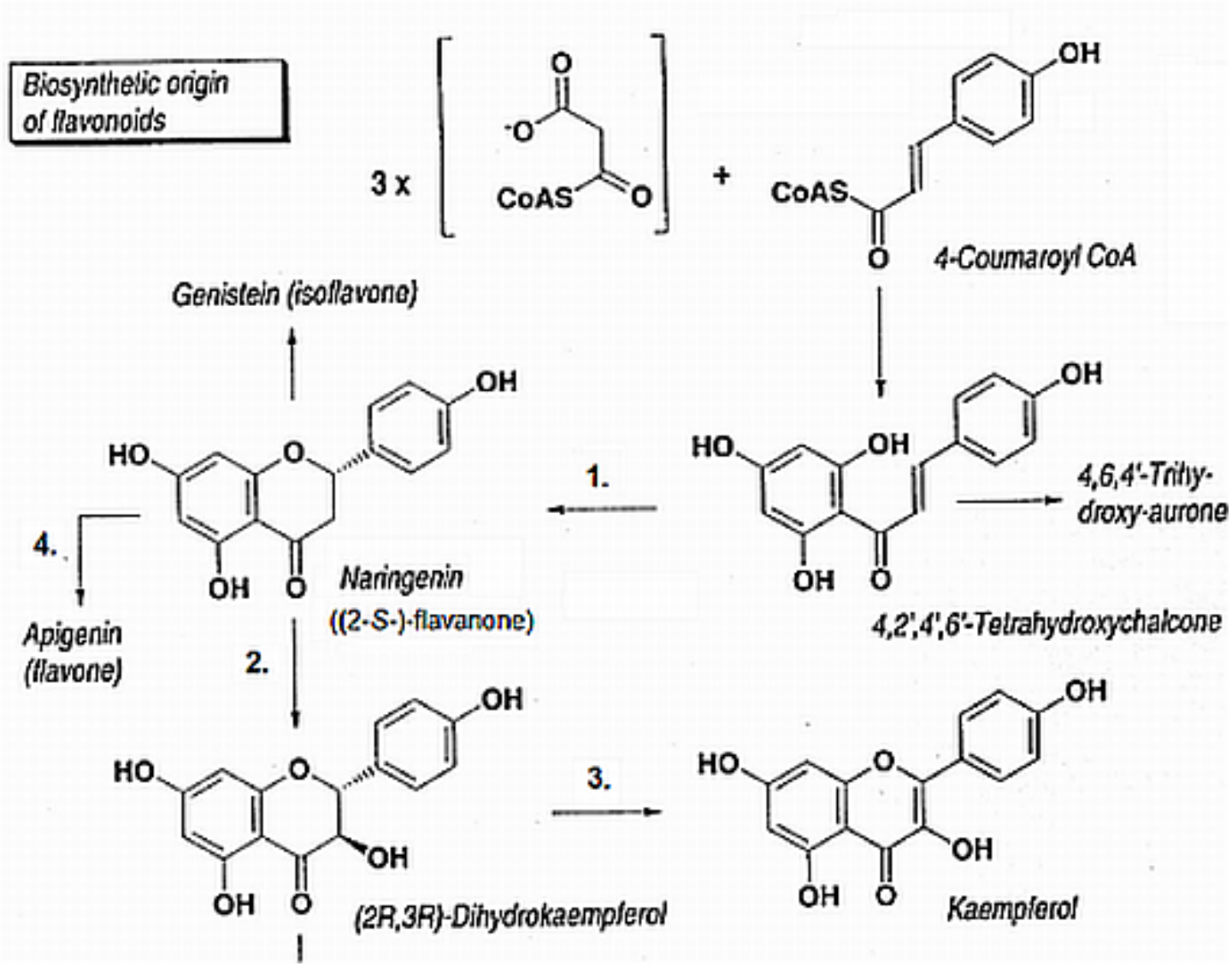
- More than 300 known structure.
- The **bond** is established **between**
  - the **asymmetric carbon on the sugar** (often glucose, also galactose or a pentose) and
  - the **C-6 or C-8 of the aglycone** (often a flavone, but another type is also possible, including a flavonol or a chalcone).
- **Structural types**:
  1. mono-*C*-glycosylflavonoids (e.g., scoparoside in Scotch broom);
  2. di-*C*-glycosyl-flavonoids (e.g., isoschaftoside in tea);
  3. *C*-glycosyl-*O*-glycosylflavonoids (e.g., saponarin [= 7-*O*-glucosylisovitexin] in passion flower);
  4. acyl- *C*-glycosyl-flavonoids (e.g., 4”- *O*-acetyl-2’‘-rhamosylvitexin in hawthorn).
- **Wessely-Moser isomerization.**
  - In derivatives of the *5-hydroxy-C-glycosyl- flavone type,* the heterocycle opens readily in acidic conditions, which explains their facile isomerization (6 <—> 8, 8 <—> 6).
  - This isomerization remains of interest for the **structure elucidation** of these compounds.

BIOSYNTHETIC ORIGIN I.

- The **key step** in the formation of flavonoids is the **condensation**, catalyzed by **chalcone synthase**, of three molecules of **malonyl-CoA** with an ester of coenzyme A and a hydroxycinnamic acid, as a general rule **4-coumaryl-CoA**.
- The reaction product is a chalcone, generally 4,2',4',6'-tetra- hydroxychalcone.

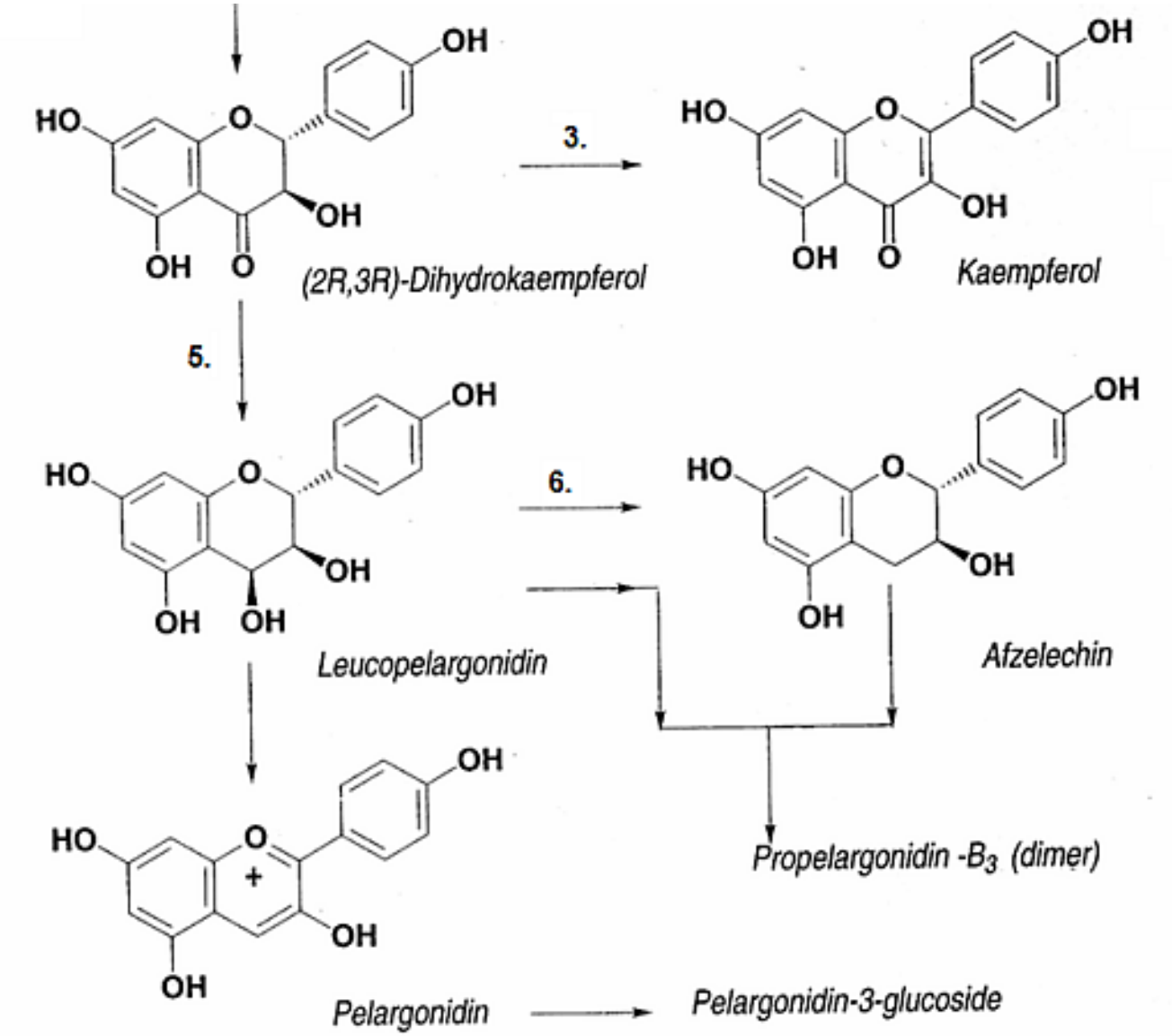


BIOSYNTHETIC ORIGIN II.



1. Chalcone **isomerase** , 2. Flavanone 3-**hydroxylase**, 3. Flavanol **synthase**, 4. Flavanone **synthases** I.& II.

BIOSYNTHETIC ORIGIN III.



5. Dihydroflavonol 4-reductase , 6. Leucoanthocyanidine 4-reductase

BIOLOGICAL PROPERTIES

- The main property that is recognized for flavonoids is “venoactivity”, in other words their ability to decrease capillary permeability and fragility.
- In animal models, they can decrease the signs of experimental vitamin C deficiency (“vitamin P”; lack of defeciency syndrome: “vitamin P factors” → “P factors”).
- Flavonoids are now referred to as “venoactive” or “vascular protective and venous tonic agents”.
- The US Food and Drug Administration (= FDA) does not recognize any activity for them; their therapeutic value is generally minimized (in reference books).
- Nevertheless, flavonoids and flavonoid based preparations are—in France, Germany, Spain, Italy—widely prescribed, often recommended by pharmacists, and commonly self-prescribed to **treat minor circulatory disorders**.
- In addition, a few compounds in this class have proven **efficacy**, at least **at high doses**.
- Fields of flavonoid reserach interest:
  - interaction between **flavonoids and free radicals**, and its potential applications in **preventive therapy**,
  - activity of flavonoids on the **cells and systems** involved in **immune responses** and **inflammation**

Flavonoids and Free Radicals

- Biochemically, **free radicals** are thought to be responsible for nucleic acid alterations, mutations, initiation/promotion of carcinogenesis, and cellular damage, because of their ability to react with membrane phospholipids, among other reasons.

- Despite the lack of absolute proof, and despite the fact that their physiological role is not yet completely elucidated,
- **free radicals** are believed to be in part responsible for the genesis of atheromatous lesions, the beginning of some cancers, and neurological degeneracy.

- This has spurred research, including **epidemiologic studies** on the potential **role of antioxidants** (i.e., free radical scavengers) **flavonoids**, some lignans, other metabolites found in the daily diet, **in preventive therapy**.

- The antagonist effect towards free radical production can be studied experimentally.
  - Using experimental free radicals (hydroxyl radical, diphenylpicryihydrazyl radical),
  - **radical scavenging capability can be measured** in vitro (by ESR, or colorimetrically) on a lipid peroxidation model, or else
  - the activity can be evaluated *in vivo* by comparison with that of a reference antioxidant.
- Many flavonoids in the broad sense and many other phenols (especially tocopherols [= vitamin E]) react with free radicals **to prevent the degradations** linked to their intense reactivity.
- It appears that the **antioxidant capability** of a flavonoid depends on its affinity for free radicals, consequently on its **structure** (in vitro, flav**a**nols > flav**o**nols, > flav**anon**es, etc.).

Conclusions of two large-scale studies (from the late 1990s):

- **No relationship** → between **incidence of cancer/mortality** and **flavonoid intake** ;
- **Inverse correlation** → between flavonoid intake and cardiovascular disease mortality. (Flavonoids protect the LDLs from oxidation,was postulated later)

Flavonoids: Enzyme Inhibitors (*in vitro*)

- **histidine decarboxylase** inhibition (by quercetin, naringenin); **elastase** inhibition;
- **hyaluronidase** inhibition (flavones, proanthocyanidins), which would maintain the intercellular ground substance in the perivascular sheath;
- non specific inhibition of **catechol-*O*-methyltransferase**, which would increase in catecholamine availability leading to a decrease in vascular fragility;
- inhibition of **cAMP phosphodiesterase** partly might explain → anti-platelet aggregation activity;
- inhibition of **aldose reductase** — known to be involved in cataract pathogenesis—
  (by quercitrin, methoxyflavones , in rodents, per os);
- inhibition of **protein-kinase** (by luteolin)
- Several flavonoids—cirsiliol, hypolaetin—are potent **inhibitors of 5-lipoxygenase**, consequently are inhibitors of the production of the leukotrienes mediating inflammation and allergic reactions. Several flavonoids (luteolin, apigenin, chrysin) **inhibit cyclooxygenase** and **platelet aggregation**. → These properties may explain anti-inflammatory and antiallergic properties
- **Flavonoids: stimulants of enzymatic activity (**rare):

  Proline hydroxylase stimulation: would favor the formation of cross-links between collagen fibers, reinforce their strength and stability, and prevent their denaturation. This **protective activity on collagen** seems to be due mainly to **flavanol oligomers** (proanthocyanidins).

  ! **Superoxide anion radica**l appears to be involved in the **non-enzymatic proteolysis of collagen**. In vitro, **anthocyanins inhibit** this degradation process.

Flavonoids: Other Biological Properties

- Flavonoids are often promoted to be **anti-inflammatory agents**—which is compatible with what is known of their (in vitro) interactions with polymorphonuclear leucocytes, thrombocytes, or the metabolism of arachidonic acid.

- They can be **antiallergic,**
- **hepatoprotective (**e.g., isobutrin, hispidulin, flavanolignans), or
- **antispasmodic** (flavonoids of thyme and other Lamiaceae)
- they can **decrease blood cholesterol levels**, be **diuretic**,
- **antibacterial**, or **antiviral** in vitro (non-glycosidic 3-hydroxy-/3-methoxyflavones).

- **Anticancer** and **tumor growth inhibitory effects** *in vitro*:
  - they interact with the enzymes of the metabolism of xenobiotic compounds;
  - they have effects against initiation, or promotion, or both; or
  - they are cytostatic or even cytotoxic.
- Most flavonoids are **antimutagenic** *in vitro*; in contrast, some flavonols are **mutagenic** on the same models.

- The extrapolation of all of these data requires the greatest caution:
  - generally low **bioavailability** of flavonoids in humans ,
  - frequent lack of correlation between in vitro and in vivo effects
  - the results are obtained with glycosides being probably hydrolyzed in the digestive tract
  - lack of structure-activity relationships, despite of intesive pharmacological research
  - With a few exceptions, no relevant study has shown any clinical benefit whatsoever.

CHIEF FLAVONOIDS ON THE MARKET I.

Citrus (Rutaceae)

- Trees of oriental origin, of which *many species, varieties and hybrids* are cultivated for their fruits and their edible endocarp.
- Widely used for their essential oils, pectins and flavonoids in the fruit pericarp.

Citroflavonoids

- **mainly flavanone glycosides** (hesperidin or hesperetin 7-O-rutinoside, neohesperidin, naringin, eriodyctin, eriocitrin).
- Structurally, these glycosides comprise
  - two rhamnoglycosides that differ by their linkage type—**rutinose** and **hesperidose**, 1 —>6 and 1 —>2, respectively—and
  - 4',5,7- **trisubstituted** aglycones (naringenin, isosakuranetin) or
  - 3' ,4' ,5 ,7-**tetrasubstituted** aglycones (eriodictyol, hesperetin) to which
  - they are bonded through their hydroxyl group at C-7.
- The pericarps also contain **flavone glycosides** (diosmin).
- Neohesperidin and naringin are found in **bitter orange**, hesperidin (0.12-0.25 g/kg) in **sweet orange**, **grapefruit** is rich in naringin (up to 0.4 g/kg).
- Citroflavonoids are **extracted** from pericarps and pulps **with water**, and isolated using different procedures (as calcium and magnesium derivatives, by adsorption onto XAD resin).

CHIEF FLAVONOIDS ON THE MARKET II.

- Currently, the pharmaceutical industry uses:
  - **a mixture of total citroflavonoids**, sometimes titrated for one particular flavonoid;
  - **glycosides of pure flavanones:** hesperidin, naringin;
  - **semisynthetic derivatives** such as hesperidin methyl chalcone (the opening of the pyran heterocycle markedly increases solubility);
  - a flavone glycoside obtained by semisynthesis, **diosmin**.

- All of these flavonoids are **used pure** (diosmin, naringin) **or in combinations** (with one another and/or with ascorbic acid, aesculetin, ruscosides, methylaesculetin, and more).
- Accepted **indications** for preparations containing **high doses of citroflavonoids** (especially diosmin, 0.3-0.6 g/unit dose):
  - to improve the symptoms of **venous and lymphatic vessel insufficiency**,
  - for the adjunctive treatment of the **functional signs of capillary fragility**, and
  - to treat the functional symptoms of the **acute attack of piles** (1.2-1.8 g/day).
- The efficacy of high doses is significantly, although only slightly greater than that of a placebo, even though, in venous and lymphatic vessel insufficiency, 50% of the patients are relieved by a placebo... and by adopting elementary elements of a healthy lifestyle.
- **Lower doses** are proposed or used for the same indications.

Rutin: quercetin 3-O-rutinoside, Industrial sources

JAPANESE PAGODA TREE, ***Sophora japonica*** L., Fabaceae,

- a **tall tree** from central and northern **China,**
- **cultivated in temperate climates** as an ornamental species
- **flower buds**: **15 to 20% rutin**

BUCKWHEAT, ***Fagopyrum esculentum*** Moench., ***F. tataricurm*** (L.) Gaertn., Polygonaceae

- an **annual pseudocereal** originated in **China**
- **cultivated in Europe** for its **edible starch-containing akenes**
- **leaves: 5-8% rutin**, in improved varieties

Other sources.

- the leaves of *Eucalyptus macrorrhyncha* F. Muell., (Myrtaceae)
- the fruits of Brazilian Caesalpiniaceae of the genus Dimorphandra.

Rutin extraction

- *S. japonica* flower buds: extraction by boiling water and crystallization upon cooling, recrystallization from ethanol.
- *Fagopyrum*: the presence of leaf pigments and photosensitizing substances (fagopyrins) complicate the extraction.

Rutin, Uses

Rutin, alone or in combination (with aesculin, citroflavonoids, or ascorbic acid) is promoted

- for the symptoms of **venous and lymphatic vessel insufficiency**,
- for the symptomatic treatment of the **functional signs of capillary fragility**,
- to treat the functional symptoms of the **acute attack of piles**, and
- for **loss of visual acuity** and **alterations of the field of vision** presumably of vascular origin.

More soluble derivatives:

- morpholinylethylrutin (ethoxazorutin, INN)
- 3’ ,4’ ,7 -tris-(hydroxy-ethyl)-rutin (troxerutin, INN)
- sodium rutosylpropylsulfonate
- Their uses are identical to those of rutin.

Rutin and its derivatives

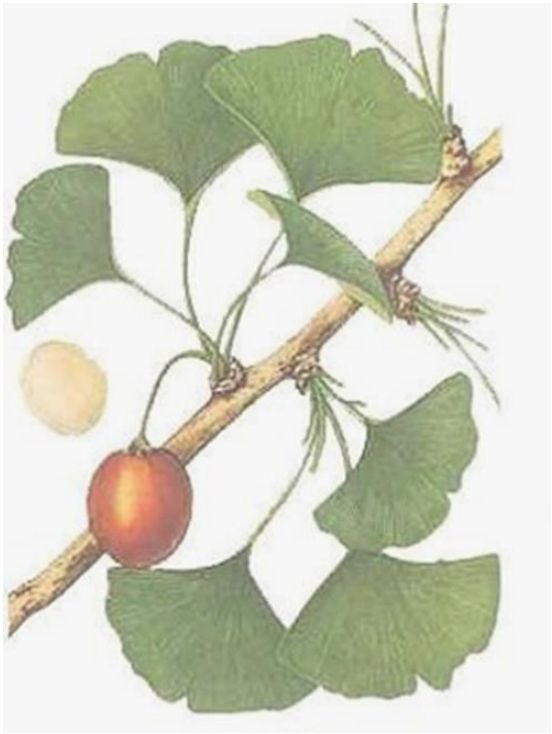
They are sometimes **combined with alkaloids** (e.g., vincamine) in proprietary drugs promoted for symptomatic **treatment for senile cerebral insufficiency**.

DRUGS FOR WHICH PART OF THE ACTIVITY MAY BE DUE TO FLAVONOIDS

MAIDENHAIR TREE, *Ginkgo biloba* L., Ginkgoaceae

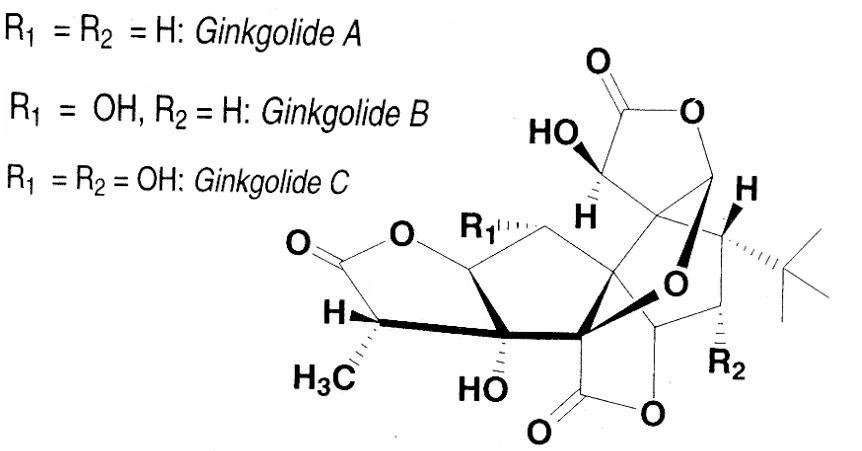
It is a dioecious species with deciduous leaves which originated in the Orient, the only survivor of an order widely represented until the end of the tertiary era.

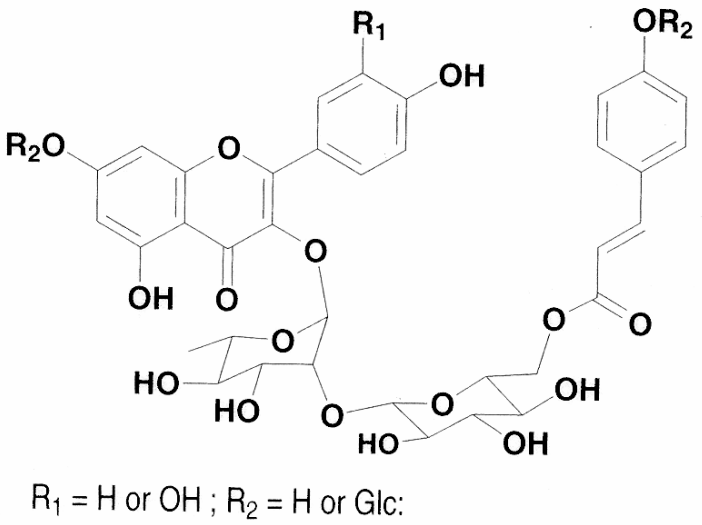
- It is characterized by
- specific reproductive organs
- a **"fruit"** with an **unpleasant odor** (in reality a fertilized ovule with pulpous aril).
- **Cultivated**
  - in Korea, the southeast of France, and the USA (South Carolina)
  - to supply the pharmaceutical market with leaves.
- **Leaves**: are commonly **bilobate**, can be almost entire or very divided.
- The petiole contains two bundles of conducting tissue which divide by dichotomy into the **blade**, giving it a very **characteristic striated look**.



Ginkgo folium，Chemical Composition

- sterols, aliphatic alcohols and ketones, 2-hexenal, organic acids,
- cyclitols, mono- and polysaccharides,
- **flavonoids** (0.5- **1%**),
  - **flavonol *0*- glucosides** (~20): quercetin and kaempferol 3-*0*-rhamnosides and 3-*0*-rutinosides, their 4- coumaric esters at C-6”’ (characteristicly with a 1”’-> 2” bond between sugars)
  - flavan-3-ols, proanthocyanidins,
  - **biflavonoids**: 3’—> 8” biflavones: eg, **amentoflavone**, **bilobetol**
- **terpene lactones**
  - **ginkgo diterpenes: ginkgolides** A, B, C, J **(**up to **0.5%)**: very specific **hexacyclic structures** with a spiro-[4,4]-nonanic sequence, a tert-butyl group, and three lactone rings.
  - sesquiterpenes : **bilobalide**





Ginkgo biloba

Fertilized ovules

- medium-chain fatty acids (C4-C8) with a **foul smell**
- fleshy part: alkenyiphenols, oxidizable to quinones forming adducts with proteins and inducing **cutaneous allergies**.
- central almond: 4'-O-methylpyridoxine (= **ginkgotoxin**), may be toxic .

Ginkgo folium, Pharmacological Activity

- **Ginkgolide B** is **an inhibitor of** the platelet activating factor (= **PAF**), a phospholipid intercellular mediator secreted by platelets, leucocytes, macrophages, and vascular endothelial cells.
- **PAF involved processes**: platelet aggregation, thrombosis, inflammatory reaction, allergy, and bronchoconstriction (this explains the trials conducted in the late 1 990s, particularly for the treatment of asthma).
- This **anti-PAF activity** and the **activities of flavonoids**, particularly **as free radical scavengers**, may explain the numerous properties of ginkgo extract observed *in animals.*

Ginkgo extract is said to be

- a vasoregulating agent (an arterial vasodilator and a venous vasoconstrictor able to decrease capillary fragility),
- an inhibitor of cyclo-oxygenase and lipoxygenase,
- an inhibitor of platelet and erythrocyte aggregation.
- It decreases capillary hyperpermeability,
- improves tissue irrigation,
- activates cell metabolism, particularly in the cortex (by increasing glucose and oxygen uptake).
- Terpene-containing fractions: prolong the survival of hypoxic rats; they protect neurons and astrocytes from damage by transient ischemia.

Ginkgo folium, Uses

Ginkgo leaves extract: titrated to contain 24% flavonoids and 6% ginkgolides-bilobalide.

- This extract has undergone several dozen human clinical trials, especially to assess its efficacy for "cerebral insufficiency".
- „Proposed" **indications** (based on trials being not nanimously well received).
  - to correct the **symptoms of senile cerebral insufficiency**,
  - for some types of **vertigo or tinnitus** or both,
  - for some types of **loss of hearing** or **loss of visual acuity** (thought to be of ischemic origin)
  - for **retinal insufficiency** likely to be of ischemic origin.
- **Recognized indication**
  - the symptomatic treatment of the intermittent claudication due to **chronic occlusive arterial disease of the lower limbs**.
- **Side effects.** → **Oral use:** rare and minor (headaches, digestive symptoms).
  Parenteral administration: serious accidents.
- **Combinations** (e.g., with heptaminol and trihydroxy**ethylrutin** [orally], or butoforme [topically]) are indicated to treat
  - the symptoms related to **venous and lymphatic vessel insufficiency**,
  - the functional symptoms of the **acute attack of piles**.

PASSION FLOWER, *Passiflora incarnata* L., Passifloraceae

- The **dried aerial parts** of the passion flower are listed in the 10th edition of the French Pharmacopoeia.
- Since the constituents responsible for the sedative activity of the drug are not known with certainty, the preparations in use systematically contain the constituents as a group.

- Grows wild in the bushes of the south of the USA and Mexico.
- **a creeping plant** with alternate leaves with a finely serrate margin and long petioles.
- **Axillary tendrils** allow the plant to climb onto supports.
- The **large solitary flowers** (5-9 cm in diameter):
- five thick sepals, white on the underside,
- **five white petals**, a double crown of petaloid appendices, crimson red on the edge,
- stamens with **orangy anthers**,
- an **unilocular ovary** with **three stigmas**.
- The **fruit** is **ovoid** and **resembles a** small, flattened, greenish to brownish **apple** with yellow flesh.


Пассифлора инкарнатная

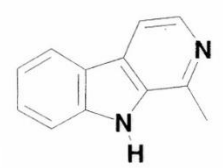
Passiflorae herba

- **Passiflorae herba:** fragments of lignified, hollow, grayish stem, with slender and smooth tendrils. The **leaf** has a long petiole and is **deeply divided into three lobes** with the middle one being most developed.

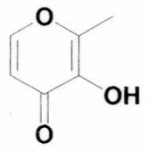
- The drug may be falsified with the stems and leaves of
  - ***P. edulis*** Sims. (**leaf blade is dentate)**, and
  - ***P. carulea*** L., (**pentolobate leave).**

Passiflorae herba, Chemical Composition

- Phenolic acids, coumarins, phytosterols,
- I mL/kg essential oil, cyanogenic glycosides (gynocardin),
- 0.05% **malto**l (or 2-methyl-3-hydroxypyrone, maybe an artefact)
- traces of indole alkaloids: **harman**, harmol, harmine
- **flavonoids** (**up to 2.5%**)



Harmane

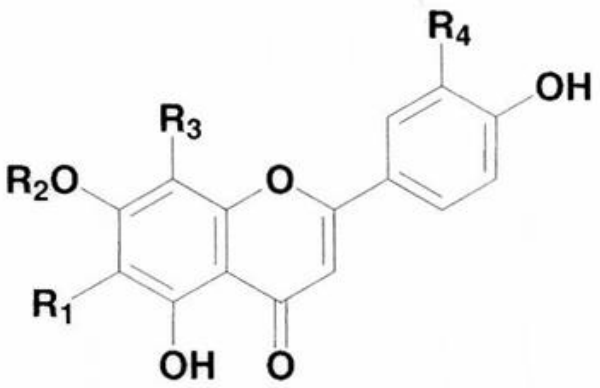


Maltol

The major ones are

- **flavone di-C glycosides** (shaftoside and isoshaftoside = **apigenin *C*-glucosyl-*C*-arabinosides** = 8,6 and 6,8 isomers) and
- **2”-O-glycosides of** isovitexin and iso—orientin (i.e., apigenin and luteolin C-sophorosides).

The qualitative composition can vary widely. In general**, isovitexin** and its glucosyl derivative are dominant.



	R_1	R_2	R_3	R_4
vitexin	*H*	*H*	*glc*	*H*
isovitexin	*glc*	*H*	*H*	*H*
orientin	*H*	*H*	*glc*	*OH*
iso-orientin	*glc*	*H*	*H*	*OH*
saponarin	*glc*	*glc*	*H*	*H*
schaftoside	*glc*	*H*	*ara*	*H*
isoshaftoside	*ara*	*H*	*glc*	*H*
vicenin-2	*glc*	*H*	*glc*	*H*

Passiflorae herba

Pharmacological Activity

- Tradition attributes to the passion flower **sedative, antispasmodic**, and "**tranquilizing"** properties partially confirmed by animal experiments (IP route).
- A body of **observations** supports the usefulness of **"neurosedative**" preparations of this drug.
- Other experiments confirm the **activity** of the passion flower extract **on the CNS of the rat** and
- point to the existence of two active compounds not yet identified.

Uses

- **The drug** (in infusions), **its galenicals** (powders, extract, tincture, nebulisate) and the **phytopharmaceuticals** containing it are **traditionally used** by the **oral route**
  - to treat **abnormalities of the cardiac rhythm** in the adult (normal heart)
  - to treat the **symptoms of nervousness** in adults and children, particularly **minor sleeplessness**
- Frequently **combined with hawthorn**, which has the same indication; **with valerian** and **other sedative plants**.
- The drug is reputed to be **harmless**.
- **Commission E** indications: **mild sleeping difficulties** and **gastrointestinal signs of nervous origin.**

THYME, *Thymus vulgaris* L., *T. zygis* L., Lamiaceae

- This Mediterranean Lamiaceae is an antibacterial and spasmolytic, but above all it is an “essential oil-containing drug”.
- **Spasmolytic activities** are recognized for the **aqueous preparations of the flower and flowering tops.**
- It has been shown that
- the concentration of the volatile essential oil phenols in these preparations is insufficient to account for the spasmolytic activity,
- which is due to **polymethoxyflavones and di-, tri-, and tetramethoxylated flavones**, **all substituted at C-6**.

ROMAN CAMOMILE, *Chamaemelum nobile* (L.) All., Asteraceae

Roman camomile flower = the dried flower-heads of the cultivated double variety of *Chamaemelum nobile* (L.) All. [...]" (Eur. Ph., 3rd Ed.). The activity attributed to this drug is in part due to flavonoids.

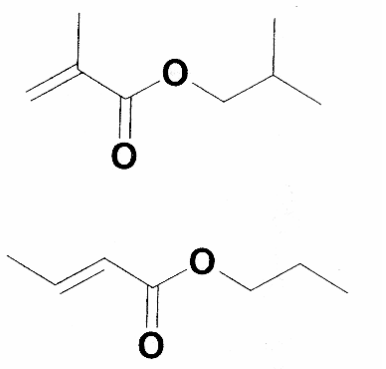
- Roman camomile is a **perennial plant** with
- ramified stems and pinnatisect pubescent leaves of a whitish-green color.
- The **capitulums** of the cultivated variety have a diameter ranging **from 8 to 20 mm**. They practically only have **ligulate flowers**, white, sterile, and inserted onto a solid receptacle which bears, between the flowers, elongated and translucent paleas.
- The **capitulum involucre** is reduced to two to three rows of tight and imbricate bracts which are scarious on the edges.
- The ligules are lanceolate with three veins and five teeth.
- The pharmaceutical market is supplied by culture (in France and Belgium, among other countries).

ROMAN CAMOMILE, *Chamaemelum nobile* (L.) All., Asteraceae

Chemical Composition

- **sesquiterpene lactones** (0.6%), specifically **germacranolides**: nobilin, 3-epinobilin, and close derivatives((with a bitter taste)
- **essential oil** (4 to 15 mL/kg; >7 mL/kg Ph. Eur, odorous)
  - over 85% **mono- and bifunctional esters of aliphatic acids and alcohols of low molecular weight** (i.e., C4, C5, or C6) themselves arising from leucine, isoleucine, or valine metabolism: angelates, tiglates, methylacrylates, crotonoates, isobutanol, 3-methylbutan-1-ol, or 2-methylbutan-1-ol butyrates, and so forth.
  - monoterpenes; azulenes, only in trace amounts
- phenolic acids, coumarins,
- **flavonoids**: apigenin and luteolin glucosides.

Esters from Chamaemelum *essential oil (examples)*



Pharmacological Activity

- **Anti-inflammatory, antispasmodic**
- may be linked to **apigenin, luteolin, and their glycosides**, whose activity has been established in the mouse.

ROMAN CAMOMILE, *Chamaemelum nobile* (L.) All., Asteraceae

Chemical Composition

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  - monoterpenes; azulenes, only in trace amounts
- phenolic acids, coumarins,
- **flavonoids**: apigenin and luteolin glucosides.

Tests

- **macro- and microscopic examination**
- **TLC of the flavonoid fraction of a methanol extract** (spraying with aminoethanol diphenylborate).
- The proportion of capitulums of diameter smaller than 8 mm must be not more than 3%.
- Essential oil level >7 mL/kg.

Roman camomile-based medicines, Uses

- Orally
  - for the symptomatic treatment of **gastrointestinal disorders** such as epigastric bloating, impaired digestion, eructations, flatulence
  - as an adjunct in the treatment of the **painful component of functional digestive symptoms.**
- Topically
  - as an emollient and itch-relieving adjunct in the treatment of **skin disorders**
  - as a trophic protective agent for **cracks, abrasions, frostbites, chaps, and insect bites**;
  - for **eye irritation or discomfort** of various etiologies (for example eye strain, seawater or swimming pool water, or smoky atmospheres);
  - an antalgic **in diseases of the oral cavity, oropharynx,** or both (collutoria, lozenges);
  - a **mouthwash** for oral hygiene.
- German Commission E indications: **dyspepsia** and **inflammation of the mouth**.
- Ingredient of color-lightening shampoos.

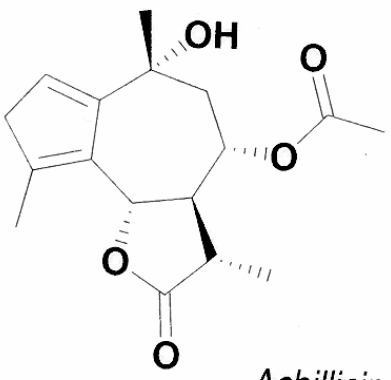
YARROW, *Achillea millefolium* L., Asteraceae

- **Folk medicine** attributes to the flowering tops of yarrow **astringent**, **antispasmodic**, and **healing properties**.
- The drug contains not less than **2 mL/kg essential oil** and not less than **0.02% proazulenes** (Eur. Ph., 3rd Ed., add. 1999).

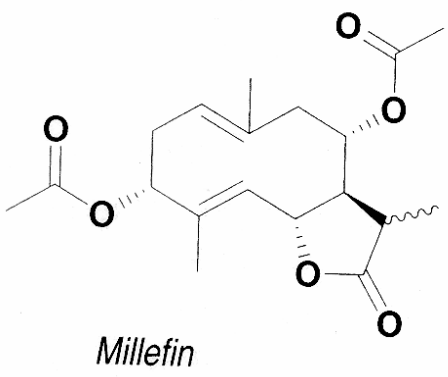
- *A.millefolium*: a cosmopolitan species with sessile, deeply divided, and pubescent leaves.
- Flowers gathered in capitulums, themselves grouped in dense corymbs.
- A polymorphous species.
- **Hexaploid subspecies** (western Europe) :
  - ssp. ***millefolium***,
  - ssp. ***sudetica*** (Opiz) Weiss
  - ssp. ***ceretanum*** Sennen (a hardy form, *A. monticola* Martin-Donos considered by different authors as a subspecies)

Achillea millefolium, Chemical Composition

- **Sesquiterpene lactones:** achillicine [= 8α-acetoxy- 10-epiartabsin], achillin, achillifolin, millefin, leukodin, dihydroparthenolide, and balchanolide
- Polyalkynes,
- **Essential oil (**2-10 mL/kg): azulene-containing
- **Flavonoids: luteolin**, **apigenin**, and their 7-*O*-glucosides; **flavones** and 6-methoxylated or di- and trimethylated **flavonols** (pectolinarigenin, 3-methylbetuletol, 3,6,4'-methyl- quercetagetin).
- Occurece of proazulenes yielding azulenes upon steam distillation:
  - only in tetraploid species and populations of *A. millefolium* in the broad sense
  - *A. collina* of central Europe is rich in proazulenes: achillicine and other derivatives of 10-epiartabsin (8α -angeloyloxy- and 8α -tigloyloxy-).
  - The essential oil of hexaploids only contains traces of azulenes.



Achillicin



Millefin

Achillea millefolium, Pharmacological Activity and Uses

- **Anti-inflammatory** and **antispasmodic** properties are attributed, without proof, to the drug.
- These properties **can be due to the flavonoids** being abundant in all subspecies (in contrast to azulenes).

Yarrow is **traditionally used**

- **orally**
  - for the symptomatic treatment of **gastrointestinal disorders**: epigastric bloating, impaired digestion, eructations, flatulence,
  - as an adjunct in the treatment of the painful component of **functional dyspepsia.**
- **topically**,
  - as an emollient and itch-relieving adjunct in the treatment of skin disorders
  - as a trophic protective agent.

Commission E indications:

- internally, for **gastrointestinal distress** (dyspepsia, cramp-type abdominal pains, lack of appetite);
- topically (sits bath) for **pelvic congestion pains** in women.

The use of yarrow is contraindicated in subjects allergic to Asteraceae because of the sesquiterpenic lactones.

FIELD HORSETAIL, Equiseti Herba, *Equisetum arvense* L., Equisetaceae

- Ph. Eur.: "Whole or cut, dried **sterile aerial parts** of *E. arvense* L."

Common in Europe, fond of moist and even swampy soils, prefers siliceous clay.

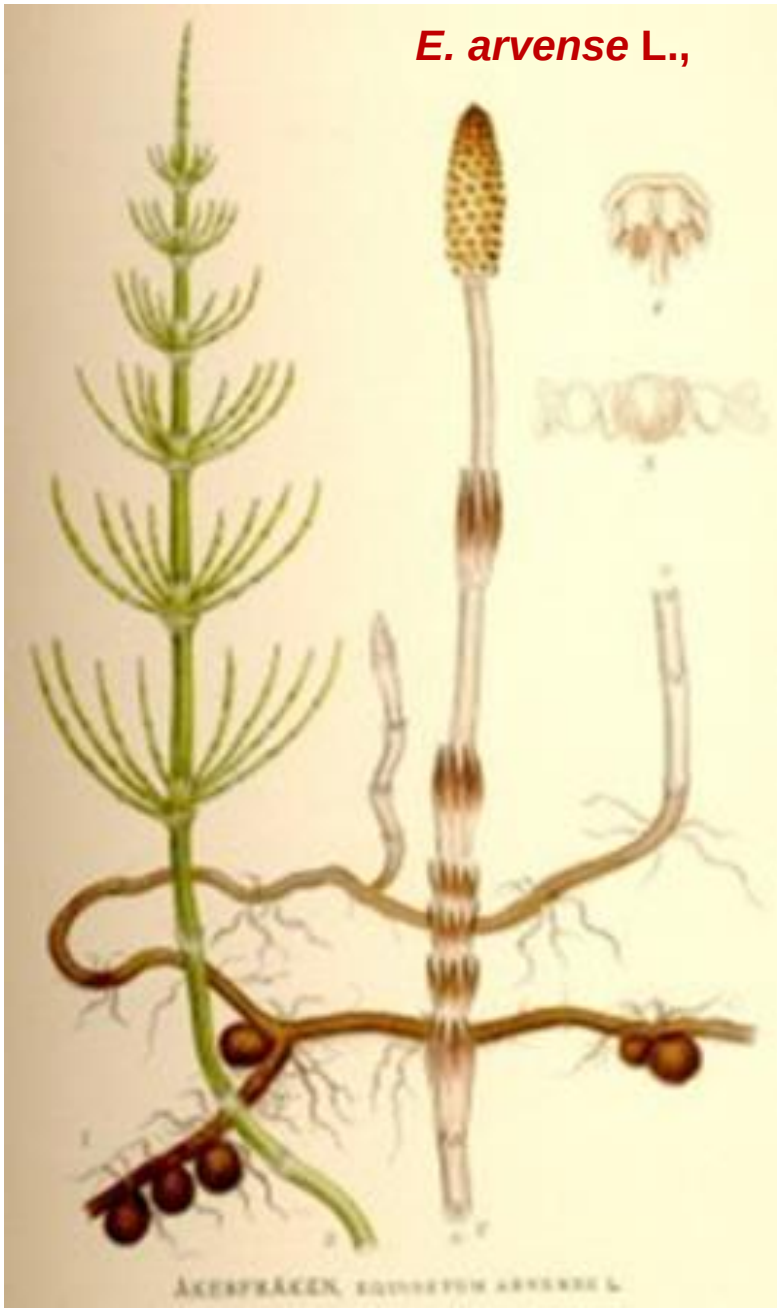
- Characterized by two types of stems: **fertile stems** appearing in the spring, and **sterile stems** following later.
- The fertile stems have **no chlorophyll** and support a **sporangium-bearing oblong spike**.
- **The sterile branches** (0.2-0.8 m) have hollow stems articulated at the nodes and are ribbed with 6-12 shallow grooves, and secondary branches with a quadrangular cross-section.
- **At the nodes** are inserted **whorled leaves of very small size**; shaped like black-tipped teeth and fused, they **form a sheath around the stem**.

E. palustre L., a species reputed **toxic** (to humans?) and possible adulterant,

only has one type of branch with stems with six to eight deep grooves,

with secondary branches with a cross-section with four or five angles, whorled in groups of eight, and hollow.

- The occurrence of alkaloids does not seem to be an absolutely distinctive characteristic.



***E. arvense* L.,**

E. palustre L.,

Equisetum stem, Equiseti herba

Chemical Composition

- **Silicon** (5-10 %): found as opaline concretions deposited onto epiderms, peripheral collenchymas, and the endoderm of stems and branches (insoluble silicon , hence the roughness of these organs).
- Sterols;
- Ascorbic acid; and **phenolic acids**: cinnamic acids, dicaffeoyltartaric and 5-O-caffeoylshikimic acids;
- **Flavonoids** (sterile stems):
  - In total, nearly 20 flavonoids in two chemotypes: Asian and American, and European.
  - **Asian and American**: **flavone 5-*O*-glucosides**, particularly luteolin 5-*O*-glucoside and its malonic ester at C-6" (50-60% of the total flavonoids). The other, European, is devoid of it.
  - **Both chemotypes:** a large amount of quercetin 3-*O*-malonylglucoside (ester at C-6") the major compound {**30-50%**} **in the European chemotype**), quercetin 3-*O*-glucoside, and other **flavonol glycosides** .
  - In the North Atlantic region (Scandinavia, Scotland): intermediate composition.

Equisetum stem, Equiseti herba

Tests

- TLC to show the flavonoids contained in an ethanol extract (visualization with aminoethanol diphenylborate under UV light).
- TLC detection of substitution by *E. palustre*:
  - carried out on the residue of acidic (H_2S0_4) extraction followed by purification by re-extraction (diethyl ether, aqueous ammonia);
  - visualization with the iodoplatinate reagent must not reveal any spots.
- **HPLC analysis** allows the detection of contamination of the drug by *E. palustre*, which contains a specific flavonoid, *kaempferol 3-O-rutinosyl-7-O-glycoside*. *E. telmateia* Ehrh. is characterized by *acetylated kaempferol glycoside**s***.

Pharmacological Activity

- Based on tradition, and on older animal experiments, the field horsetail was considered diuretic.
- More recent experimental data only show, at best, a slight **increase in water excretion**.

Toxicity

- **Horsetails**, especially ***E. palustre***, can cause intoxications of **herbivorous animals**. Intoxication in **horses** is especially serious and has every aspect of an **acute vitamin deficiency** (loss of motor coordination).

Equisetum stem, Equiseti herba, Uses

- Horsetail-hased phytopharmaceuticals recognized indications:
  - to enhance urinary and digestive elimination functions (“traditional uses”);
  - to enhance the renal excretion of water,
  - as an adjunct in weight loss programs.
- Phytotherapists frequently recommend the horsetail for bone fragility, cramps, and more.
- Contraindication: edema due to impaired cardiac or renal function
- **Commission E monograph** lists, among the uses for this “mild diuretic”,
  - post- traumatic and static edema,
  - inflammatory or bacterial disorders of the urinary tract, and kidney stones.
  - **Externally**: used as an adjunct in the treatment of **wounds that don’t heal well**.
- The horsetail and its preparations are widely used **in cosmetology (**prevention of wrinkles, vibex, and cellulitis).

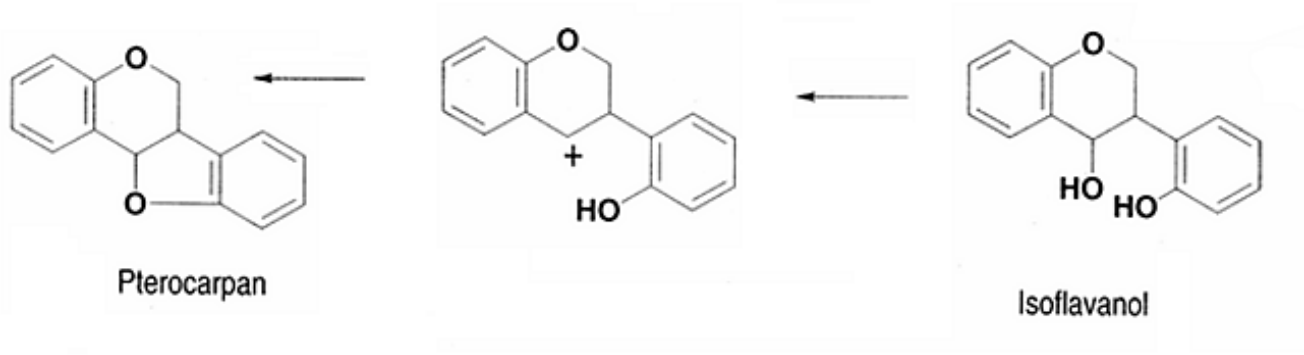
ISOFLAVONOIDS

Isoflavonoids, I

- All molecules in this group can be related to the **skeleton of 3-phenylchromane**.
- Distribution : rather limited, isoflavonoids are almost specific to the **Fabaceae.**

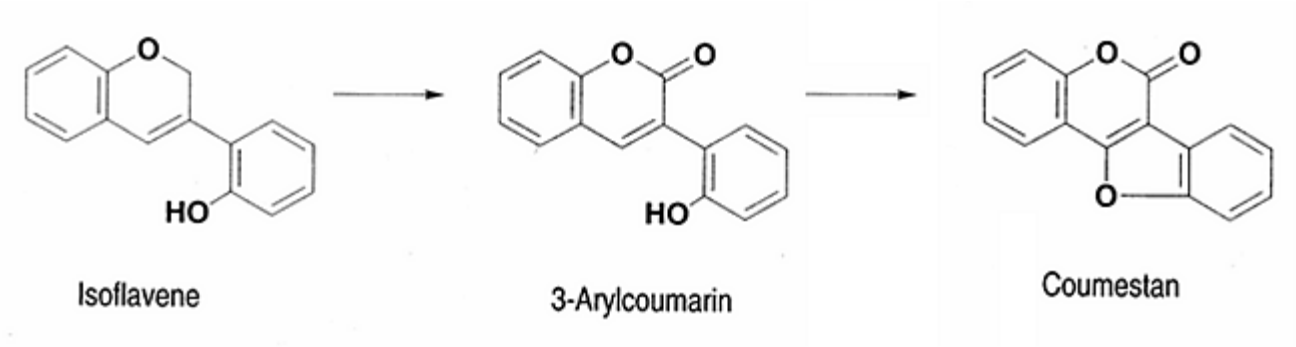
Structural diversity:

- Structural types can be differentiated by their **degree of oxidation** and by the existence of **added heterocycles**.
- High frequency of **isoprenylated derivatives**, and consequently, of **furan-, dihydrofuran-**, and **pyran-type structures**.
- The most common compounds are **isoflavones**, which occur in the free state, or as glycosides (O-glycosides mainly). Related structures include isoflavanones, isoflavenes, and isoflavanes.
- **Isoflavonoids with an additional ring**: arising from the cyclization of a 2' -hydroxylated derivative: **pterocarpans** and their derivatives (pterocarpenes and 6a-hydroxypterocarpans), and also of **coumaranochromones**.

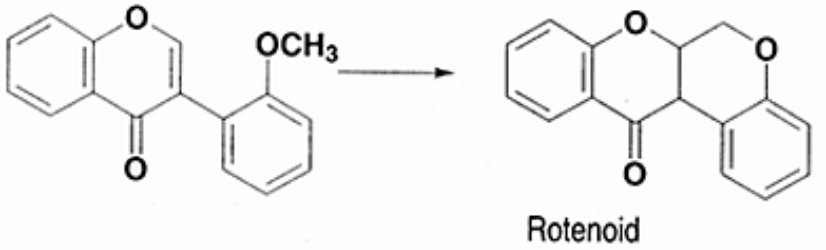


Isoflavonoids, II

- **Isoflavonoids with a coumarinic structure** resulting from the oxidation of an isoflavene: consider the **3-arylcoumarins** (of Glycyrrhiza spp.) and the cyclization products of a 2'-hydroxy-3-arylcoumarin: **coumestans** (i.e., oxidized pterocarpans).

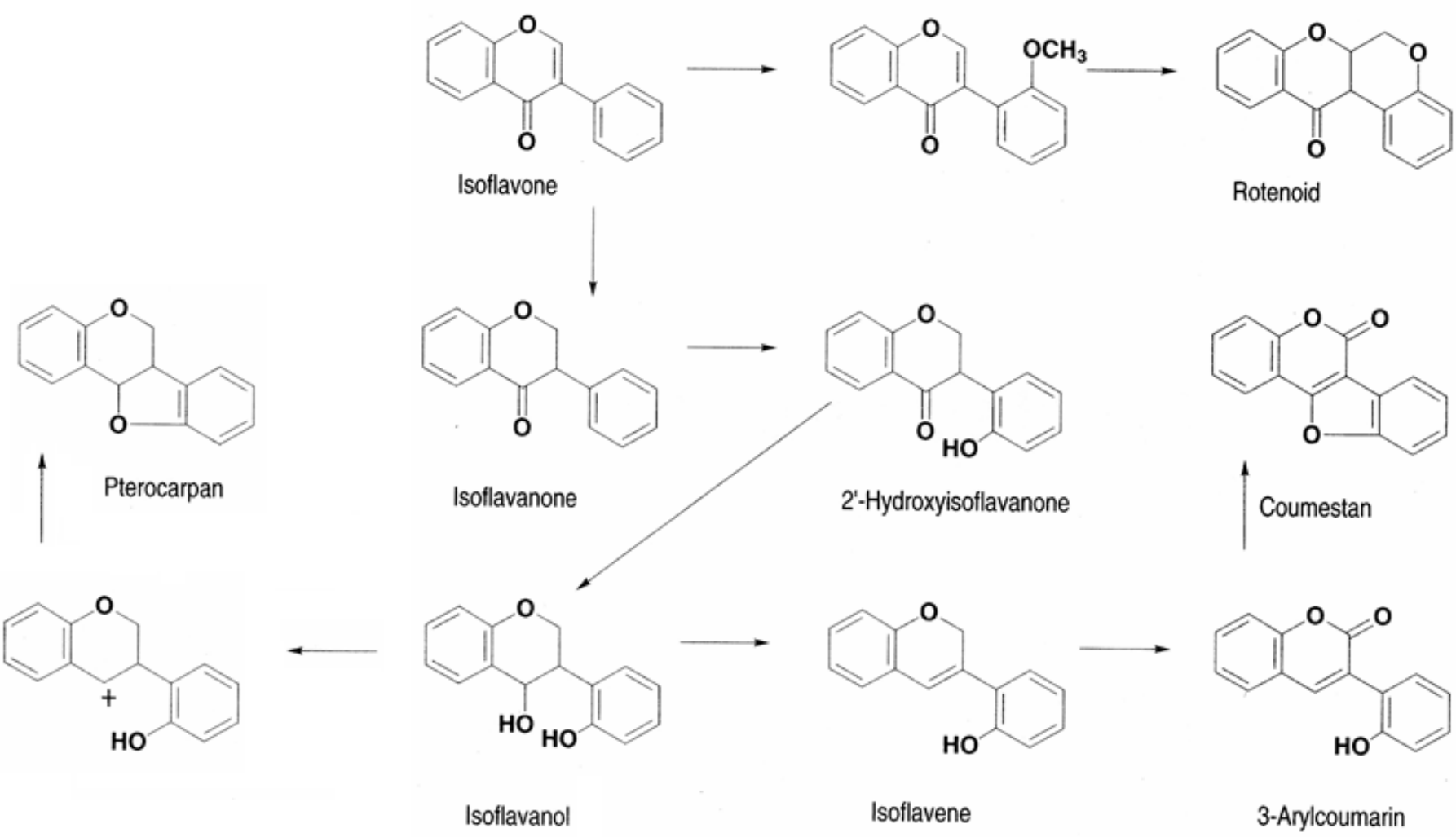


- **Polycyclic compounds with additional carbon atom**: e.g., **rotenoids** arising from the oxidative cyclization of a 2'-methoxyisoflavone.

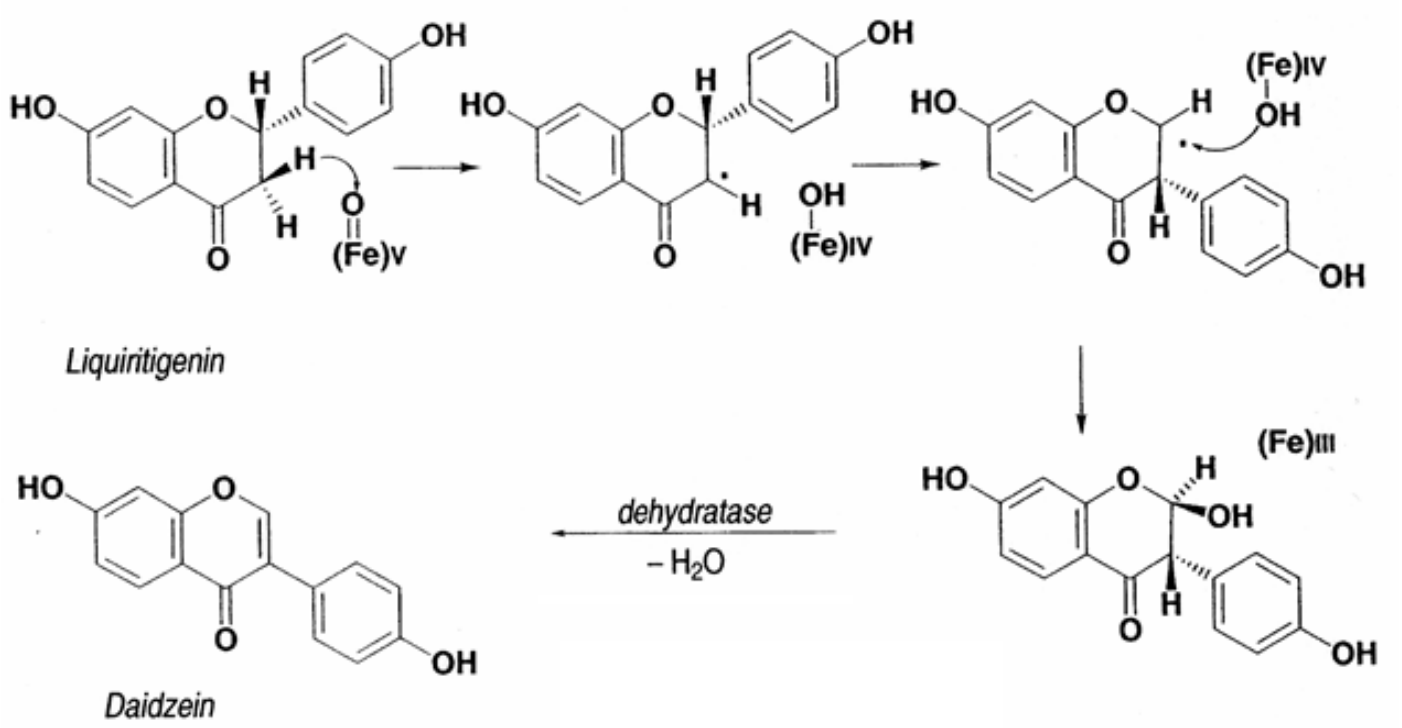


- **Benzofurans :** isoflavonoids formed (maybe?) by the loss of the C-6 of coumestan.
- Isoflavonoids can form **dimers** and **oligomers** (e.g., bisisoflavanes, isoflavane-flavanone,) as well as adducts with cinnamic acids (isoflavano-lignans).

Chief types of isoflavonoids and interconversion



BIOSYNTHESIS, LIKELY ORIGIN OF ISOFLAVONES



- The mechanism proposed consists of two steps.
- The first one is the **oxidation** of a flavanone and its **rearrangement**,
- the second one is a **dehydration,** yielding isoflavone (e.g., (2S)- liquiritigenin —> daidzein).
- The first step is catalyzed by **isoflavone-synthase** (a cytochrome P450-dependent mono-oxydase) in the presence of NADPH and oxygen and leads to a 2-hydroxyisoflavanone.
- The mechanism is thought to involve a radical, with hydroxylation taking place at the same time as **aryl migration**.

ISOFLAVONOIDS, BIOLOGICAL ACTIVITY

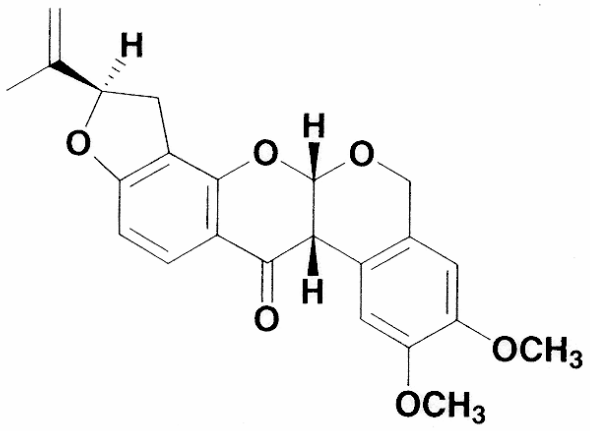
- **Phytoalexins**: may be considered as the natural defense, mostly antifungal, products of the plants that produce them.
- The pharmacological properties of isoflavonoids are seldom known.
- **Insecticide activity** of rotenoids has found an application.
- **Estrogenic properties**: isoflavonoids cause infertility in sheep that ingest massive quantities of clover; cows seem less susceptible, perhaps because of a difference in metabolism.

Estrogenic activity of isoflavonoids

- **Isoflavones in food →** possible impact on **human health**. **In the soybean**, the concentration of **daidzein** (7,4'-dihydroxyisoflavone**),** **genistein** (5,7,4'- trihydroxyisoflavone) and their glycosyl derivatives (**glucosides-** and **6" -O-malonylglucosides**) can reach 3 g/kg.
- These **isoflavones** and their intestinal metabolites (equol. demethylangolensin) **bind to estrogen receptors**, frequently with a **weak estrogenic activity**.
- As **tyrosine-kinase inhibitors** may have a role in the **transformation and cell proliferation phenomena**.
- **Soybean-based diet: decreases mammary and prostate carcinogenesis** in animal models.
- **Genistein**: an **anticarcinogen** (mammary tumors of the female rat during the neonatal period, colon microadenomas; *in vitro* result with cell culture lines).
- In humans, **soybean isoflavones**
  - **seem to have** a **preventive effect on breast-, prostate-, and colorectal cancer** (suggested by epidemological studies).
  - **decrease the symptoms of menopause** (hot flashes and others) and **reduce the risk of osteoporosis**.

ROTENOIDS

- These compounds, biogenetically related to isoflavonoids, have in common a **four-ring structure**: a **chromanochromanone**.
- The **two oxygenated cycles** are cis fused, and the biological activity is maximal for those derivatives that possess a **dihydrofuran ring**.
- The chief representative of the group is **rotenone**, the major active principle **in the roots of various tropical Fabaceae** of the genera Derris, Lonchocarpus, Milletia, Mundulea and Tephrosia.



Rotenone

ROTENOIDS

- ***Derris elliptica*** (Roxb.) Benth (Fabaceae)
  - is **a vine species** from Malaysia and Burma,
  - also introduced and cultivated in Africa.
  - The drug consists of the **root**.
  - **Derris extract**: enriched and titrated to contain about 30% rotenone.
  - **Derris powder:** with rotenoid level ranging from 3 to 10%.
- The **roots** of South American ***Lonchocarpus*** (***L. urucu*** Killip & Smith, ***L. utilis*** A.C. Smith) are used in the same forms and for the same purposes.
- **Rotenone** has **insecticidal properties**;
  - active upon contact as well as by ingestion,
  - it **inhibits mitochondrial respiration** (NADH-dependent dehydrogenase).
- **Rotenoids-containing Fabaceae** (powder, extracts, rotenone) is used
  - by phytopharmacy (treatment of house plants, and, sometimes, of vegetable gardens), and
  - for the extermination of ectoparasites of domestic animals

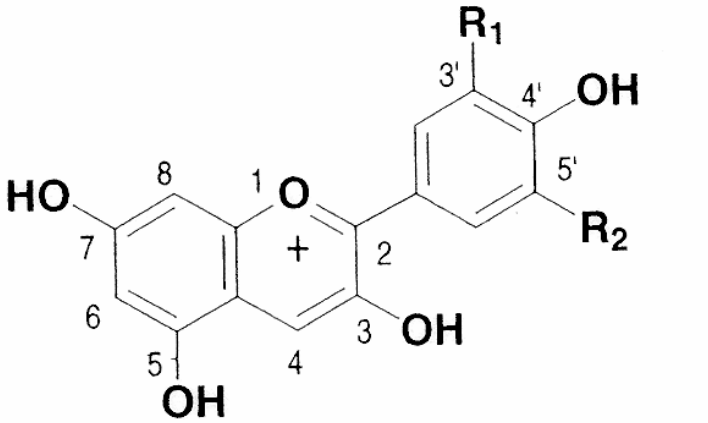
ANTHOCYANINS

Anthocyanins

- Anthocyanins = antocyan glycosides, Anthocyanidins = aglycons

Aglycons

- Anthocyanidins occur in acidic medium as cations.
- They are always hydroxylated at C-3 and, most often, penta(3,5,7,3',4') or hexasubstituted (3,5,7,3',4',5') by hydroxyl groups, or methoxyl groups, or both.
- The most common aglycones (they are virtually ubiquitous) are pelargonidin (scarlet), cyanidin (crimson), and deiphinidin (purple).



Structures of the chief anthocyanidins

$R_1 = R_2 = H$: *Pelargonidin*

$R_1 = OH$, $R_2 = H$: *Cyanidin*

$R_1 = OCH_3$, $R_2 = H$: *Peonidin*

$R_1 = R_2 = OH$: *Delphinidin*

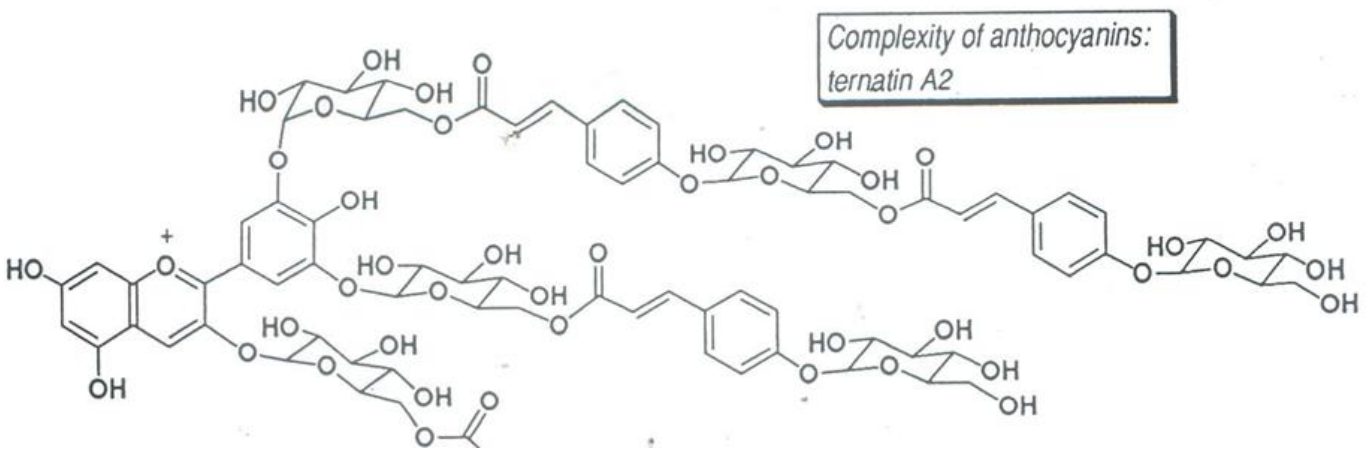
$R_1 = OCH_3$, $R_2 = OH$: *Petunidin*

$R_1 = R_2 = CH_3$: *Malvidin*

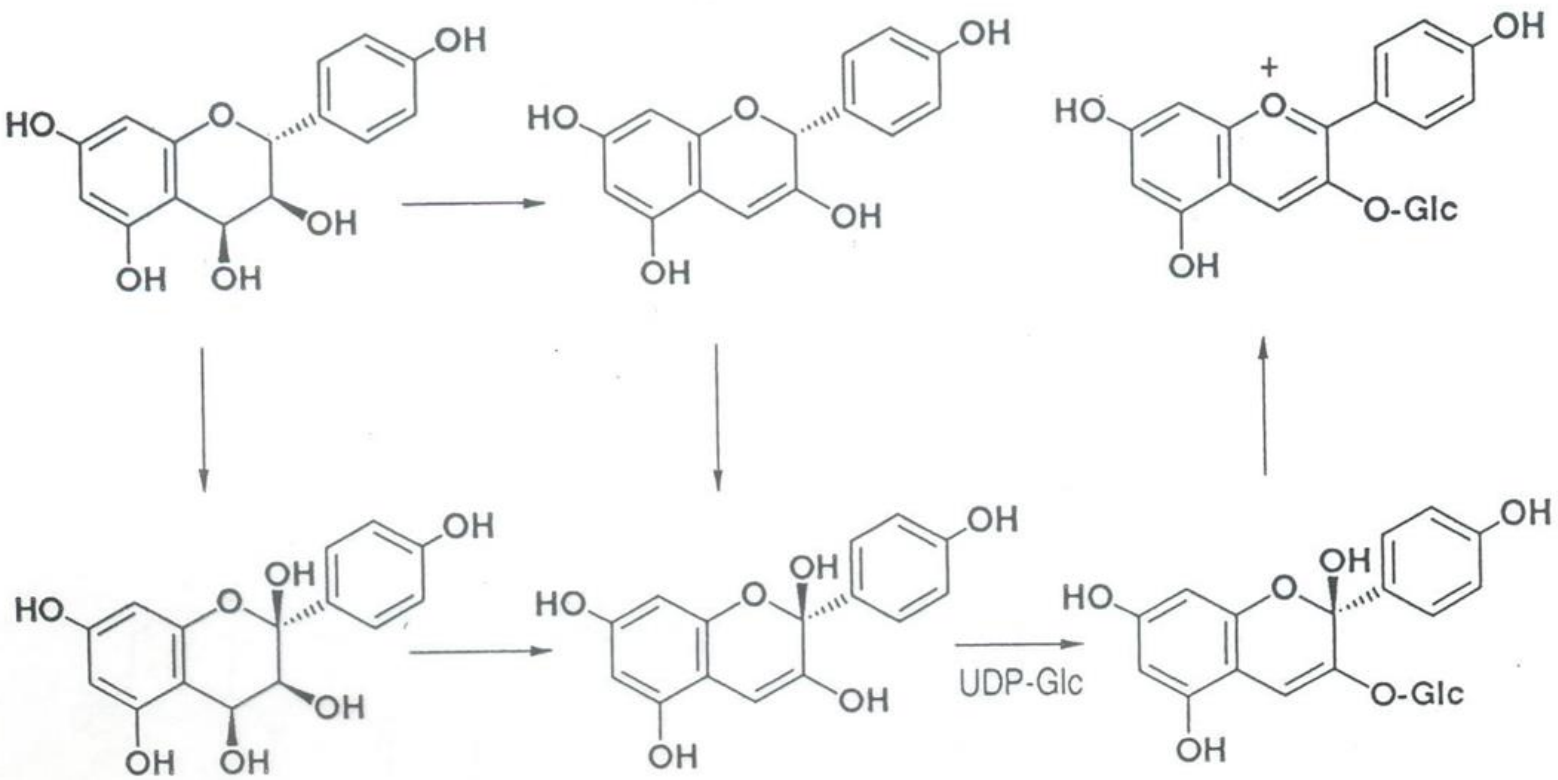
Anthocyanins

Glycosides

- At **least one hydroxyl group** at C-5, C-7, or C-4' **must remain free** to allow the formation of the colored quinonoid structures.
- (In contrast, anthocyanidins are unstable because their 3-hydroxylgroup makes the flavylium ion very reactive.)
- In fact, the **3-hydroxyl group is always linked to a sugar** (very often glucose) to form a stable and water soluble anthocyanin.
- 3- **monosides** and 3,5-**diglycosides** are the most common forms, 3,7-diglycosides and **triglycosides** (for example 3,5,3'-triglycosides) are less common.
- **Sugar residue:** can be a monosaccharide (e.g., glucoside, galactoside, rhamnoside), a disaccharide (e.g., rutinoside, xylosylglucoside), or, less often, a trisaccharide.
- Acylation: frequent, by phenylpropanoic acids (e.g., p-coumaric, caffeic, ferulic,sinapic acids) or benzoic acids (gallic acid), generally at C-6". ; Also known by dicarboxylic aliphatic acids (e.g., malonic, malic, oxalic, succinic acids).

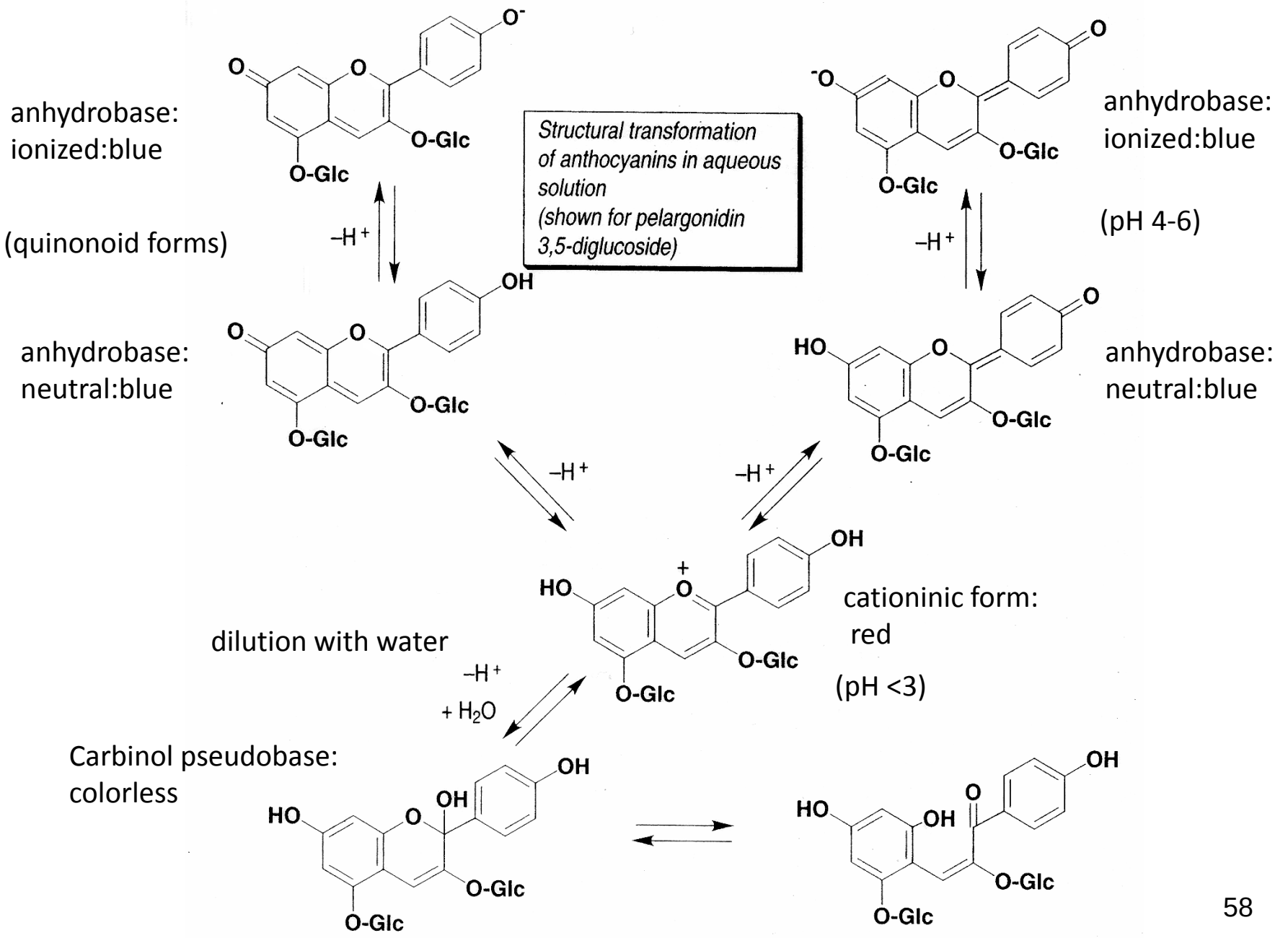


Biosynthetic Origin → general metabolism of flavonoids →precursors: 2,3- trans-dihydro-3,4-cis-dihydroxyflavonols → the diols undergo a hydroxylation (at C-2) and a double dehydration→ glucosylation (UDP-glucose) probably occurs late

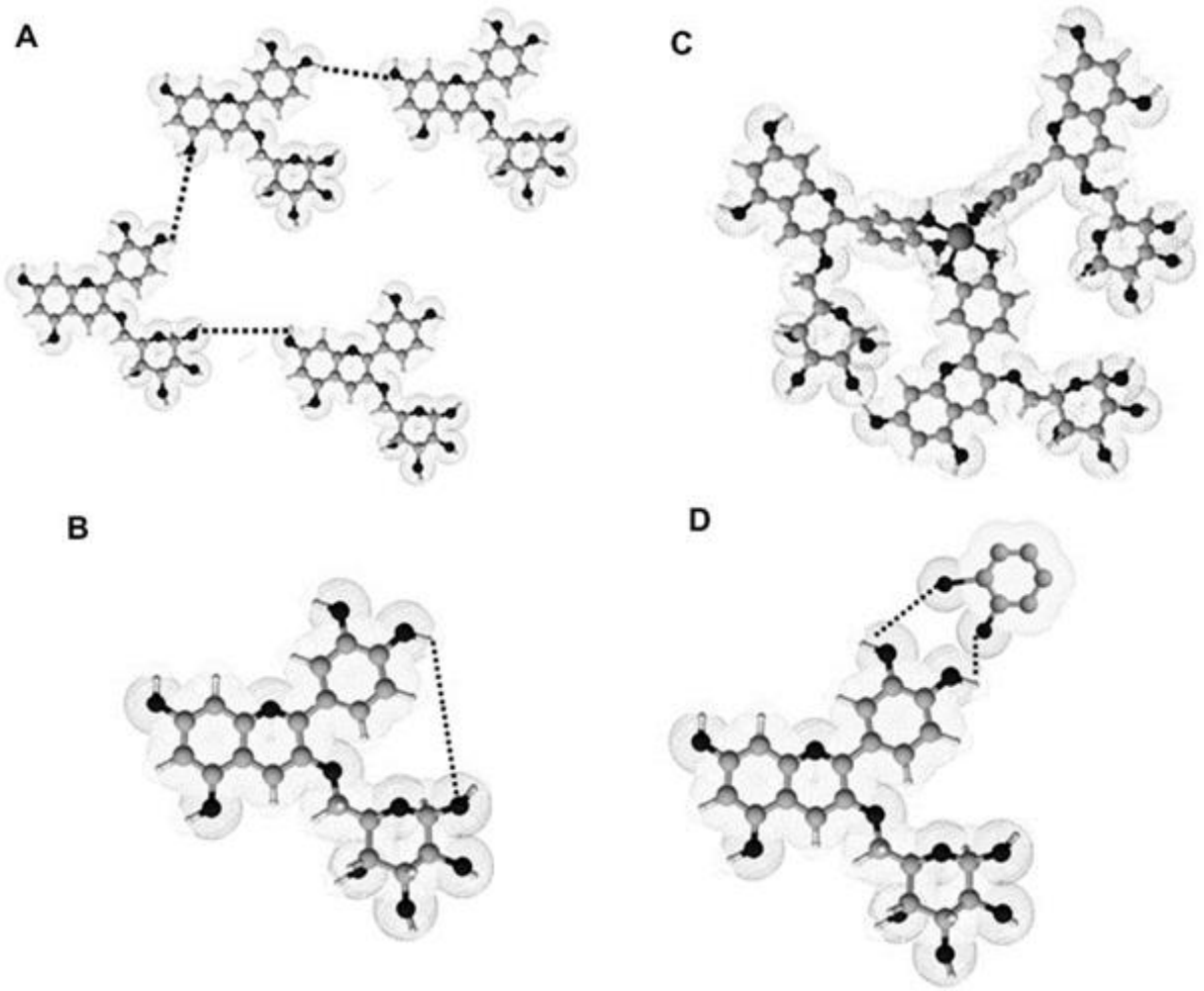


Possible biosynthetic origin of anthocyanins

Properties of 2-phenylbenzopyrylium cation: a weak diacid and a good electrophilic reagent.



Anthocyanins interaction. (A) self-association, (B) intramolecular copigmentation, (C) metal complexation, (D) intermolecular copigmentation (Castañeda-Ovando et al.).



Co-pigmentation is a phenomenon in which the pigments and other colourless organic compounds, or metallic ions, form molecular or complex associations, generating a change or an increment in the colour intensity

Some investigations suggest that the co-pigmentation of anthocyanins with other compounds (co-pigments) is the main mechanism of stabilisation of colour in plants. http://dx.doi.org/10.1016/j.foodchem.2008.09.001,

EXTRACTION AND CHARACTERIZATION

- Anthocyanins are **soluble in** water and alcohols, **insoluble in** apolar organic solvents, and
- They are **unstable in** neutral or alkaline medium.
- They are **generally extracted with an alcohol** (methanol, preferably ethanol if the product is intended for use in food) **in the presence of** a small amount (**0.1-1%**) **of hydrochloric acid**.
- To avoid esterification of the free carboxyl group of acylated anthocyanins by a diacid, and especially **to prevent** their **deacylation**, it is better to use other acids, either **weak acids** (acetic, tartaric, citric) or **volatile acids** (trifluoroacetic), or to work in a **neutral medium** (alcohol mixtures), and to work at **low temperature** (< 30 °C).
- Anthocyanin solutions are very unstable, and they can only be kept under nitrogen, at low temperature, and in the dark.
- **Industrial preparation of anthocyanin extracts**.
  - Classical procedure: extraction in aqueous medium containing sulfur dioxide, followed by acidification to regenerate the anthocyanins.
  - More recent procedures: ultrafiltration on cellulose membranes, chromatography on ion-exchange resins.
- **Separation of anthocyanins** is achieved by **chromatographic techniques** (column chromatography on polyamide supports, on polyvinylpyrrolidone supports, or on ion-exchange resins, preparative TLC on cellulose-coated plates, or semipreparative HPLC).

EXTRACTION AND CHARACTERIZATION

HPLC is the method of choice to analyze anthocyanin-containing drugs.

- The **separations** are most often carried out **on reverse phases with acidic water and alcohol gradients**, in which the cationic forms can be **detected at about 500-550 nm**.
- As for flavonoids, **diode array detectors** represent a considerable method enhancement.
- The more complex methods (**LC-MS, MS-MS**) are only available in specialized research laboratories.

As a general rule, **anthocyanin quantitation** is performed by spectrophotometry.

- At the wavelengths of maximum absorption of these compounds, interferences are exceptional:
- quantitation can be done directly on an acidic solution in alcohol (cationic form) or on an acidified juice.
- To prevent anthocyanin **self-association**, which would result in a positive deviation from the Beer-Lambert Law, **dilute solutions must be used**.

Quantitative estimates of the constituents of an anthocyanin mixture are now obtained directly by HPLC.

ANTHOCYANINS

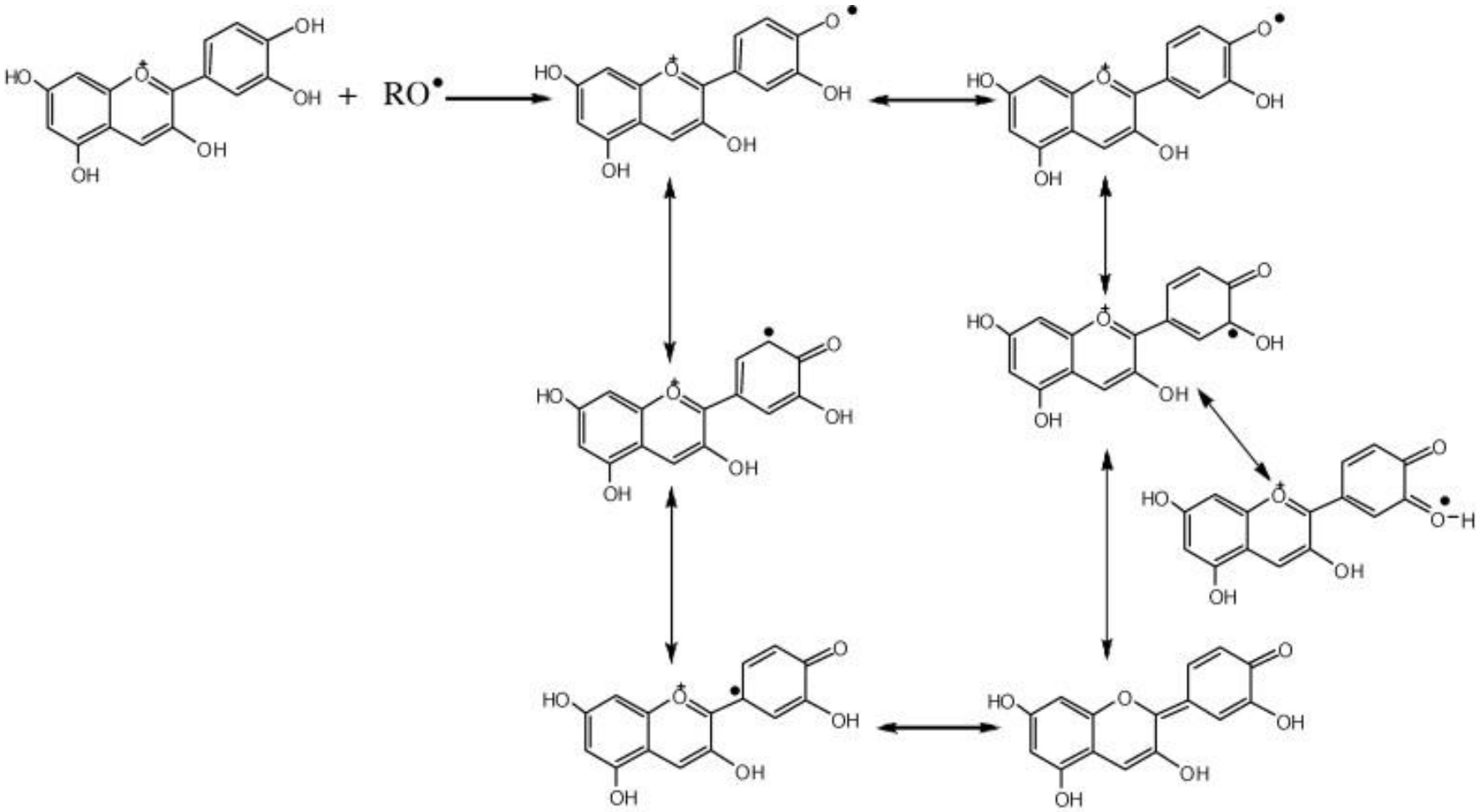
PHARMACOLOGICAL ACTIVITY

- **decrease capillary permeability and fragility** (confirmed by biological tests on animals based on the diffusion of dyes)
  - participation of the collagen of the vascular wall in the control of the permeability of that wall.
  - inhibition of the proteolytic collagen degradation enzymes (elastase, collagenase); (It has been shown in vitro for black currant extracts)
- **antiedema activity**,
- **increase in regeneration of** 'visual purple' or **rhodopsin** (see bilberry
- act like **radical scavengers** in vitro (antioxidant activity).

USES

- for the symptomatic treatment of venous and lymphatic insufficiency and capillary fragility (in phlebology, proctology, or gynecology).
- in ophthalmology
  - to treat circulatory disorders of the retina or choroid,
  - to improve vision at dusk.

Proposed mechanism for the stabilisation of the cyanidine semiquinone radical (resonance) (Castañeda-Ovando et al.).



http://dx.doi.org/10.1016/j.foodchem.2008.09.001, **Chemical studies of anthocyanins: A review,**

ANTHOCYANINS, Other uses

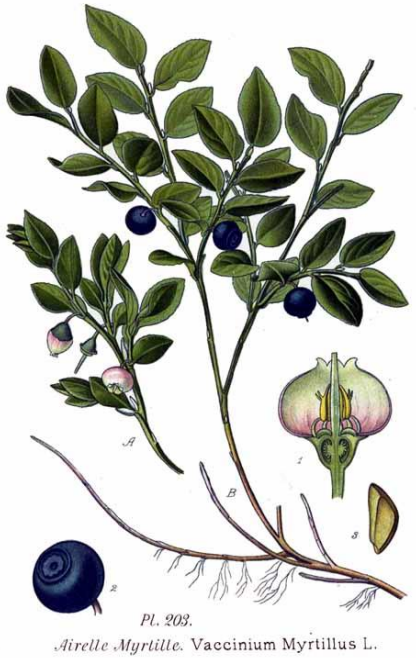
The chief industrial application of anthocyanins is **coloring**

- they are natural pigments for which no animal toxicity has been found, be it acute or chronic.
- Sources:
  - **unfermented grape juice**: → **liquids** titrated to contain 0.5-1% anthocyanins, or
    →**nebulisates** titrated to contain 1-5% anthocyanins.
  - **elderberry** or **red cabbage** leaves, which are more expensive, but provide a more stable coloring agent.
- Difficulties (restricts the scope of applications):
  - instability in aqueous media (high): it results in color changes as a function of the pH, and in sensitivity to heat, light, sulfites (often used as preservatives), and metals (food cans).
  - The common occurrence of proanthocyanidins and gallotannins in the extracts can also be a problem (for example it makes **gelatin precipitate** in jams).
  - insolubility of anthocyanins in lipids.
- Anthocyanins are extracted from edible fruits and vegetables and may be used as **food additives** (Eur. id. code E163), for example in beverages (30 mg/L), jams, and confectionery products, to name only a few.

CHIEF ANTHOCYANIN-CONTAINING DRUGS

BILBERRY, *Vaccinium myrtillus* L., Ericaceae

- The bilberry is a subshrub with **coriaceous leaves**.
- The **bell-shaped flowers** grow solitary or in pairs at the base of the leaves.
- The fruit is **a multiseeded** tetra- or pentalocular globose **berry** with a fleshy mesocarp;
- on the flattened top, the remains of the style and the calyx form a **small disc with a dull edge**.



- Blueberries are particularly abundant in the woods that grow on siliceous soils in the mountains of the northern hemisphere.
- The French market is largely dominated by imports (from Poland).
- Other species (e.g., V. corymbosum, cultivated in Germany) are also used in the food industry.

Vaccinium myrtillus, Chemical Composition

Blueberries

- Water (up to 90%), sugars (3 to 7%), and organic acids.
- Phenolic acids, **flavonoids** (hyperin = hyperoside], quercitrin), **proanthocyanidins** (procyanidins B-1 and B-4), and monomeric flavan-3-ols (catechin and epicatechin).
- **Anthocyanins** (in the **fresh fruits**: about 0.5%): C-3 *O*-glucosides, *O*-galactosides, and *O*-arabinosides of cyanidin,peonidin, delphinidin, malvidin , and petunidin.

Bilberry leaf

- Phenolic acids, **flavonoids** (rhamnoglucosyl-, arabinosyl-, and glucuronylquercetin).
- Traces of quinolizidine alkaloids (myrtine, epimyrtine).
- **Proanthocyanidins** and catechin (up to 10 %).

Uses

- **Water –soluble bilberry powder** titrated to contain 70 % anthocyanins. It is an ingredient of drugs used to treat
  - the functional symptomes of **venous and limphatyc vessel insufficiency** ,
  - cutaneous **capillary fragility**, and
  - mesopic and scotopic **vision** (nyctalopia, myopia).

Vaccinium myrtillus, Uses, Indications

In France

- **Bilberry fruit, fresh or dried**, and **bilberry leaf**: traditionally used
  - to treat the subjective symptoms of **venous insufficiency**, such as fullness in the legs, and
  - to relieve the **symptoms of piles**.
- **The fruit, fresh or dried**: is traditionally used for the adjunctive therapy of the painful component of **functional dyspepsia**.
- **Only the fresh fruit:** for the symptomatic treatment of the functional symptoms of **capillary fragility**,
- B**ilberry leaf or dried fruit**: for the symptomatic treatment of **mild diarrhea**.

In Germany, Commission E:

Bilberry fruit:

as an **adstringent**, in case of **diarrhae**;

as a topical **anti-inflammatory** in case of **irritation** of the **mucous membranes** of the mouth and throat.

CRANBERRY, *Vaccinium macrocarpon* Aiton, Ericaceae

- Cranberry grows wild in eastern North America, from the Carolinas to Canada.
- Cultivated in the United States since the beginning of the nineteenth century,
- It produces small dark red fruits
- widely consumed as such (**fresh or frozen**) and as cranberry j**uice** (pure or as a cocktail sweetened with corn syrup), cranberry **sauce**, and so on
- The **fresh fruit** is very rich in **acids** (citric. quinic. benzoic);
- it also contains **anthocyanins** (3-O-galactosides and 3-O-arabinosides of cyanidin and peonidin),
- **catechin**, and **flavonoids**.

Vaccinium macrocarpon

- The beneficial—**bacteriostatic**-—-**effect** of cranberry juice in the treatment of urinary infections is confirmed by secular use.
- It is now postulated that the activity is due to the **inhibition of bacterial adhesion onto mucous membranes.**
- This has been demonstrated in the case of *E. coli* adhesion onto urinary tract epithelial cells, using cranberry juice as well as the urine of mice or humans collected after administration of cranberries.
- The active constituent (possibly a procyanidine) inhibiting the adhesins specific to the pathogenic strains of E. coli has since been isolated from cranberry juice—and also from bilberry (blueberry) juice.
- More recently, a placebo-controlled, double- blind clinical trial showed that
- the **daily consumption of 300 mL of** a commercially- available **cranberry juice induced**, in elderly women (average age 78.5 years), a very **significant decrease in the frequency of urinary bacterial contamination**, after 4-8 weeks of treatment, a delay which may correspond to an initial action on the intestinal bacterial flora.
- In the United States, the recommended use of cranberries is as a dilute juice; dried juice capsules are also available.

BLACK CURRANT, *Ribes nigrum* L., Grossulariaceae

- This bushy shrub is cultivated for its edible fruits (in the Burgundy region of France and in central Europe).
- The black currant leaf as well as the fruit are used in pharmacy (Fr. Ph., 10th Ed.).

- Tri- to pentalobate leaves, the underside of which are pale, pubescent, and scattered with yellow secretory glands.
- Flowers : reddish, grouped in dangling racemes, a pubescent calyx which is longer than the corolla.
- Fruit : a fragrant black berry on top of which the remains of the calyx can be seen.

Constituents

- **Fruit**: sugars (10-15%); organic acids; flavonol glycosides ; anthocyanins : cyanidin and deiphinidin glycosides.
- **Leaves:** a small amount of essential oil; flavonoids: hyperin, astragalin, rhamnoglucosides and glucoxylosides of quercetin and kaempferol; dimeric and trimeric prodelphinidins.

BLACK CURRANT,

Uses

- **Fruit**
  - **t**o prepare extracts enriched in anthocyanins
  - With therapeutic indications identical to those of the bilberry

- **Leaves** (in phytopharmaceuticals, traditionally)
  - to facilitate urinary and digestive elimination functions,
  - to enhance the renal excretion of water, and
  - as an adjunct in weight loss programs.
  - orally and topically, for the symptomatic treatment of minor painful symptoms of the joints.

Contemporary phytotherapy prescribes the *preparations based on the* **buds** in the same fashion.

- These are rich in **diterpenoid acids** (hardwickiic acid), and are
- prized for their **essential oil**, which is used in food technology.
- **Essential oil composition** varies with cultivars, but the **chief constituents** are almost always **hydrocarbons** (A3- carene, sabinene, phellandrenes, and limonene).

VINE, *Vitis vinifera* L. (tinctoria varieties), Vitaceae

- The term “vine” designates cultivars with black grapes, red pulp, and leaves that turn red in the fall, partially or completely .

- The **dried vine leaf** was the subject of a monograph in the 10th edition of the **French Pharmacopoeia.**
- The pharmaceutical industry also uses **grape seeds.**

Constituents

- Anthocyanins (up to 0.3%): 3-O-glycosides of cyanidin and peonidin;
- monocaffeoyltartaric acid, phenylpropanoic acids,
- flavonol glucosides,
- hydrolyzable tannins (esters of glucose and of gallic and dehydrohexahydroxydiphenic acids),
- proanthocyanidins.

VINE

Uses

Vine leaf-based phytopharmaceuticals are traditionally used (orally and topically) to treat

- the functional symptoms of capillary fragility such as ecchymosis and petechiae,
- the subjective symptoms of venous insufficiency such as fullness in the legs, and
- the symptoms of hemorrhoids.
- Topically, they are traditionally used for eye irritation or discomfort of various etiologies (e.g., eye strain, seawater or swimming pooi water, or smoky atmospheres).

EUROPEAN ELDER, *Sambucus nigra* L., Caprifoliaceae

- The flower—it is the subject of a monograph in the European Pharmacopoeia .
- The fruit, a source of extracts used as food coloring.

- The European elder is a shrub widespread in western Europe.
- Its bark has small cracks and
- its leaves are imparipinnate.
- Large (20 cm) inflorescences of strong-smelling flowers,
- black berries with their purplish-red juice and three seeds.
- The flower is fairly easy to identify.

- However, to verify the absence, in the drug, of flowers of dwarf elder (*S. ebulus* L)—with red instead of yellow anthers—the French Pharmacopoeia requires a TLC analysis of the flavonoid content of a methanol extract.
- It contains sambunigrin

cyanogenic glycosides .

S. ebulus L

EUROPEAN ELDER

Flowers

Constituents

- Flavonoids (>0.8%, Ph. Eur.): rutin, isoquercitrin,
- Derivatives of caffeic acid, free and esterified.
- Triterpenes
- Essential oil
  - smells like muscat grapes, has a pasty consistency,
  - contains fatty acids, 3,7-dimethyl-1,3,7-octatrien-3-ol, linalol, cis-hexenol, and rose oxides, among others.

Uses

Elder flower

- **In France** it is traditionally used
  - to enhance the urinary and digestive elimination functions,
  - as an adjunct in weight loss programs, and
  - to enhance the renal elimination of water
- **In Germany**, Comission E indications
  - As a sudorific, causing an increase in bronchial secretion
  - Colds and coughs

EUROPEAN ELDER

Fruits

- **Constituents**
  - Cyanidin glycosides: 3-*O*-glucosyl-, 3-*O*-sambubiosyl-, 3,5-diglucosyl-, and 3-sambubiosyl-5-glucosyl-cyanidins.
  - Flavonoids, acids (citric, malic), saccharides, 0.1 mL/kg essential oil.
  - The seed contain cyanogenic glycosides .
- The ripe fruit,
  - edible fresh or as a jam ,
  - is the source of an extract used as food coloring (e.g., to color cherry or pomegranate syrup)
- Traditionally used – in France – for medicinal purposes, for the same indications as the leaf.

Phenoloids in Zingiberaceae family

(diarylheptanoids and arylalkanones)

TURMERIC, *Curcuma domestica* Val. = *C. longa* L., Zingiberaceae

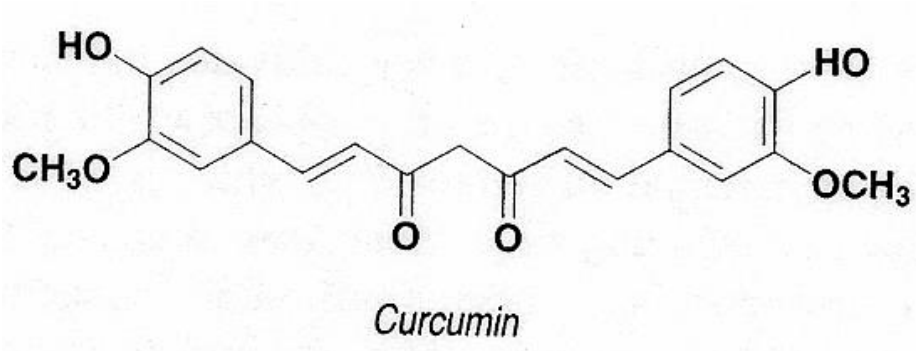
- **Perennial** by a rhizome,
- turmeric has **large sheathing leaves** with an elliptic blade and pinnate veins.
- The **flowers** are **yellow**, **gathered into a spike** with bracts, and
- have an **irregular corolla** with a developed **posterior petal**,
- an androecium reduced to one fertile stamen and staminodes forming a petaloid label, and
- a gynoecium with three carpels.

- **Several cultivars** grown in India, Sri Lanka, Indonesia, China, and Jamaica. For the most part (80%) the world production comes from India.
- Commercial turmeric commonly consists of the ovate primary rhizomes (“bulb” or “round” turmeric), the cylindrical secondary rhizomes (“fingers”), or a mixture of both.
- Fingers: gray and grooved surface and a diameter of about 1 cm.
- Break with a clean fracture, reddish-yellow inside; odor aromatic, taste warm,somewhat bitter.

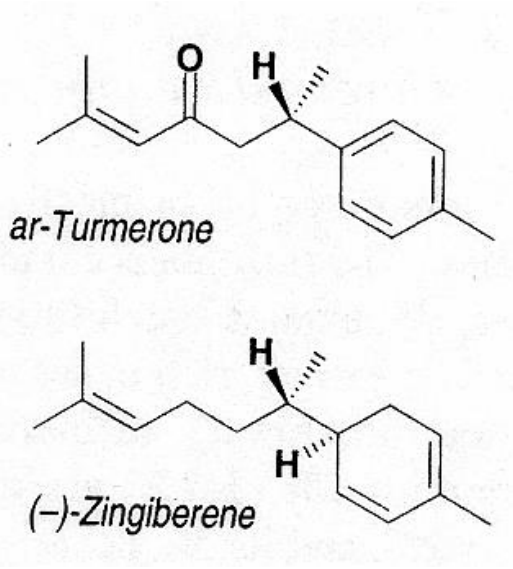
TURMERIC

Chemical Composition

- Starch (45-55%); arabinogalactans (ukonans)
- **Essential oil** (2.5 to 6%) rich in monocyclic sesquiterpenes :
  - hydrocarbons: **zingiberene,** - and 6-curcumene ; their oxygenated derivatives : **turmerone**, *S*-(+)-ar-turmerone, curlone, α- and y-atlantone;
- **Curcuminoids** (can reach 8 %):
  - The coloring principles in the drug, structurally related to a diarylheptane,
  - **curcumin** (50 to 60%), desmethoxycurcumin, bisdesmethoxycurcumin, dihydrocurcumin .



Curcumin



ar-Turmerone

(−)-Zingiberene

TURMERIC, Pharmacological Properties

Curcumin

- Its **anti-inflammatory activity** has been demonstrated in animal experiments and promoted by observations reported in India in man.
- Is apparently devoid of side effects.
- The mechanism of action remains poorly-understood:
  - inhibition of the increase in activity of lysosomal enzymes,
  - effect on the synthesis of prostaglandins, or
  - interference with the response of granulocytes to stimuli linked to the inflammatory phenomenon.

The drug has a definite action on the

- Hepatic parenchyma: the **hydroalcoholic extract prevents the cytotoxic effects of carbon tetrachloride** in vivo in the mouse and in vitro in cultured rat hepatocytes.
- Stomach: the **ethanolic extract** (0.5 g/kg in the rat) is **active against ulcers** and protects cells .

TURMERIC

Uses

- **Food coloring:** turmeric cultivars with the highest curcumin content (e.g., Allepey, > 6.5%).
- **Curcumin** (>90%):
  - It is a **nontoxic authorized color** (Eur. id. code E1oo).
  - It is **heat resistant** and scarcely sensitive to changes in pH.
  - It is used as the rhizome powder, or the oleoresin, or extracts and curcumin solutions of variable concentration, sometimes adsorbed onto hydrocolloids.
- **Spice:** Madras (3.5% curcumin) and other cultivars . **Turmeric** is, alongside coriander and other spices, **one of the main ingredients** of **curry powders** (these may also contain chili, ginger, clove, fenugreek).
- **Oleoresin** is also used in food technology.
- **In phytopharmaceuticals** : traditionally used
  - as a choleretic and cholagogue,
  - for functional dyspepsia attributed to a hepatic origin,
  - as an appetite stimulant.
  - Biliary tract obstruction is a contraindication (Commission E).

TEMU LAWAQ, *Curcuma xanthorrhiza* Roxb., Zingiberaceae

- Temu lawaq is botanically very close to turmeric, and is a cultivated **Indonesian species.**
- **The rhizome** is cut after being harvested, so the drug appears as thin round slices.

Constituents

- Starch (30-40%),
- **Essential oil** (up to 12%): rich in **sesquiterpenes**: **zingiberene**, **ar-curcumene,** (*R*)(—)-xanthorrhizol, turmerones, bisacurones, bisacumol, and bisacurol.
- **Curcuminoids** (1-2%): **curcumin**, its monodemethoxylated derivative, and its di-, hexa-, and octahydrogenated derivatives. Monophenolic and non-phenolic analogs have been isolated from rhizomes collected in Thailand.

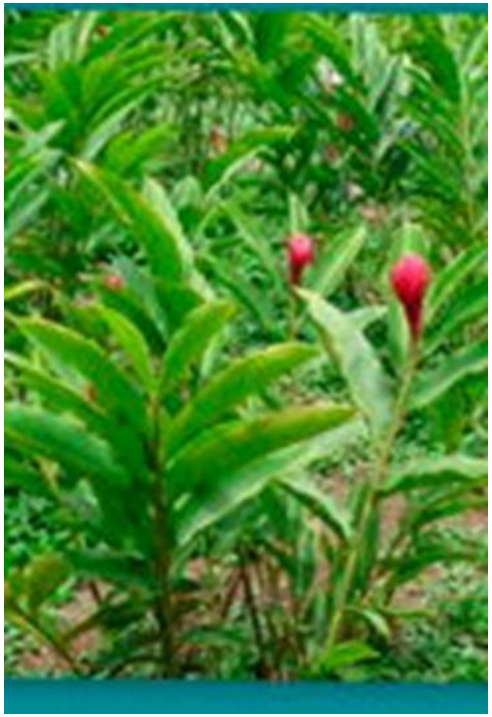
Uses

- A traditional folk remedy in southeast Asia: it is used as **cholagogue** and **choleretic.**
- In Germany, Commission E :
  - its use acceptable **for gastrointestinal symptoms**,
  - biliary tract obstruction is a contraindication,
  - prolonged use can cause gastric irritation.

GINGER, *Zingiber officinalis* Roscoe, Zingiberaceae

This **spice**, is used in the oriental traditional medicines, especially for **functional dyspepsia**.

- Originally from India, ginger is cultivated in India, China, and all of southeast Asia (Indonesia, Philippines), and in the tropical regions of Africa (Nigeria).



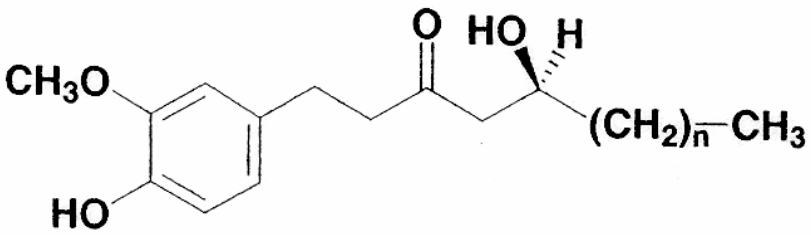
- Large herbaceous perennial plant,
- lanceolate leaves,
- thick inflorescence with overlapping lateral bracts,
- pale green flowers with purple label.

- The rhizome is ramified within one plane.
- Appearance, depending on the mode of preparation: gray with a wrinkled surface (coated or unscraped), white with a smooth surface (uncoated or scraped), or prepared (preserved).
- Fibrous and granular fracture; odor aromatic, taste warm and pungent.

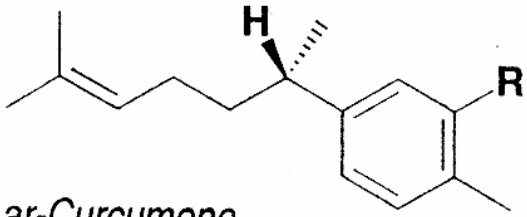
GINGER

Chemical Composition

- Starch (60%), proteins, fats (10%), from 10 to 25 mL/kg essential oil, and a resin.
- **Essential oil** (composition highly depends on geographical origin):
  - **Sesquiterpene hydrocarbons** (30-70%): (—)-**zingiberene**, (+)-**ar-curcumene**, (—)-β-sesquiphellandrene, *E,E*-β-farnesene, β-bisabolene. Monoterpene aldehydes (citrals) and alcohols.
- **Gingerols =** 1 -(3' -methoxy-4' -hydroxyphenyl)-5-hydroxyalkan-3-ones:
  - [3-6]-, [8]-, [10]-, and [12]-**gingerols** bearing a side chain with 7-10, 12, 14, or 16 carbon atoms, respectively;
  - alongside the **corresponding ketones**, and dehydration products (**shogaols**).
- Labdane-type diterpenes, galonolactone and its dialdehyde derivative.



Gingerols (n=1-4,6,8,10)



R = H: *ar-Curcumene*
R = OH: *Xanthorrhizol*

GINGER, Pharmacological Properties

- Used since remote times in India and China.
- **Animal experiments** :
  - oleoresin is a **cholesterol lowering** agent (in rodents),
  - [6]-gingerol is a **cholagogue** (in the rat by the intraperitoneal route),
  - [8]-gingerol is a **hepatoprotective** agent (prevents the toxic effects of carbon tetrachloride in rat hepatocytes).
  - the acetone extract and zingiberene have an **antiulcer effect** in the rat
  - The drug has **an anti-inflammatory activity** (possibly acting on prostaglandin and leukotriene production).
- **Human studies** (on antiemetic properties)
  - Most trials reveal an activity superior to that of a placebo for **motion sickness**, **post-operative nausea**, or **morning sickness** (at the usual dose of 1 g per day).
  - These trial results are divergent ; (among the reasons: ginger products used were not standardized).
- **Antiemetic action**: may be the consequence of direct effects on the gastrointestinal tract: in the mouse, the stimulation of gastrointestinal motility by the acetone extract (75 mg/kg), by [61-shogaol (2.5 mg/kg), or by gingerols is comparable to that of metoclopramide (10 mg/kg).
- Other authors, however, noted the lack of effect of ginger powder on the rate of gastric emptying in healthy humans.
- The drug is not toxic and has no side effects.

GINGER

Uses

- Used (especially for functional dispepsia) for over 25 centuries in the formulation of countless traditional Oriental remedies (China, Japan).
- **In France**, in phytomedicines: traditionally used **for motion sickness**.
- In Germany: the rhizome powder is used for gastrointestinal distress and to prevent motion sickness (2 g/day).
- Commission E:
  - Ginger is a spasmolytic in animals
  - in humans, it has antiemetic, positive inotropic, and stimulant effects (intestinal peristalsis, salivary and gastric secretions).
  - Ginger must not be used to prevent morning sickness in pregnant women.

KAVA, *Piper methysticum* Forst. f., Piperaceae

- *P. methysticum,* a pepper tree which grows in the islands of western Polynesia (Papua New Guinea, Tonga, Samoa, Fiji, Vanuatu) and as far as Tahiti.

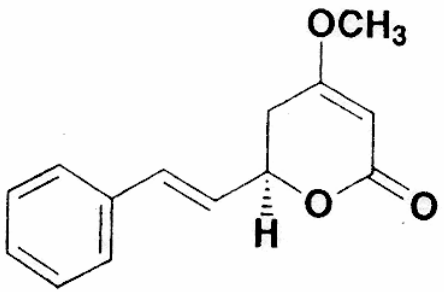
- A perennial dioecious **subshrub** with
- **cordate leaves.**
- **Decaploid** and **sterile**.
- Multiplies by vegetative propagation.

- The term **kava** designates **a beverage** prepared by soaking in water the rhizome or root fragments, after grinding them with a pestle or chewing them.
- It has been consumed for centuries according to a ceremonial described in 1875 by Captain Cook.
- This ritual beverage **induces a sensation of well-being**.
- It continues to play an important role in the culture of that part of the globe.

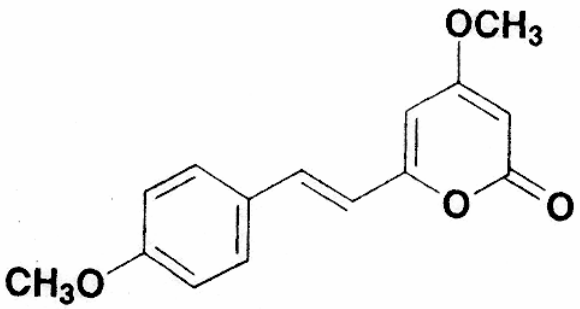
KAVA

Constituents

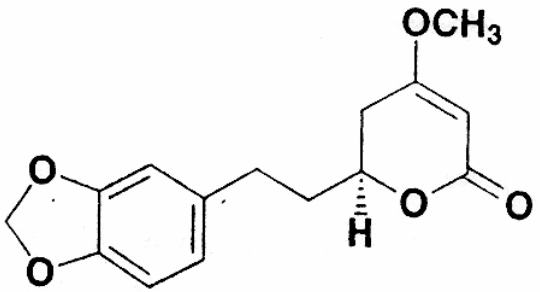
- Mono- or di-**unsaturated α-pyrones**, substituted by a styryl or phenethyl group, itself substituted (methoxyl, methylenedioxyl) or not.
  - They include **yangonin**, (+)-methysticin, (+)-**dihydromethysticin**, (+)-**kawain,** (+)-dihydrokawain**,** demethoxyyangonin, and minor products (e.g., dehydrokawain, 7 ,8-dihydroyangonin, 10- and 11 -methoxyyangonins).
- **Resin** *content* can fluctuate *from 3 to 20%* depending on cultivars and location (rhizome, lateral roots) and its *composition* varies with the *chemotype*.



Kawain



Yangonin



Dihydromethysticin

KAVA, Pharmacological research

- The **pyrones**
  - **induce sleep** in rodents (per os) and **are sedatives** in rodents, cats, and rabbits.
  - cause **muscle relaxation** and several are **anticonvulsan**t (strychnine, electroshock).
- The **kavapyrones** (DHK, DHM) are analgesics and weak local anesthetics.
- The **aqueous extract** and the **lipid-soluble fraction** decrease spontaneous movement, but
- the (mild) sedation induced by the aqueous extract is not accompanied by a loss of muscular tone;
- the resin induces sleep, but the aqueous extract does not (mouse, IP).
- **Kava** and **kawain induce sleep** by **acting on the limbic system** (EEG in cats)
- **Klinical trials** indicated that **a kava extract** is more **efficacious** than a placebo in patients who suffer from **non-psychotic anxiety**.

KAVA, Uses

- In Germany, **pharmaceuticals based on standardized extracts** (i.e., 35-120 mg kavapyrones) were promoted **as sleep disorder and anxiety medicines**.
- **Nowadays their application is contraindicated** due to the
- **hepatotoxic adverse reactions** reported in association with the use of **all types of kava products** in the South Pacific Islands, Australia, Europe, and the US.
- It appears that **poor quality of the kava material** was responsible for the **liver toxicity**.
- Therefore, a sophisticated approach to establish kava quality standardizations is needed for safe human use of kava as
  - relaxing traditional beverages,
  - the anxiolytic drugs, and
  - recreational dietary supplements.