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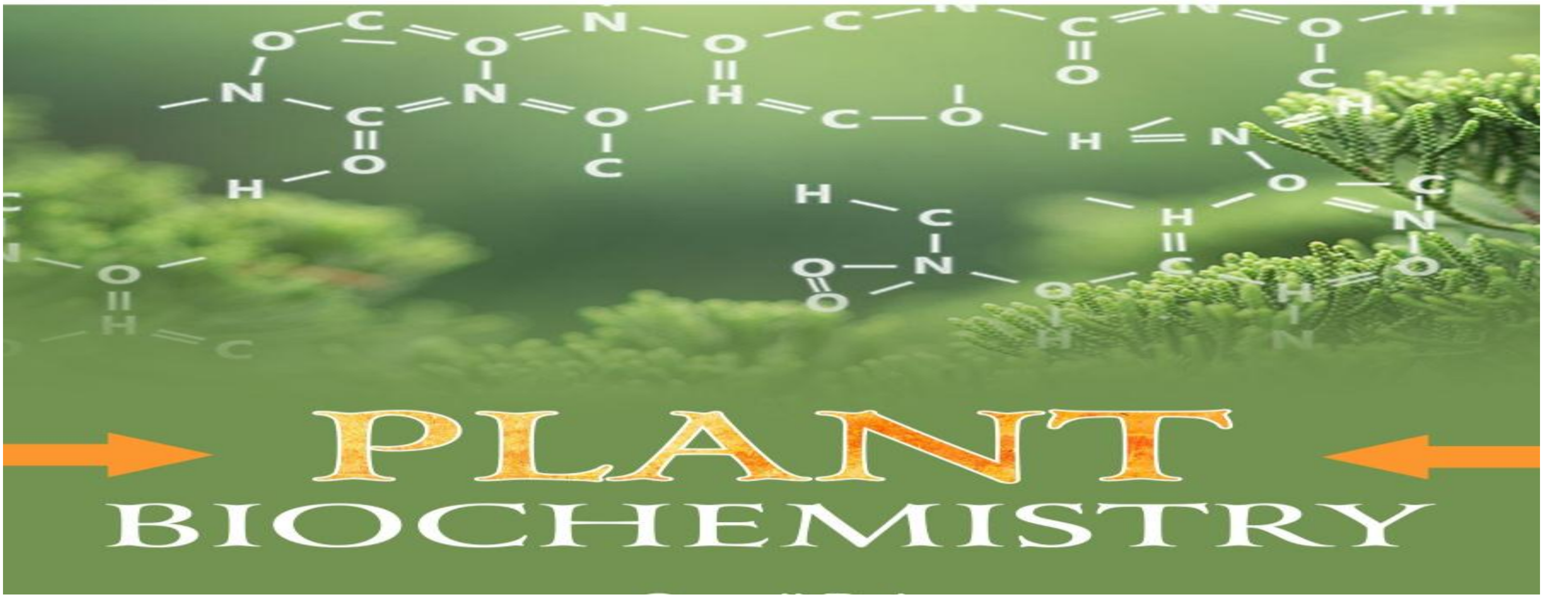

Plate 39. Polluted-water algae.

PLANT BIOCHEMISTRY & PHYCOLOGY

FOR 4TH YEAR SPECIAL BOTANY

BY

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Phycology

Definition of Algae

Algae are a diverse group of simple, plant-like organisms. Like plants, most algae use the energy of sunlight to make their own food, a process called photosynthesis. However, algae lack the roots, leaves, and other structures typical of true plants. Algae are the most important photosynthesizing organisms on earth. They capture more of the sun's energy and produce more oxygen (a byproduct of photosynthesis) than all plants combined. Algae form the foundation of most aquatic food webs, which support an abundance of animals. Algae vary greatly in size and grow in many diverse habitats. Microscopic algae, called phytoplankton, float or swim in lakes and oceans. Phytoplanktons are so small that 1000 individuals could fit on the head of a pin. The largest forms of algae are seaweeds that stretch 100 m (300 ft) from the ocean bottom to the water's surface. Although most algae grow in fresh water or seawater, they also grow on soil, trees, and animals, and even under or inside porous rocks, such as sandstone and limestone. Algae tolerate a wide range of temperatures and can be found growing in hot springs, on snow banks, or deep within polar ice.

Habit and Habitat

The algae are predominantly aquatic and are found in fresh or salt waters. Fresh water forms occur abundantly in ponds, lakes, slow flowing streams and water reservoirs. In habit they may be free swimming, free floating or attached to the bottom in the shallow water. Some are terrestrial and grow in wet situations, such as, on damp soil, damp shaded sides of trees and walls or even rocks and thus have adapted themselves to a life in the air. They may live as epiphytes as well as epizoics. A few occur in association with fungi. Some algae are endophytic whereas a few live in symbiotic relationship with the higher plants. Most of the marine forms are *Sea Weeds.* They inhabit vast area of the ocean. Some of the marine forms are free floating. A few are attached. The free floating and free swimming initiate algae together with similar other organizations constitute the *plankton* of the ocean and lakes.

According to the habitat, the algae may be classified as follows:

1. Aquatic algae
2. Terrestrial algae
3. Aerophytes
4. Cryophytes
5. Thermophytes
6. Algae of unusual habitats

1. Aquatic algae: Majority of the algal genera are aquatic and found either completely submerged or free floating on the surface of water. Aquatic algae usually occur in ponds, pools, tanks, ditches, streams or in slow running rivers and are called fresh water forms. Marine algae are found in sea and macroscopic large thalli of brown algae are commonly known as "sea weeds". Fresh water algal forms like *Chlamydomonas, Volvox, Hydrodictyon* are found in stagnant waters, whereas *Cladophora, Oedogonium, Ulothrix* and few species of *Vaucheria* occur in slow running water bodies. Most of the members of Phaeophyceae and Rhodophyceae are found in sea either floating on the surface of sea water or attached with rocks or any other substratum. The free floating and free swimming microscopic algal forms together with other similar organisms constitute the *Planktons* of water bodies. Plankton forming algae may either be free floating from very beginning and are never attached (Euplanktons) *e.g., Microsystis, Chlamydomonas, Scenedesmus* and*, Cosmarium* or in the beginning may be attached but later on they get detached and become free floating (Tychoplanktons) *e.g., Zygnema, Oedogonium, Cladophora, Cylindrospermum, Rivularia* etc.

2. Terrestrial algae: Many algal genera are found on or beneath the moist soil surface are called terrestrial algae. The algal forms occurring on the surface of soil *e.g.* few species of *Vaucheria, Botrydium, Fritschiella* and *Oedocladium* are called *Sapophytes* while algal genera having subterranean habit *e.g.,* few species of *Nostoc, Anabaena* and *Euglena* are known as *Cryptophytes.*

Factors affecting the soil algae: A number of factors have been found to be associated with the growth and diversity of soil algae.

Moisture: it is one of the important factors for the growth of soil algae which is required to complete the life cycle.

Temperature: Many blue-green algal forms can tolerate temperature between 60-90 ^{0}C and due to this they are common in tropical soils. Diatoms can survive very low temperatures.

Light: Although many algae can withstand bright sunlight but their growth is maximum in less bright light.

pH and salinity: pH has an important role in determining the algal groups. Many green algae can be found in a wide range of pH while blue-green algae prefer neutral or alkaline pH. Blue-green and diatoms do not prefer acidic soils.

Soil texture and chemical composition: The chemical properties and the texture of soil are important factors which decide the type and growth of algal flora.

3. Aerophytes: Such algal forms as are adapted for aerial mode of life and occur on the trunks, moist walls, flower pots, rocks, and get their water and carbon dioxid requirements completed directly from atmosphere are called Aerophytes. *Trentepohlia* is found the bark of trees in moist and humid climatic conditions while *Phormidium, Scytonema* & *Hapalosiphon* have been observed to grow on bark of trees along with Bryophytes.

4. Cryophytes: These algae are found on the mountain peaks covered with snow and impart attractive colours to the mountains. *Haematococcus nivalis* gives red colour to Arctic and Alp regions while *Chlamyodomonas yellowstonensis* along with some species of *Ankistrodesmus* is responsible for the green colour of the snow of the mountain of European countries particularly in Arctic region.

a) Those algae which are found on snow and not on ice *e.g.,* some species of *Raphidone* and *Chlamydomonas.*

b) Those algae which can grow only on ice and result in "ice bloom" *e.g., Ancyclone, Mesotaenium.*

c) Those algae which can grow on snow and ice both *e.g.,* and *Cylmdrocys*

d) Those algae which are not true cryophytes and have their temporary growth on ice snow e.g., *Phormidium* and *Gloeocapsa*.

5. Thermophytes: The algal genera occurring in hot springs at quite high temperature are called thermophytes. There are certain algae which are known to tolerate the temperature up to 85 ^{0}C e.g. few genera belonging to family Chroococcaceae and Oscillatoriaceae. *Oscillatoria brevis, Synechococcus elongatus* and *Haplosiphon lignosum* are some common examples of thermophytes which can survive up to a temperature of 70°C at which generally plant life is not possible.

6. Algae of unusual habitats: Many algae are found at various interesting places and according to their habitats may be of following types:

a) Halophytic algae. These algae are found in saline water containing high percentage of salts e.g., *Dunaliella, Stephanoptera* and *Chlamydomonas chrenbergii*.

b) Lithophytic algae. Usually the members of Cyanophyceae grow on moist rocks, wet and other rocky surfaces. Blue green algae *Rivularia* and *Gloreocapsa* occur on exposed rocks, whereas *Nostoc* is found growing in damp shady habitats. Several marine belonging to Rhodophyceae and Phaeophyceae are lithophytic in habit and grow on submerged rocks or rocky surface *e.g., Ectocarpus, Polysiphonia* etc.

c) *Epiphytic algae.* Such algal forms which grow on the other aquatic plants are called *Epiphytic algae.* Green algae *Chaetonema* has been found growing on *Tetraspora* and *Batrachospermum. Rivularia* are observed to grow on Angiospermic plant.

d) *Epizoic algae.* Many algae grow on the shells of molluscs, turtles and fins of fishe are known was *epizoic algae. Cladophora* is found on snails and shells of bivalves.

e) *Endozoic algae.* Contrary to epizoic algal forms endozoic algae are found inside the aquatic animals *e.g., Zoochlorella* is found inside *Hydra viridis* while *Zooxanthe* known to occur inside the fresh water sponges.

f) *Parasitic algae. Cephaleuros virescens* which causes 'red rust of tea' is a striking example of parasitic algae and causes heavy damage to tea foliage. *Chlorochytrium* and *Phyllosiphon* are other examples of parasitic algae. *Polysiphonia festigata* a member of Rhodophyceae is reported as semiparasite on *Ascophyllum nodosum.*

g) *Symbiotic algae.* Several members of Cyanophyceae grow in association with other plant and lichens exhibit good example of it. Almost all the plant groups are known to have symbiotic association with blue green algae *e.g., Nostoc* is found within the thalli of *Anthoceros* and *Notothylas, Anabaena cycadeae* is reported in the coralloid roots of Cycas, *Anabaena azollae* occurs in *Azolla* etc. The association of *Chlorella* with nitrogen fixing bacterium *Azotobacter chrooccocum,* and with that of certain species of *Ceratophyllum* and mosses are other examples of symbiotic algae.

Organisation of the thallus

The algae exhibit a great diversity in the organisation of the plant body. The simplest forms are motile or non-motile unicells (*Chlamydomonas* and *Chlorella*). In many species, the cells are grouped into aggregations called colonies *e.g., Volvox* and *Pediastrum.* These assume various forms and may be a hollow sphere, a flat plate or a filament. The filamentous types are usually multicellular and the filament may be simple *e.g., Ulothrix, Zygnema* or branched or an aggregation of filaments or a*,* highly organised thallus of a large size. In some multicellular forms the cells may perform both functions, vegetative and reproductive as in *Oedogonium,* while in others special reproductive cells or organs may be developed, e.g., *Chara*, *Sargassum* etc.

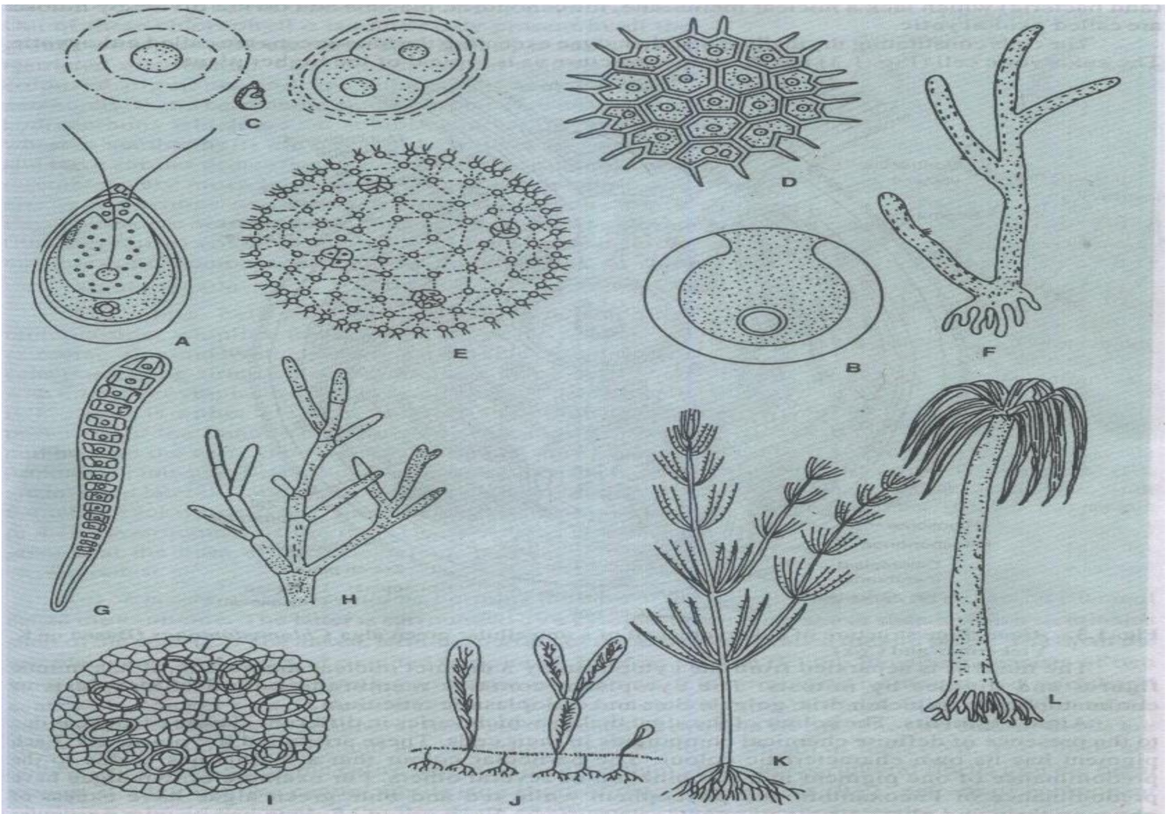


Fig. 1.2. Range of thallus organisation in Algae. (A). *Chlamydomonas*, (B). *Chlorella*, (C). *Gloeocapsa*, (D). *Pediastrum* (E). *Volvox* (F). *Vaucheria* (G). *Ulothrix* (H). *Cladophora*, (I). *Coleochaete* (J). *Caulerpa* (K). *Chara* (L). *Postelsia*

Structure of algal cell

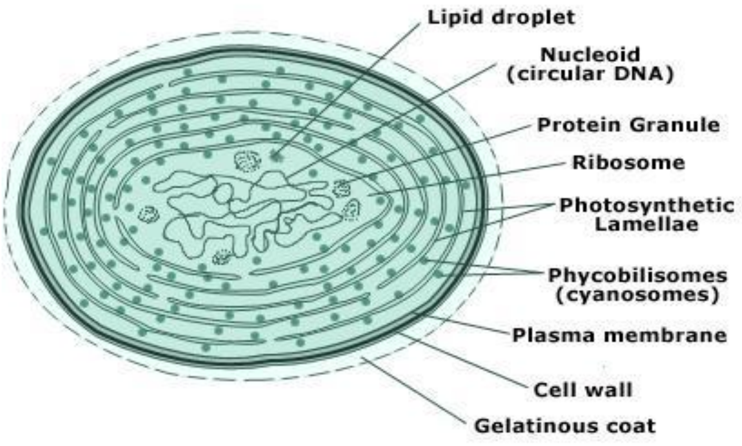


Fig.2. Cell Structure of Cyanophyta

The cells constituting the algal thalli are basically of two kinds, prokaryotic and eukaryotic. The prokaryotic cells (Fig.2) which constitute thalli of cyanophyta (blue-green algae) have a cell wall which contains a specific strengthening component not found in the cell walls of other algae. The central nucleoid has many irregularly arranged fine strands of DNA.

The chlorophyll-pigment is bound to photosynthetic lamellae or thylakoids which may be arranged in parallel layers in the periphery of the cytoplasm or form a network extending throughout the cell cytoplasm. They are not organised into grana. The chloroplasts are thus absent and so are the mitochondria, golgi body and endoplasmic reticulum. The ribosomes are, however, present. The nuclear division does not take place by mitosis and no cell plate is formed. Such simple cells of blue-green algae (and bacteria) which lack a nuclear membrane, mitochondria, and plastids and do not divide by mitosis are called prokaryotic. The cells constituting the thalli of all other algae excepting the blue-greens are called eukaryotic. The eukaryotic cell (Fig.3) has the same structure as is typical of the higher plants.

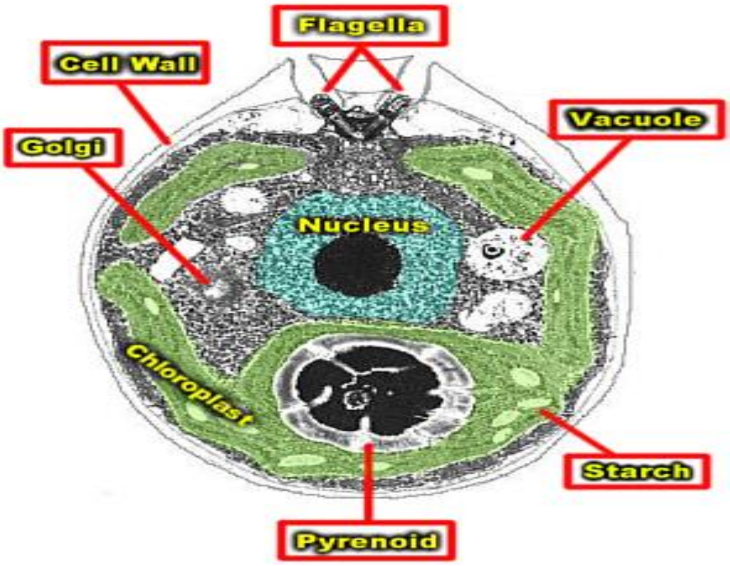


Fig.3. Cell structure of green algae

The nucleus is separated from the cytoplasm by a distinct nuclear membrane. It has mitotic figures and divides by mitosis. The cytoplasm contains membrane bound chloroplasts or chromatophores, mitochondria, golgi bodies and endoplasmic reticulum.

Algal pigments

The colour of the algal thallus which varies in different classes of algae is due to the presence of definite chemical compounds in their cells. These are called the pigments; each pigment has its own characteristic colour. The particular colour that a thallus has is due to the predominance of one pigment in a combination of several others. For example brown algae have predominance of Fucoxanthin and phycophein while red and blue green algae have excess of phycoerythrin and phycocyanin pigments respectively. Each group of algae has its own particular combination of pigments and a characteristic colour which is not found in the other algal groups. In fact the various algal groups show striking differences of colour. Thus the nature of the pigments present in the algal cells forms a quick guide to the primary classification of algae into divisions. The photosynthetic pigments in algae are of three kinds, namely, **chlorophylls, carotenoids** and **phycobilins** or **biliproteins**. The algal chlorophylls are characterized by green colour and in solution they show the phenomenon of fluorescence and emit red light.

1. **Chlorophyll** pigments are fat soluble compounds and are of five

different types. chlorophyll a, b, c, d, and e. Out of these chlorophyll a is universally present in all the groups of algae whereas chlorophyll b, c, d and e have restricted distribution.

Chlorophyll a - present in all higher plants and algae.

Chlorophyll b - present in all higher plants and green algae.

Chlorophyll c - diatoms and brown algae.

Chlorophyll d - red algae.

Chlorophyll e - xanthophyta.

(Chlorophyll a is present in all photosynthetic organisms that evolve O_2.)

2. Carotenoids are fat soluble yellow coloured pigments and are subdivided into carotene, xanthophylls and carotenoid acids.

a) Carotens are unsaturated hydrocarbons which are fat soluble and yellow in colour.

b) Xanthophylls are oxygen derivatives of carotenes and have similar properties like carotenes.

c) Carotenoid acid resembles very much with carotenes and xanthophyll, and are hydrocarbons consisting a chain of carbon atoms.

3. Phycobilins are water soluble blue (phycocyanin) and red (Phycoerythrin) coloured pigments and are present in the members of Cyanophyceae and Rhodophyceae.

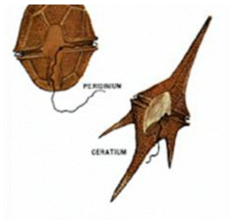
Taxonomic Group	**Photosynthetic Pigments**
Cyanobacteria	chlorophyll *a*, phycocyanin, phycoerythrin
Green Algae (Chlorophyta)	chlorophyll *a*, chlorophyll *b*, carotenoids
Red Algae (Rhodophyta)	chlorophyll *a*, chlorophyll d, phycocyanin, phycoerythrin, (phycobilins)
Brown Algae (Phaeophyta)	chlorophyll *a*, chlorophyll c, fucoxanthin and other carotenoids
Golden-brown Algae (Chrysophyta)	chlorophyll *a*, chlorophyll c, fucoxanthin and other carotenoids
Dinoflagellates (Pyrrhophyta)	chlorophyll *a*, chlorophyll *c*, peridinin and other carotenoids

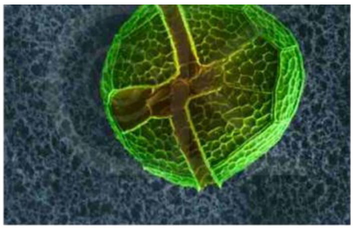
Dinoflagellates

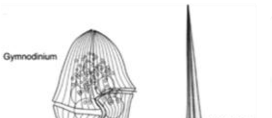
About 75% of the known toxic marine algal species are dinoflagellates which are microscopic. Dinoflagellates are mostly unicellular. The cell wall of these algae is only present when in the cyst stage. Most species of dinoflagellates have two flagella. One typical flagellum extends behind the cell. The other, usually shorter flagellum lies in a groove encircling the cell.

Dinoflagellates are an important component of plankton, the primary producers of organic material in the oceans. While this makes them important as a food source, some species of dinoflagellates are poisonous.

Some dinoflagellate species are photosynthetic, some eat other organisms and some do both. There are around 2,000 species of dinoflagellates in the world. While most species are not harmful, about 30 dinoflagellates produce potent neurotoxins that are capable of producing poisoning in human consumers of contaminated seafood. Some species can grow rapidly, accumulating near the sea surface and discoloring the water in a phenomenon called a red tide. While red tides usually have benign effects, some species produce toxins as they redden the sea. Other algal species produce toxins but do not discolor the sea at all The Red Tides common off the coasts of Florida and Mexico are caused by dinoflagellates and can kill millions of fish.







Algal flagella

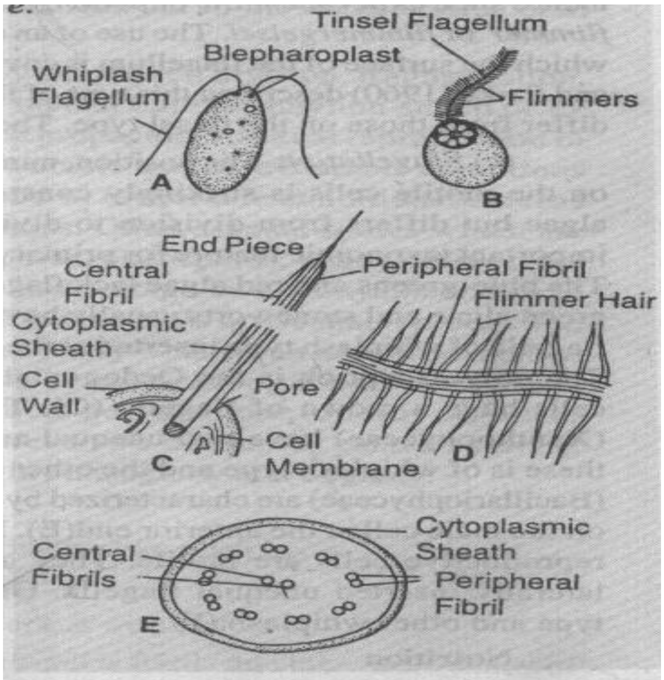


Fig. 1.4. (A-E). *Algae*. Kinds of Flagella and their structure. (A), Whiplash; (B), Tinsel; (C), L.S. Whiplash Flagellum; (D), L.S. Tinsel Flagellum; (E), C.S. Eukaryotic Flagellum.

The motile cells of algae are provided with fine, protoplasmic, whiplike threads, the flagella (A). They are extremely fine and hyaline emergencies of the cytoplasm. In cells possessing firm cell walls, the flagella are connected with the inner cytoplasm through small pores in the cell wall (C). There is either a single anterior flagellum (rarely posterior) or the flagella occur in pairs (A), rarely in great numbers on the cell. The flagella on the cell may be equal **(**isokont**)** or unequal **(**heterokont**)** in length. When the flagella are inserted laterally one is directed forwards in motion and the other backwards. They function as the locomotory or propelling structures of the cell. Usually there is a single granule at the base of each flagellum. It is known as the blepharoplast.

a) Kinds of flagella

They are of two main types, whiplash (A) and tinsel (B)*.* The whiplash flagellum has a smooth surface. The tinsel flagellum bears longitudinal rows of fine, minute flimmer hairs arranged along the axis almost to the tip of the flagellum. There may be a single row of hairs as in the Euglenophyta and Pyrrophyta or two as in Chrysophyceae and Phaeophyceae. The hairs arise from the margins of the peripheral fibrils. The whiplash or smooth flagella are also known by other names such as acronematic or peitchgeisel. The other names for the tinsel flagella are pantonematic**,** flimmer or flimmergeisel. The use of electron microscope has revealed a third kind of flagellum in which the surface of the flagellum is covered by scales (*Chara*) and minute, short, stiff hairs. The hairs differ from those on the tinsel type. They can be easily detached.

b) Flagellation

The position, number and kinds of flagella on the motile cells are strikingly constant in each division of algae but differ from division to division. Thus it forms an important taxonomic feature for primary classification of algae. The blue – green and red algae lack flagella. The motile cells in green algae and stoneworts usually have two, rarely four equal flagella of whiplash type inserted at the anterior end (A and B) the only exception is the Oedogoniales in which the motile cells have a crown of flagella (C) . The yellow green algae (Xanthophyceae) have two unequal anterior flagella. One of these is of

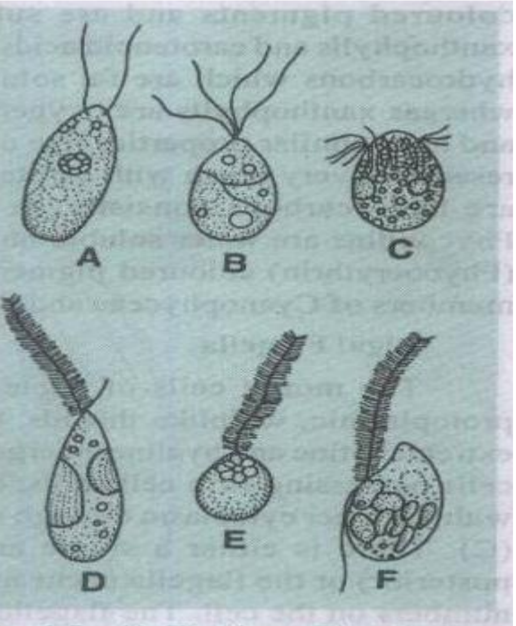


Fig. 1.5. (A-F) *Algae.* Flagellation., Chlorophyceae (A-C); Xanthophyceae (D); Bacillariophyceae (E) and Phaeophyceae (F).

whiplash type and the other tinsel (D). The diatoms (Bacillariophyceae) are characterized by a single tinsel flagellum on the male cell at the anterior end (E). In brown algae only the reproductive cells are motile. They are furnished with two unequal flagella. One of these is of tinsel type and other whiplash (F).

Nutrition

With respect to their nutrition the algae are eutrophic. All or most of the cells of the thallus normally contain chlorophyll. The green cell can manufacture their carbohydrate food from carbon dioxide and water through the agency of sunlight. The aquatic forms obtain water and carbon dioxide by osmosis and diffusion respectively from the medium in which they grow. The aerial forms obtain water from the damp substratum and carbon dioxide from the air. The algae are also to synthesize oil and proteins from the carbohydrates which they manufacture and soluble forms of nitrogen and other minerals available in solution in the water in which they grow.

Food reserves

The food materials which accumulate as food reserves in the form of polysaccharides, however, vary from group to group and thus provide useful data for preliminary classification of algae. True starch is typical of only two algal divisions namely, Chlorophyta and Charophyta. The two other kinds of characteristic starches are the cyanophycean starch (Cyanophyta) and floridean starch (Rhodophyta). The three other important polysaccharides which accumulate as reserve food are laminarin found in the brown algae, paramylon characteristic of Euglenoids and leucosin peculiar to the Xanthophyta, Bacillariophyta and Chrysophyta. Besides, a proteinaceous compound cyanophycin is found only in the cells of blue-green algae. Mannitol which was formerly considered to be unique to the brown algae has recently been reported to occur in a few red algae. Fats occur as reserve food in the cells of Xanthophyta, Bacillariophyta and Chrysophyta.

Factors affecting algal growth

1. Light

The light of the important factors affecting the growth of algae in the waters of rivers and lakes and in rivers, but the light may be less often because of turbidity, which accompany the rivers, which lead to block the light from the sun except a thin layer of surface water.

2. Macronutrient

Nutrients, major inorganic Macronutrient such as: phosphate and Nitrate.

Are the major nutrients of the key factors that determine the number and types of algae, especially nitrates and phosphates and the element is added to the silica for the diatoms.

3. Temperature

Various factions of algae for micro-, super- and optimal temperature in which to grow, for example, the optimum temperature for the growth of Diatoms ranging from 18-30 °C, while green algae thrive at temperatures ranging from 30-35 °C, and blue-green algae thrive at temperatures ranging between 35 -40°C.

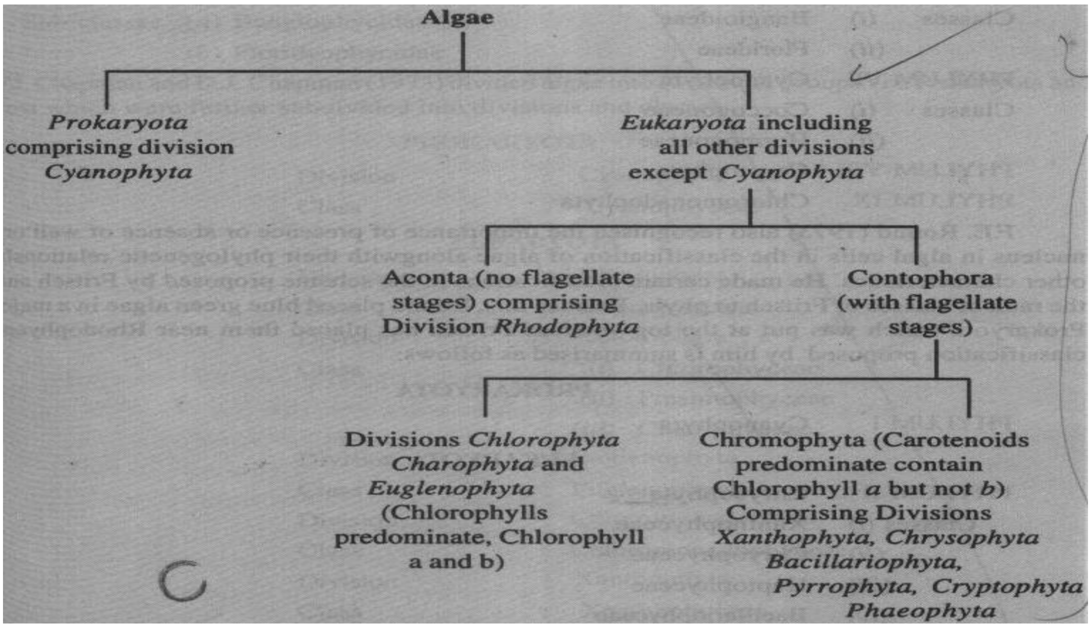
4. Micronutrient

Most algae grow best when the concentration of iron in the water ranges between 0.2 to 2 mg/l, while the observed toxicity of iron when the increased focus on the 5 mg/l.

5. Biological factors: such as

a) Competition between aquatic organisms on nutrients.

b) The rate of other organisms feeding on algae.



Classification

Christensen (1964) proposed a new scheme of primary classification of algae into Procaryota and Eucaryota on the basis of difference between the Procaryotic and Eucaryotic cells. It is briefly given below:

Economic important of algae

Benefical role of algae

1. Algae as human food: From ancient times large numbers of algae have been used as human food. They are often mixed with rice and fish and consumed as salad, soups etc. *Spirulina* is a blue-green microalga with a long history as a food source in East Africa. As it is high in protein and other nutrients it is currently used as a food supplement and as a treatment for malnutrition. *Chlorella elliposidea* is used successfully with tea in Japan.

2. Algae as fodder: Seaweeds are classified as Rhodophyta (red algae), Phaeophyta (brown algae) or Chlorophyta (green algae) depending on their nutrition and chemical composition. The Chlorophyta (green algae) is an important group of marine algae, which is important both ecologically and scientifically. Seaweeds mainly the members of brown algae are used as feed for domestic animals in different parts of the world. *Sargassum, Fucus* and *Laminaria* are consumed by cattles in Scotland. The use of dried seaweeds as cattle food has enhanced ten percent milk yield. The milk does not any taste of algae after the Seaweed diet. *Rodymenia* is a common cattle-feed in France. Macrocystis is used for cattle-feed because it is rich in vitamins A and E. in Japan *pelvetia* is used as a cow- feed. Hens which feed on *Ascophyllum* –meal and *Fucus*- meal produce eggs with increased iodine content.

3. Algal role in fisheries: Various algae have utilized in fish culture. The members of planktonic and periphytic algae serve as primary food for fishes and other aquatic animals. Species of *oedogonium, Microspora, Ulothrix, Spirogyra, Cladophora,* diatoms, etc. sever as fish food in freshwater systems. It has been estimated that about a hundred Kg of algae produce approximately one Kg of fish. As oxygen is vital for the fish life and the higher concentration of Carbone dioxide is lethal, the role of photosynthetic algae is very important in aquaculture, the removal of carbon dioxide and the release of oxygen during photosynthetic process. A fish named *Tilapia* uses only the members of Cyanophycea and Chlorophyceae as its food. Many fishes depend on their food only on diatoms.

4. Algae as nitrogen fixation: conversion of atmospheric nitrogen into nitrogenous compound i.e., nitrogen fixation is one important attribute of blue-green algae. A large number of members belonging to order Chroococcales and Nostocales have been found to perform this function.

5. Algae in soil fertility and land reclamation: The species of *Nostoc*, *Scytonema*, *Lyngbya, Anabaena,* and *cylindrospermum* grow extensively on moist soil surfaces. Many of these are fix atmospheric nitrogen and increase the soil fertility due to their mucilaginous sheath they are able to prevent soil erosion by binding the soil particles firmly. They also reduce the evaporation of water because of thick covering on the soil. Due to the presence of these algae, water holding capacity is increased by 40 % and pH is reduced from 9.7 to 7.6.

6. Algae as fertilizer: These algae increase the water holding capacity besides the addition of their chemical constituent in the soil. Seaweeds, particularly brown algae improve the fertility of soil in cultured fields as their algin content helps in conditioning the soil, facilitating aeration, moisture retention and adsorption of nutrient elements. Seaweed liquid fertilizers will be useful for achieving higher agricultural production, because the extract contains growth promoting hormones (IAA and IBA), gibberellins, cytokinins, trace elements and vitamins and increased resistance to diseases upon treatment in various crops have been reported, among them, Fucus spp., Padina spp., Laminaria spp., Sargassum spp., and Turbinaria spp. are used as biofertilizers in agriculture**..** They are either mixed with some other organic materials or are allowed to rot in the field as such.

Fucus is used as common manure by Irish people. Genera like *Lithophyllum*, *Lithothamnion* and *Chara* are used in the deficiency of calcium in the field.

A 30% increase in the total production of rice grains was reported by algologists at Central Rice Research Institute, Cuttack, when the rice fields were inoculated by some nitrogen –fixing blue- green algae.

7. Algae as oxygen donors and link of food Chain: Algae oxygenate the environment by photosynthesis. They are primary link of many diverse food chains. Aquatic animals depend ultimately for food on algae.

8. Algae in sewage treatment: In sewage oxidation tanks, presences of algae oxygenate the sewage to a great extent. The released oxygen is utilized by bacteria in rapid decomposition of the sewage. Algae used in sewage ponds mainly belong to Chlorococcales, Volvocales (Chlorophyceae) and Euglenophyceae. The common members are *Chlorella, Scenedesmus, Pediastrum Euglena; Phacus* etc. algae of sewage disposal ponds utilize nutrients to breakdown sewage.

9. Algae as research material: *Chlorella* culturing tanks are used in space vehicles because the alga restores the oxygen by it photosynthesis. Algae like *Chlamydomonas, Chlorella* etc. are very much useful in physiological, cytological and genetical studies. Species of *Nitella* are used in the studies of cytoplasmic movement, ion accumulation etc. *Valonia* and *Halicystis* help in understanding the phenomenon of permeability.

10. Algae and medicinal use: Brown algae are used in various goiter medicines due to their high iodine content. *Sargassum* which is used against goiter and other glandular troubles. Insect diseases to humans are treated with extract from *Corallina, Digenia, Codium, Alsidium* and *Durvillea*. Fucoidin and compounds of laminarin are used as anticoagulant while carrageenin acts as blood coagulant. Algae are used in the treatment of kidney, bladder and lung disease in China and Japana. *Gelidium* is used in stomach disorders and in heat induced illness.

Antibiotic chlorellin is extracted from *Chlorella vulgaris* which inhibits the growth of certain bacteria and a few algae. The growth of Escherichia coli (E. coli) is found to be reduced by *Nitzschia palea* (diatom). Microcystis reveals inhibitory action to *Staphylococcus, Closteridium* and zooplanktons like *Cyclops* and *Daphnia*.

11. Algae as source of growth promoting substances: Algae have been to increase the yield even when the soils are well manured. Seed treatments with *Phormidium* results in profuse tillering, increased height and multiple rice yields. There is also an increase in protein contents of grains of treated plants. *Phormidium* is not a nitrogen-fixing alga.

12. Algae in uptake of heavy metals and radioactive wastes: There are important role of algae in absorbing radioactive wastes and heavy metals. Species of *Chlorella, Euglena, Spirogyra, Cladophora, Scenedesmus* and *porphyra* have been found to absorb the radioactive elements and heavy metals.

13. Lens paper: For cleaning optical articles, lens paper is produced from *Spirogyra*.

14. Algae in the origin of petroleum and gas: The plankton of the seas is probably of great importance as a source of organic matter. Organic compounds produced from photosynthesis get accumulated in bottom deposits and in an oxygen-free environment gradually converted into oil and gas. Natural gas (largely methane) can be produced by certain kinds of anaerobic bacteria and oil formation is also associated with this.

15. Algae and limestone formation: Some members of green algae, blue green, red algae and flagellates deposit calcium carbonate on their cell wall or gelatinous sheath both in fresh and salt water.

Algae also play an important role in formation of beds of limestone rocks.

16. Algae as indicator to the source of water : Can identify the source of the water potential of a sample of surface water, by selecting the numbers and types of algae and associated in this sample, this is possible because the numbers and types of micro-organisms that may be established linked to the quality of water sources where they live. For example, green algae are more prevalent in the open lakes of fresh water while hosting the blue-green algae in lakes closed.

17. The role of Algae in industries: Many commercial products are extracted from algae and their cell walls. Here only brief accounts of these are given.

a) Alginates (Alginic acid): derived from cellulose free middle lamella and primary wall of the members of phaeophyceae like *Macrocystic, Laminaria, Ascophyllum* ,*Lessonia* etc. Alginic acid content varies with the location, seasones, temperature and parts of the plants. Its content approximately 30-40% in brown algae on dry weight. It is similar to cellulose and pectic acids in composition consisting of a long unbranched chain of β-d-mannuronic acid joined by 1:4 glycosidic linkages. The soluble calcium salt of alganic acid is algin.

As algin has remarkable water absorbing capacity it is used in many industries where there is the requirement of thickening, suspending, emulsifying, gel-forming, and stabilizing. Sodium salt of algainic acid is used in textile industry as they form excellent polishing and dressing material. Alginates are used also in food industry for filing creams, thickening soup, sauces, in cosmetics industry as dispensing agents in ointments, creams tooth pastes, shampoos, in paint industry for suspension of pigments, stabilization of emulsion; in pharmaceutical industry as emulsifyers and as filters in the manufacture of tablets, pills. Aginats are also used as gel in the freezing of fish, antibiotics and in the treatment of shocks.

b) Agar (Agar-agar): This is dried gel-like non-nitrogenous, gelatinous extract obtained from many red algae. This is one the best known algal products and used as a solidifying agent in the preparation of microbiological culture media. Dried agar is insoluble in cold water but soluble in hot water. The important algae used for the extraction of agar are *Gelidium, Gracilaria, Pterocladia, Gigartina, Chondrus*. Gelling property varies with the species but it will set at from 35to 52 ^{0}c. The major component of agar is agarose. Uroinc acid, pyruvic acid, polysaccharides like agarose and agaropectin are also present in agar-agar.

Besides most important use of agar in the preparation of culture media. It has also been used in food industry, cosmetics, leather, textile industry, pharmaceuticals, dental impression mold and meat packing, for clotting of blood and as emulsifyers, laxatives.

c) Carrageenan (Carrageenin): it is carbohydrate mucilage named after Irish village Carrageenin which is extracted from red alga *Chondrus crispus* and to a lesser extent from *Gigartina*. The compound is a cell wall polysaccharide complex of D-galactose-3, 6-anhydro-D-galactose and monoesterified sulphuric acid. These compounds are used like alginates in food, textile, leather, and industry, pharmaceutical and brewing industries. This gelatinous carbohydrate is variously used with pudding, consumed with milk, fruit and ice-cream. It is used as clearing agent in beer preparation.

d) Iodine and other compounds: Members of brown algae such as *Laminaria digitata* and *Fucus* spp.are known for the extraction of iodine. The maximum percentage of iodine (1.23%) has been obtained in Laminarias of Britsh Coasts. Seaweed are also known for the presence of macronutrients useful for human consumption like iron, manganese, zinc, copper etc. bromine , formic acid , acetone, acetic acid are also extracted from seaweeds. Seaweed ash is also used as source of salt and soda.

Harmful roles of algae

Besides many uses, algae are also known to create problems for human beings.

1. **Death of aquatic animals and fishes:** Many of blue green algae produce toxins which cause death to domestic animals and fishes. Important among these algae are *Microcystis, Anabeana* and *Aphanizomenon*. Besides death many harmful effects like loss of weight, weakness, abortion etc. have been reported in cattles. Phycocyanin pigment found in blue- greens is sensitive to light and when they enter in the blood capillaries of cattle cause an internal and peeling of the skin.

2. **Disease and death to human beings:** The direct consumption of dinoflagellates through fishes has resulted in human death. Dinoflagellates like *Gonyaulax*, when consumed with shell fish, produce several diseases. Reports about the occurrence of paralysis, respiratory failure and death within 2 to 12 hours after consumption of dinoflagellates are available.

3. **Algae and water supply:** algae interfere and create problems in water supply and storage reservoirs in many ways like in a) profuse growth and interference with filtration process, b) production of bad odour by their decomposition, c) imparting objectionable tastes, d) formation of slimes in water, colouration to the water, f) corrosion of pipes, g) interference in disinfection process by imparting turbidity, h) formation of sediments in service reservoirs.

The increase in the rate of excessive growth of algae to changes in physical properties, chemical and biological agents in water bodies. Where that rise to the phenomenon of the eutrophication is characterized by a high concentration of phosphates and nitrates, which lead to a significant increase in the rate of growth of some species of algae over some of the other races and species usually prevail in the unwanted presence of water bodies because of their detrimental effect on the rest of the organisms that coexist with them and the quality of the water.

The water quality is divided into a water body according to the abundance of nutrients, depending on the rate of growth of algae as follows:

a) Oligotrophic Water

It is characterized by the water that the concentration of major nutrients is rare, so do not lead to an increase in the numbers and types of algae. Featuring the region that the water is pure and transparent and the bottom of the river is relatively free from sediment, and inorganic materials such as calcium, magnesium, iron, sulfur, and dissolved half carbonate are found in small quantities.

b) Mesotrophic Water

Water containing medium concentrations of major nutrient which allows an increase of algae growth rate slightly

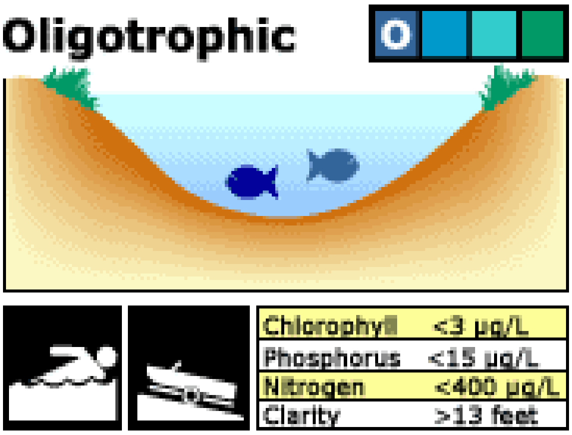
c) Eutrophic Water

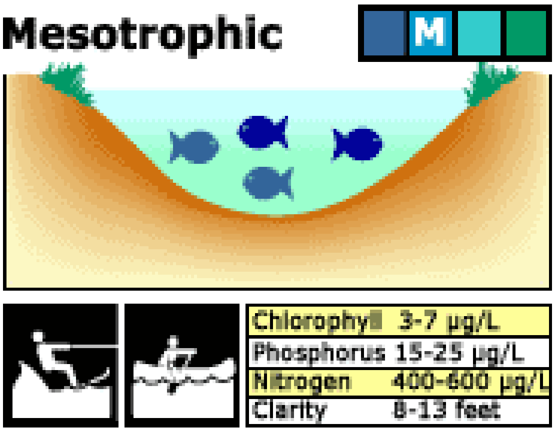
A water with a high concentration of major nutrients leading to increased algae growth rate substantially, which in turn leads to a phenomenon of the Euotrophication, which is usually accompanied by a phenomenon of algal bloom and which is characterized by a predominance of certain species of harmful algae, especially those excreted phycotoxins or algal toxins that produce certain chemicals that occur a change in the taste and smell of water.

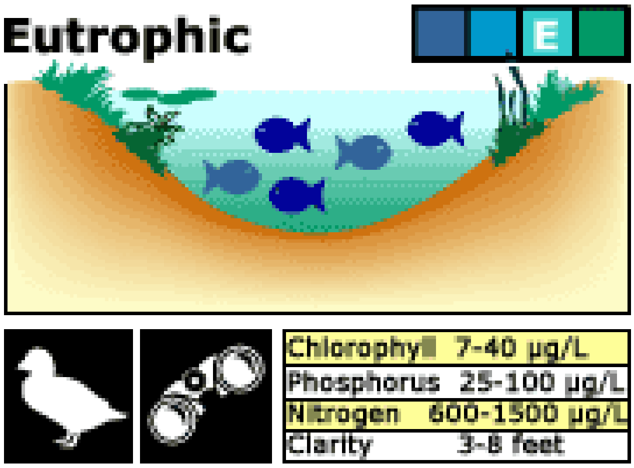
d) Hypereutrophic Water

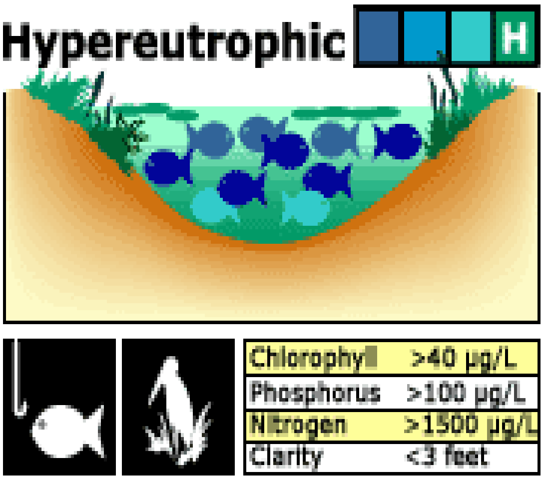
It is water that is characterized by high abundance of nutrients in the major, which in turn leads to a dramatic growth and the significant increase in the numbers and types of algae

Algae causing these problems belong to cyanophyceae (*Oscillatoria, Microcystis, Scytonema, Cylindrospermum, Chroococcus* and *Anabaena*), Chlorophyceae (*Ankistrodesmus, Chlorella, Pediastrum*), Euglenophyceae (*Euglena and Phacus*).









4. Damage to building: During rainy season, on moist wall surface many blue- green algae grow. Gradually these spoil the walls of the buildings. *Scytonema, Tolypothrix* and *Chlorococcum* are some common algae causing such damage in tropical countries.

5. Accidents due to blue-green algae: Due to intensive growth of blue-greens, ground surface becomes slippery by which cattles and human beings get slipped.

6. Parasitic algae: Many algal forms are true parasites which cause severe damage to economically important parts of the plants.

Algal bloom

An algal bloom (water bloom) occurs when the numbers of algal cells increase rapidly to reach concentrations usually high enough to be visible to the naked eye. Algal bloom concentrations may reach millions of cells per milliliter. Many types of algae form blooms. Algal blooms are often green, but they can also be yellow-brown or red, depending on the species of algae. Not all algal blooms are toxic. Some, such as the blooms of diatoms in the early spring, are very important to the health of the ecosystem.

Bright green blooms are a result of blue-green algae, which are actually bacteria (cyanobacteria). Blooms may also consist of macroalgal, not phytoplankton, species. These blooms are recognizable by large blades of algae that may wash up onto the shoreline. "Black water" is a dark discoloration of sea water, first described in the Florida Bay in January 2002.

Algae causing water blooms

Microcystis aeruginosa, M.viridis, M.flos-aquae, Anabaena circinalis, A.microspora, A.spiroides, Lyngbya limnetica, Oscillatoria planktonica, *Spirulina gomontiana, Nostoc linckia, Noduaria spumigena* (cyanophyta*), Chlamydomonas, Pandorina, Volvox, Scenedesmus, Botryococcus, Cosmarium, Zygnema, Mougeotia, Odogonium* (Chlorophyta*) and Tabellaria* are the common bloom-forming algae.

Factors of bloom formation

There is no single factor which causes an algal bloom. A large number of factors have been found to be associated with the formation of water blooms such as water temperature, water movements, light and inorganic nutrients (mainly phosphorus and nitrogen). When phosphates are introduced into water systems, higher concentrations cause increased growth of algae and plants. Algae tend to grow very quickly under high nutrient availability, but each alga is short-lived, and the result is a high concentration of dead organic matter which starts to decay. The decay process consumes dissolved oxygen in the water, resulting in hypoxic conditions. Without sufficient dissolved oxygen in the water, animals and plants may die off in large numbers. Excess carbon and nitrogen have also been suspected as causes, although a study suggested that this is not the case. Major elements and heavy metals like zinc play an important role in the bloom formation. Blooms of dinoflagellates are related to cobalamine begin washed out in the sea from the soils which posses sufficient amount of this vitamin. As the planktonic blue-green algae have floating device (gas vacuoles), blooms of the members of this class are mostly of permanent nature. Within a few days, a bloom can cause clear water to become cloudy. The blooms usually float to the surface and can be many inches thick, especially near the shoreline. Cyanobacterial blooms can form in warm, slow-moving waters that are rich in nutrients such as fertilizer runoff or septic tank overflows. Blooms can occur at any time, but most often occur in late summer. They can occur in marine and fresh waters, but the blooms of greatest concern are the ones that occur in fresh water, such as drinking water reservoirs or recreational waters.

Measurement

Algal blooms are monitored using biomass measurements coupled with the examination of species present. A widely-used measure of algal and cyanobacterial biomass is the chlorophyll concentration. Peak values of chlorophyll a for an oligotrophic lake are about 1-10 µg/l, while in a eutrophic lake they can reach 300 µg/l. In cases of hypereutrophy, such as Hartbeespoort Dam in South Africa, maxima of chlorophyll a can be as high as 3,000 µg/l.

Harmful effects

HAB stands for harmful algal bloom. There are many species of single-celled organisms living in the oceans, including algae and dinoflagellates. When certain conditions are present, such as high nutrient or light levels, these organisms can reproduce rapidly. This dense population of algae is called a bloom. Some of these blooms are harmless, but when the blooming organisms contain toxins, other noxious chemicals, or pathogens it is known as a harmful algal bloom (HAB). Of particular note are harmful algal blooms (HABs), which are marine algal bloom events involving toxic phytoplankton such as dinoflagellates of genus Alexandrium and Karenia. Such blooms often take on a red or brown hue and are known colloquially as red tides.

Examples of common harmful effects of HABs include:

1. The production of neurotoxins which cause mass mortalities in fish, seabirds and marine mammals .
2. Human illness or death via consumption of seafood contaminated by toxic algae.
3. Mechanical damage to other organisms, such as disruption of epithelial gill tissues in fish, resulting in asphyxiation.
4. Oxygen depletion of the water column (hypoxia or anoxia) from cellular respiration and bacterial degradation.

Mechanism of algal bloom causing fish mortalities

These algae like *Microcystis* and *Anabaena* usually have gas vacuoles which expand in warm water and rise to the surface. The dense plankton in surface layers absorbs and reflects the sunlight causing an inadequate penetration of light for photosynthesis. Due to fall in light intensity, oxygen concentration decrease to 1ppm at depth. After few weeks when there is no oxygen or insufficient oxygen in the greater depth, fish moves into the upper water where oxygen is sufficient. But when the oxygen deficient water up wells to the surface, fish of all types are trapped and killed.

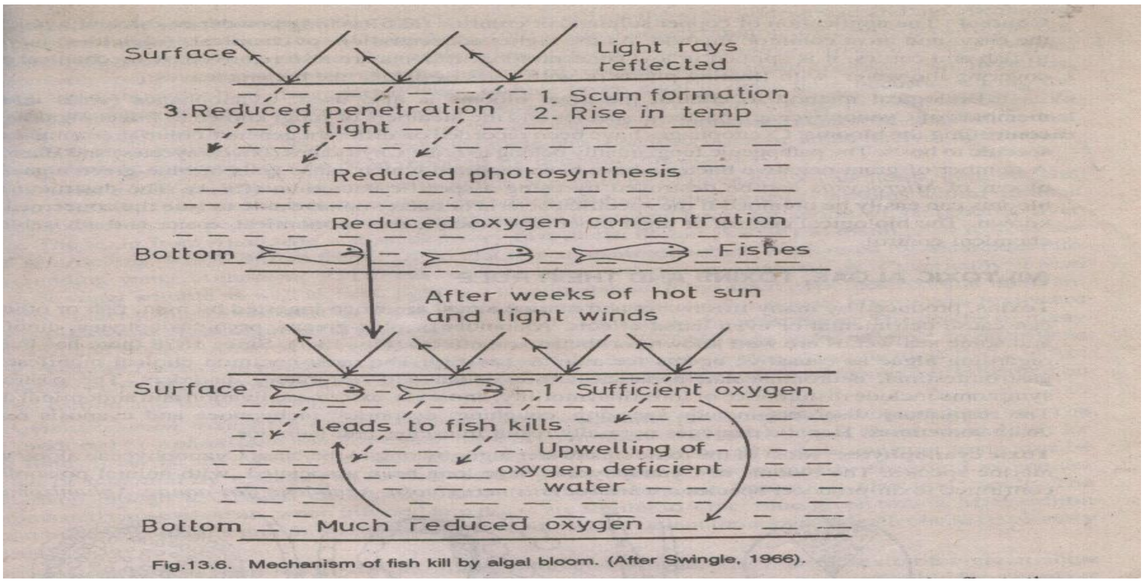


Fig.13.6. Mechanism of fish kill by algal bloom. (After Swingle, 1966).

If the HAB event results in a high enough concentration of algae the water may become discoloured or murky, varying in colour from purple to almost pink, normally being red or green. Not all algal blooms are dense enough to cause water discolouration.

Red tides

"Red tide" is a term often used to describe HABs in marine coastal areas, as the dinoflagellate species involved in HABs are often red or brown, and tint the sea water to a reddish colour. The more correct and preferred term in use is harmful algal bloom, because:

1. these blooms are not associated with tides
2. not all algal blooms cause reddish discoloration of water
3. not all algal blooms are harmful, even those involving red discoloration

Control of blooms

1. The chemical method: the application of copper sulphate or chlorine (as bleaching powder or calcium hypochlorite) is the easy and most common method. As the higher concentration of chemicals (algicides) may be lethal to fish and cattles, it is applied in low concentration.

2. Mechanical methods: by covering the water with floating plants or with parts of plants like banana leaves.

3. Biological methods: by used Cyanophages (virus infecting the members of Cyanophyceae), bacterial species and the members of fungi known to infect algae are used for controlling the blooms. The pathogenic fungi (Phycomycetes) and a number of gram negative bacteria have been reported which cause lysis of blue-green algae. The algal blooms of *Microcystis* can be destroyed by using a specific bacterium (CP-1). The biological control of algal blooms is much more economical, easier and advisable than the chemical control.

Water treatment

Algal blooms sometimes occur in drinking water supplies. In such cases, toxins from the bloom can survive standard water purifying treatments. Researchers at Florida International University in Miami are experimenting with using 640-kilohertz ultrasound waves that create micropressure zones as hot as 3,700 °C. This breaks some water molecules into reactive fragments that can kill algae.

Toxic Algae

Death and sickness to pets, livestock, wildlife and even man have been attributed to the presence of certain algae, mostly blue-green forming species, in water supplies. Lethal substances produced by these algae are retained within the cells and released after death or are secreted from living cells. Many unattended farm ponds and other waters contain some of these toxic forms, posing a threat to human health and the environment. Medical case histories, biologist reports and laboratory tests show some of the Possible effects of toxic algae.

Humans: Exposure to and ingestion of algae caused a variety of "discomforts" including: skin rashes, headaches, nausea, vomiting, diarrhea, fever, muscular pains and eye, nose and throat irritation.

Livestock: There have been reports of rapid deaths of a great variety of animals after drinking water containing high concentrations of blue-green algae such as *Microcystis*, *Aphanizomenon*, *Nostoc, Rivularia, Nodularia*, and *Anabaena*. Fatal poisonings have occurred among cattle, pigs, sheep, dogs, horses, turkeys, ducks, geese and chickens.
It is believed that such algae may be toxic to all warm-blooded animals.

Cyanotoxins

Cyanotoxins: Cyanotoxins are toxins produced by cyanobacteria.

Cyanobacterial Toxins and Symptoms:

Some cyanobacteria produce toxins that can affect animals and humans. These toxins have the potential to affect the liver, the nervous system, or exposed skin. In order to be affected, people, pets or wildlife have to be exposed to the toxin by drinking or playing in water with a toxic bloom.

Risks to humans and animals:

The toxins can damage the liver and neurological system of both humans and animals and in severe cases can cause death. Can cause gastrointestinal, skin, eye and respiratory irritations to humans and animals.

Chemical structure:

The chemical structure of cyanotoxins falls into three broad groups:

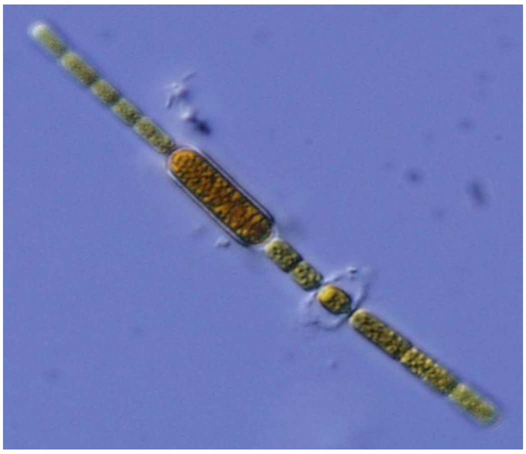
1. **Cyclic peptides**
2. **Alkaloids**
3. **Lipopolysaccharides**

Chemical structure of cyanotoxins			
Structure	**Cyanotoxin**	**Primary target organ in mammals**	**Cyanobacteria genera**

Cyclic peptides	**Microcystins**	**(Hepatotoxins)** **Liver**	***Microcystis, Anabaena, Oscillatoria (Planktothrix), Nostoc, Hapalosiphon, Anabaenopsis***
	Nodularins	**Liver**	***Nodularia***
Alkaloids	**Anatoxin-a**	**(Neurotoxins)** **Nerve synapse**	***Anabaena,*** ***Oscillatoria (Planktothrix), Aphanizomenon***
	Anatoxin-a(S)	**Nerve synapse**	***Anabaena***
	Saxitoxins	**Nerve axons**	***Anabaena, Aphanizomenon, Lyngbya, Cylindrospermopsis***
	Aplysiatoxins	**Skin**	***Lyngbya, Schizothrix, Oscillatoria (Planktothrix)***
	Cylindrospermopsins	**Liver**	***Cylindrospermopsis, Aphanizomenon, Umezakia***
	Lyngbyatoxin-a	**Skin, gastro-intestinal tract**	***Lyngbya***
Lipopolysaccharides		**Potential irritant; affects any**	**All**

		exposed tissue	

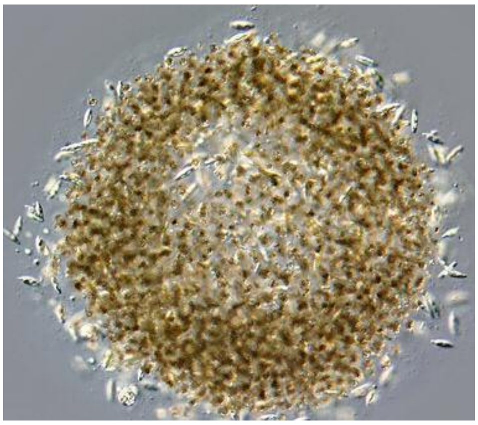
***Anabaena* spp.**



***Aphanizomenon* spp.**

Cylindrospermopsis

***Lyngbya* spp.**



***Microcystis* spp.**

Nodularia

Nostoc

Oscillatoria/Planktothrix

1. **Cyclic peptides**: A peptide is a short polymer of amino acids linked by peptide bonds. They have the same chemical structure as proteins, except they are shorter. In a cyclic peptide the links link back to the start to form a stable circular chain. In mammals this stability makes them resistant to the process of digestion and they can bioaccumulate in the liver. Of all the cyanotoxins, the cyclic peptides are of most concern to human health. The microcystins and nodularins poison the liver, and exposure to high doses can cause death.

A) **Microcystins**: Microcystins were named after the first organism discovered to produce them, *Microcystis aeruginosa*. Blooms containing microcystin are a problem worldwide in freshwater ecosystems. Microcystins are cyclic peptides and can be very toxic for plants, animals and humans. They bioaccumulate in the liver of fish, in the hepatopancreas of molluscs, and in zooplankton. They are hepatotoxic and can cause serious damage to the liver in humans.

Microcystin toxicosis may include:

- jaundice
- abdominal pain/distention
- weakness
- nausea
- vomiting
- severe thirst
- rapid/weak pulse

- death.

B) Nodularins: The first nodularin variant to be identified was nodularin-R, produced by the cyanobacterium *Nodularia spumigena*. Most common toxins present in cyanobacterial blooms in fresh and brackish waters are the cyclic peptide toxins of the nodularin family. Nodularins are potent hepatotoxins and can cause serious damage to the liver. They present health risks for wild and domestic animals as well as humans.

2. Alkaloids: Alkaloids are a group of naturally occurring chemical compounds which mostly contain basic nitrogen atoms. They are produced by a large variety of organisms, including cyanobacteria. Alkaloids act on diverse metabolic systems in humans and other animals, often with toxic effects. Almost uniformly, they are bitter tasting.

A) Anatoxin-*a*: also known as "Very Fast Death Factor‟ When it was first discovered, the toxin was called the Very Fast Death Factor (VFDF) because when it was injected into the body cavity of mice it induced tremors, paralysis and death within a few minutes.

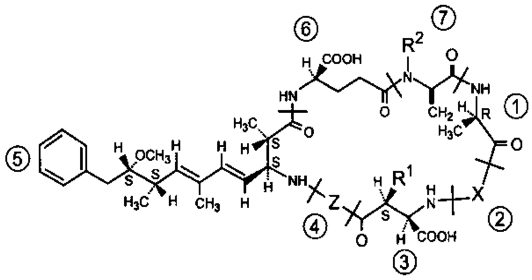
Toxic effects from anatoxin-a progress very rapidly because it acts directly on the nerve cells (neurons) as a neurotoxin. The progressive symptoms of anatoxin-a exposure are loss of coordination, twitching, convulsions and rapid death by respiratory paralysis.

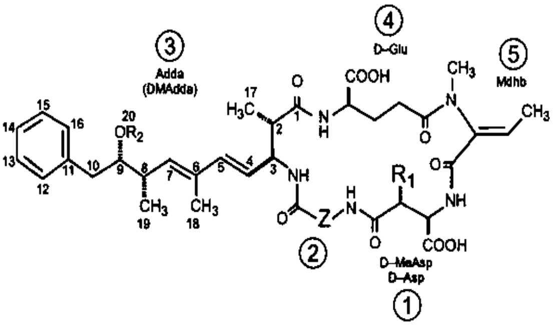
B) Cylindrospermopsins: (abbreviated to CYN or CYL).

Cylindrospermopsin was isolated from the cyanobacterium *Cylindrospermopsis raciborskii.* Cylindrospermopsin is toxic to liver, kidney, heart and other organs and is thought to inhibit protein synthesis and to covalently modify DNA and/or RNA. Symptoms of liver toxicosis include nausea, vomiting, and acute liver failure.

C) **Saxitoxins:** Saxitoxin (STX) is one of the most potent natural neurotoxins known. The term saxitoxin originates from the species name Saxidomus giganteus, whereby it was first recognized. Saxitoxin is produced by the cyanobacteria *Anabaena* sp., some *Aphanizomenon* sp., *Cylindrospermopsis* sp., *Lyngbya* sp. and *Planktothrix* sp. Saxitoxins bioaccumulate in shellfish and certain finfish. Ingestion of saxitoxin, usually through shellfish contaminated by toxic algal blooms, can result in paralytic shellfish poisoning. Death often occurs from respiratory failure.

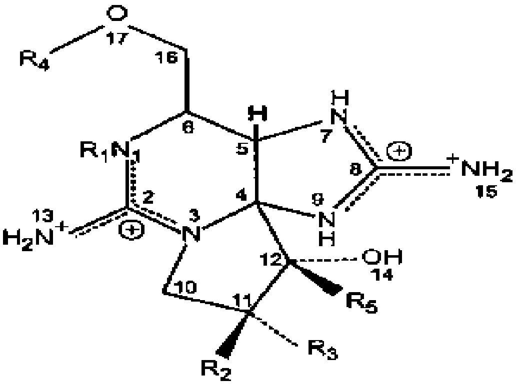
3. **Lipopolysaccharides**: Lipopolysaccharides are present in all cyanobacteria. Lipopolysachharides are less toxic than hepatotoxins or neurotoxins but are significant in terms of water supply for drinking, showering and recreation. Lipopolysacharides have been associated with outbreaks of gastroenteritis, skin and eye irritations and fever, in humans that have come into contact with algal blooms. Humans that contact Lipopolysachharides in the aerosol form (fine spray) may suffer asthma, eczema, and blisters in the lining of the nose and mouth.

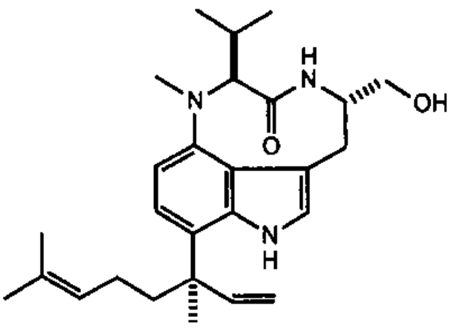




Microcystin

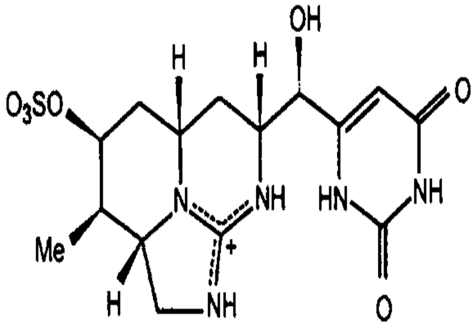
Nodularin

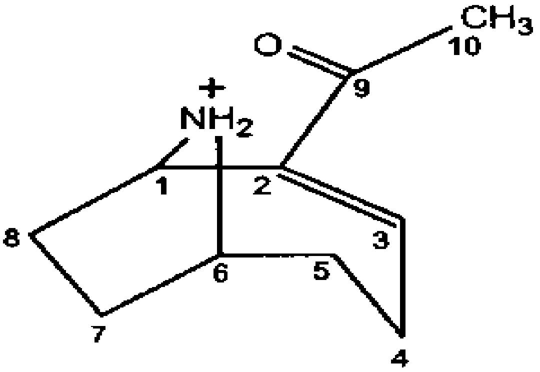




Saxitoxin

Lyngbiatoxin-*a*





Cylindrospermopsins

Anatoxins

Chemical structure of cyanotoxins

Marine algal blooms: Ocean warming has combined with nutrient enrichment to create larger, more frequent algal blooms around the world. Other environmental stresses that encourage blooms include over-harvesting of fish that feed on plankton. Marine algal blooms affect Commercial and recreational shellfish and fish harvesting, recreational swimming and diving, fish and wildlife food chain.

Toxic effects on humans:

Common human health problems caused by harmful marine algae consist of the following syndromes:

- ➢ Amnesic Shellfish Poisoning (ASP)
- ➢ Diarrhetic Shellfish Poisoning (DSP)
- ➢ Neurotoxic Shellfish Poisoning (NSP)
- ➢ Paralytic Shellfish Poisoning (PSP)
- ➢ Ciguatera Fish Poisoning (CFP)
- ✓ Each of these syndromes are caused by different species of toxic algae

Amnesic Shellfish Poisoning (ASP):

Causative organisms: *Pseudo-nitzschia*

Toxin produced: Domoic Acid

ASP is characterized by gastrointestinal and neurological disorders including short-term memory loss (Amnesia).

Gastrointestinal symptoms: Gastroenteritis usually develops within 24 hours of the consumption of toxic shellfish and symptoms include: nausea, vomiting, abdominal cramps, diarrhea.

Neurological symptoms: Neurological symptoms appear usually within 48 hours of toxic shellfish consumption. These symptoms include: dizziness, headache, seizures, disorientation, short-term memory loss, respiratory difficulty, in severe cases coma may occur followed by death.

Diarrhetic Shellfish Poisoning (DSP):

Causative organisms: *Dinophysis sp., Prorocentrum sp.*

Toxin produced: Okadaic Acid

Symptoms: DSP is characterized by gastrointestinal symptoms (nausea, diarrhea, vomiting, abdominal pain). The patients usually recover within a few days (2-3 days). Chronic exposure to DSP is suspected to promote tumor formation in the digestive system.

Neurotoxic Shellfish Poisoning (NSP):

Causative organism: *Gymnodinium breve*

Toxins produced: Brevetoxins

It is characterized by gastrointestinal and neurological disturbances usually with recovery within few days.

Symptoms:

Typical symptoms include:

Numbness, tingling, dizziness, fever, chills, muscle pains, nausea, diarrhea, vomiting, headache, reduced heart rate and pupil dilation, but is never fatal. In addition, formation of toxic aerosols by wave action can produce respiratory asthma-like symptoms.

Paralytic Shellfish Poisoning (PSP):

Causative organisms: *Alexandrium sp., Gymnodinium sp., Pyrodinium sp.*

Toxins produced: Saxitoxins

Symptoms: Symptoms include tingling and numbness of the mouth, tongue and extremities, nausea and vomiting. In severe cases the patient will exhibit more severe neurological symptoms such as ataxia, weakness, dizziness, followed by complete paralysis. The most severe cases result in respiratory arrest within 24 hours of consumption of the toxic shellfish. Death occurs due to respiratory paralysis.

Ciguatera Fish Poisoning (CFP):

Causative organisms: *Gambierdiscus toxicus* (benthic dinoflagellates).

Toxins produced: Ciguatoxin

Symptoms: Ciguatera produces gastrointestinal, neurological and cardiovascular disturbances, and recovery often takes months or even years.

Gastrointestinal symptoms include: Typical symptoms are diarrhea, abdominal pain, nausea, vomiting and followed by lots of neurological signs.

Neurological symptoms include: reversal of temperature sensation (cold objects feel hot), numbness and tingling of hands and feet, dizziness, anxiety, chills, sweating, balance difficulties, rashes.

Cardiovascular symptoms: low heart rate and blood pressure, it can very rarely cause death in humans. Extreme cases can cause death from respiratory failure.

With my best wishes

Dr. Abla A. M. Farghl

Plant Biochemistry

Plant biochemistry is the study of the biochemistry of autotrophic organisms such as photosynthesis and other plant-specific biochemical processes.

The plants and higher fungi produce through their metabolism a vast variety of chemical substances. These are important for the plant itself, but also for the environment and the recovery and use by humans. Plant Biochemistry deals with the biochemical processes of plant metabolism. The entirety of the vital processes of plants is also known as plant physiology.

Plant biochemistry is therefore a branch of Biochemistry.

A variety of chemical aspects as well as other disciplines play in this field a role, for example, natural products chemistry, phytochemistry, the chemistry of pesticides, the chemistry of vegetable dyes and more.

Chemistry of natural products deals with the chemistry of metabolites. Metabolites are naturally occurring organic compounds synthesized by plants, through metabolic activities in plants, aided by enzymes. There are two types of metabolites, viz: (a) Primary metabolites such as carbohydrates, proteins, fatty acids and glycerol, mevalonic acids, etc. (b) Secondary metabolites such as steroids, Alkaloids, Triterpenes, tannins, saponins, flavonoids, etc.

The Chemistry of natural products is dated far back to the early century. An aspect of Natural Products that deals with plants is known as Phytochemistry.

Natural products are defined as organic compounds and other chemicals synthesized by plants through metabolic processes aided by sunlight, involving CO2, H2O vapour and chlorophyll. Generally, natural products are characterized by specific functions they perform in plants and animals. Categories of natural Products are called metabolites. Primary metabolites are usually found in all living organisms such as plants and animals. They form the fundamental building block of living material e.g. mevalonic acids and nucleotides. Primary metabolites have a wide distribution in living systems and are usually involved in essential life processes. However, secondary metabolites are chemicals synthesized by plants but are not directly used by them, but are used indirectly by man as a source of pharmaceutical preparations. Secondary metabolites are generally built from primary metabolites. Secondary metabolites have restricted distribution and are characteristics of individual genera or species. Secondary metabolites are essential to the existence of the organism but play an important role to the survival of the plant. Reports have shown the presence of secondary metabolites in yeast and microbes. All living organisms from the simplest protozoan to the most developed animals contain a wide range of organic compounds. Millions of secondary metabolites have been isolated from plants and animals, some of which are known to have medicinal features. Examples are the popular Quinine-an antimalarial drug and Chloroquine isolated from medicinal plants. It is good to note that natural products are restricted specifically to major organic compounds obtained from natural origin, especially from marine plants and plants growing on the land. Plants are generally classified based on the family they belong. Each family of the plant contains a GENUS and SPECIES. Leguminoseae is a family of pant of the genus Berlinia and several species of Berlinia exists; such as *B. confusa*, *B. grandiflora*, *B. auriculata*, etc.

In the present days, millions of secondary metabolites are in existence; and majorities are isolated from natural sources such as plants. Examples are steroids, terpenoids, alkaloids, glycosides, flavonoids, etc. However, some secondary metabolites have varying structures within a group or a specific specie e.g. monoterpenoids, diterpenoids, triterpenoids, sesquiterpenoids, etc. Some are acyclic while others are cyclic in their structures. Some secondary metabolites are synthesized via biosynthesis and or biogenesis. Higher plants synthesised chemical compounds in vivo and degrade them by means of a series of chemical reactions, each aided by enzymes, by a process known as metabolism. The products of metabolic pathways are called metabolites.

Plant secondary metabolites, also referred to as natural products or specialized metabolites, constitutean enormously rich reservoir of chemical biodiversity.

The secondary metabolites have been the subject of research since the 1850s for their practical utility as dyes,polymers, fibers, glues, oils, waxes, flavors, fragrances, drugs, insecticides, and herbicides.

Today it is recognized that natural products have important ecological roles in plants, and their studyhas transitioned into the realm of modern biology.

In the absence of a valid distinction between primaryand secondary metabolites based on structure or biochemistry, a functional definition is used herein.

Secondary products are defined as substances thatinfluence communication between the plant and its environment, and primary products as substances that participate in nutrition and essential metabolic processes in the plant.

In comparison with primary metabolites, which are essentialto plant growth and development, secondary metabolites have internal roles in plants and also are integral to the communication of a plant with its environment.

The interaction between a plant and its environmenttakes many forms.

It can be an accumulation of pigments in flower petals, or a release of volatile chemicals by flowers to attract pollinators.

It can be the release of volatiles by a leaf damaged by a grazing caterpillar to attract predatory wasps in a tri-trophic interaction, or the production of bitter or toxic chemicals that serve as antifeedants.

It can also be the release by roots of secondary metabolites into the rhizosphere to attract beneficial soil microorganisms.

Terpenoids

There are many different classes of naturally occurring compounds. Terpenoids also form a group of naturally occurring compounds majority of which occur in plants, a few of them have also been obtained from other sources. Terpenoids are volatile substances that give plants and flowers their fragrance. They occur widely in the leaves and fruits of higher plants, conifers, citrus and eucalyptus.

The term terpene' was given to the compounds isolated from terpentine, a volatile liquid isolated from pine trees. The simpler mono and sesqui terpenes are the chief constituent of the essential oils obtained from sap and tissues of certain plants and trees. The di and tri terpenoids are not steam volatile. They are obtained from plant and tree gums and resins. Tetraterpenoids form a separate group of compounds called Carotenoids.

The term 'terpene was originally employed to describe a mixture of isomeric hydrocarbons of the molecular formula $C_{10}H_{16}$ occurring in the essential oils obtained from sap and tissue of plants, and trees. But there is a tendency to use more general term terpenoids which include hydrocarbons and their Oxygenated derivatives. However, the term terpene is being used these days by some authors to represent terpenoids.

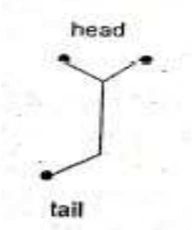
By the modern definition: "Terperioids are the hydrocarbons of plant origin of the general formula $(C_5H_8)_n$ as well as their oxygenated, hydrogenated and dehydrogenated derivatives".

Isoprene rule: Thermal decomposition of terpenoids give isoprene as one of the product. Otto Wallach pointed out that terpenoids can be built up of isoprene unit.

Isoprene rule stats that the terpenoid molecules are constructed from two or more isoprene unit.

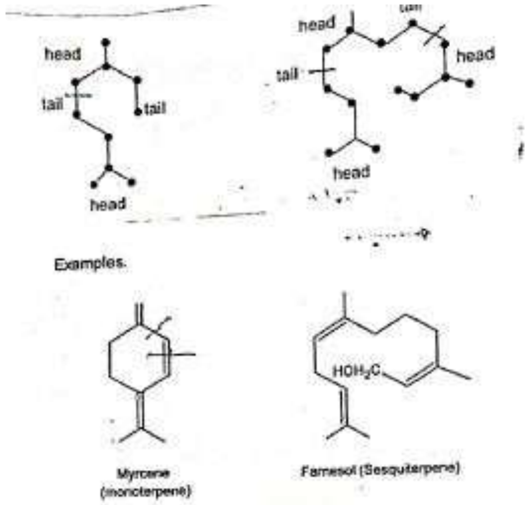
Further, Ingold suggested that isoprene units are joined in the terpenoid via a 'head to tail' fashion.

Special isoprene rule states that the terpenoid_molecule is constructed of two or more isoprene units joined in a 'head to tail' fashion.



But this rule can only be used as a guiding principle and not as a fixed rule. For example, carotenoidsare joined tail to tail at their center and there are also some terpenoids whose carbon content is not a multiple of five.

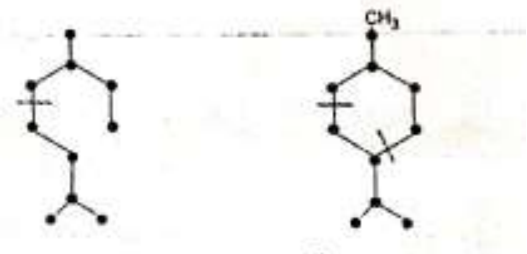
In applying the isoprene rule we look only for the skeletal unit of carbon. The carbon skeletons of open chain mono terpenoids and sesqui terpenoids are,



Ingold (1921) pointed out that a gem alkyl group affects the stability of terpenoids. He summarized these results in the form of a rule called `gem dialkyl rule' which may be stated as "Gem dialkyl group tends to render the cyclohexane ring unstable whereas it stabilizes the three, four and five member rings."

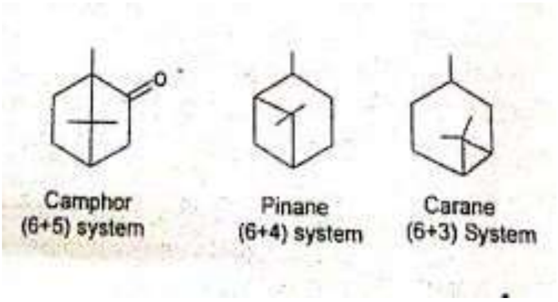
This rule limits the number of possible structures in closing the open chain to the ring structure. Thus the monoterpenoid open chain gives rise to only one possibility for a monocyclic monoterpenoid

i.e the p-cymene structure.



P-cymene structure

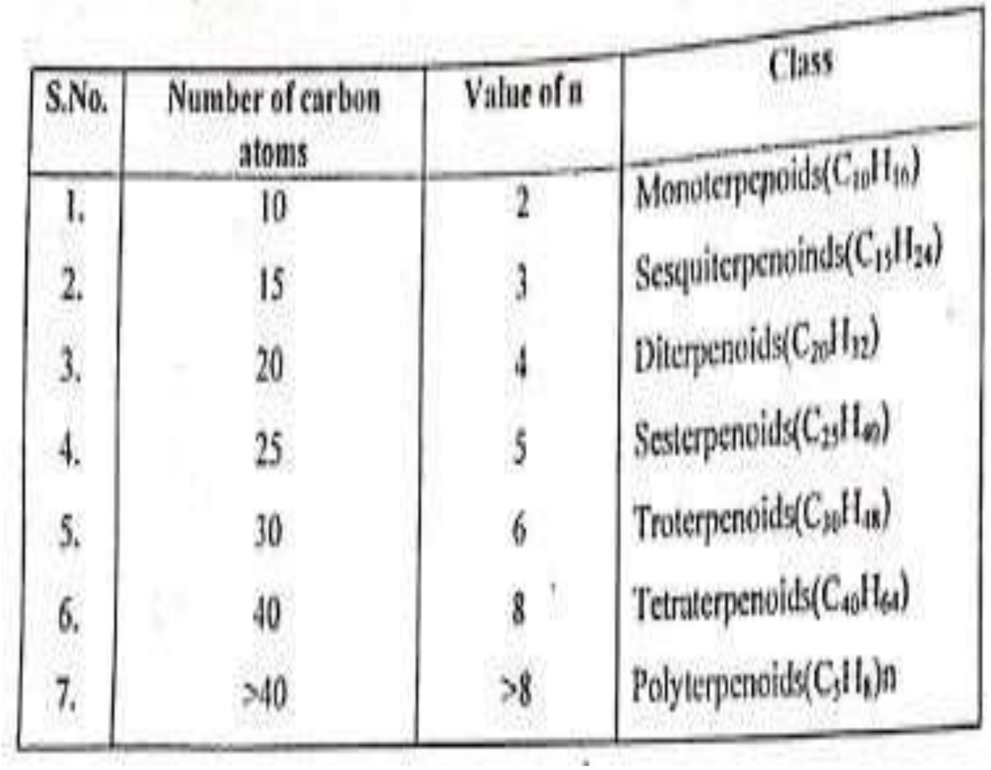
Bicyclic monoterpenoids contain a six-member and a three-member ring. Thus closure of the ten carbon open chain monoterpenoid gives three possible bicyclic structures.



Classification of Terpinoids

Most natural terpenoids hydrocarbon have the general formula $(C_5H_8)_n$. They can be classified based on the value of n or the number of carbon atoms present in the structure.

Table-1: Classification of Terpenoids



S.No.	Number of carbon atoms	Value of n	Class
1.	10	2	Monoterpepoids$(C_{10}H_{16})$
2.	15	3	Sesquiterpenoinds$(C_{15}H_{24})$
3.	20	4	Diterpenoids$(C_{20}H_{32})$
4.	25	5	Sesterpenoids$(C_{25}H_{40})$
5.	30	6	Troterpenoids$(C_{30}H_{48})$
6.	40	8	Tetraterpenoids$(C_{40}H_{64})$
7.	>40	>8	Polyterpenoids$(C_5H_8)n$

Each class can be further subdivided into subclass according to the number of rings present in the structure.

i) **Acyclic Terpcnoids**: They contain open structure.

ii**) Monocyclic Terpenoids**: They contain one ring in the structure

iii) **Bicyclic Terpenoids**: They contain two rings in the structure.

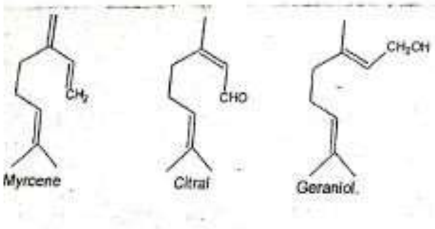
iv**) Tricyclic Terpcnoids**: They contain three rings in the structure.

v)**Tetracyclic Terpenoids:** They contain four rings in the structure.

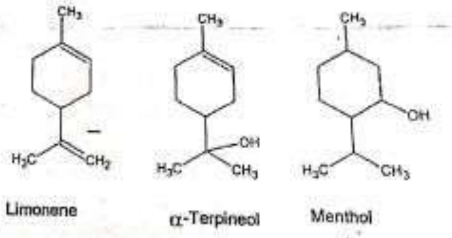
Some examples of mono, sesqui and di Terpenoids:

A) Mono terpenoids

i) Acyclic Monoterpenoids



ii) Monocyclic monoterpenoids

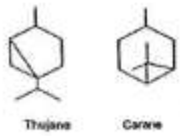


iii) **Bicyclic monoterpenoids**: These are further divided into three classes.

a) Containing -6+3-membered rings

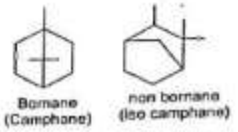
b) Containing -6+4- membered rings.

c) Contining -6+5-membered rings



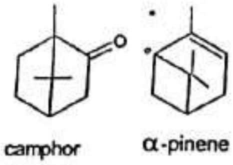
Containing -6+3-membered rings

-6+4-membered rings



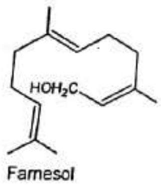
-6+5-membered rings

Some bicyclic monoterpenes are:

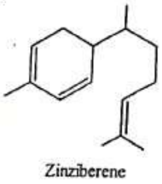


B) Sesquiterpenoids:

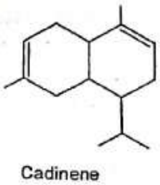
i) Acyclic sesquiterpenoids



ii) Monocyclic sesquiterpenoids

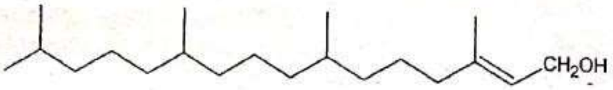


iii) Bicyclic sesquiterpenoids.



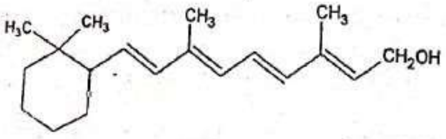
C) Diterpenoids:

i) **Acyclic diterpenoids**



Phytol

ii) **Mono cyclic diterpenoids:**



Vitamin A

Isolation of mono and sesquiterpenoids

Both mono and sesquiterpenoids have a common source i.e essential oils. Their isolation is carried out in two steps:

i) Isolation of essential oils from plant parts

ii) Separation of Terpenoids from essential oils

i) **Isolation of essentia1 oils from plant parts**: The plants having essential oils generally have the highest concentration at some particular time. Therefore better yield of essential oil plant material has to be collected at this particular time. e.g. From jasmine at sunset. there are four methods of extraction of oils.

a) Expression method

b) Steam distillation method

c) Extraction by means of volatile solvents

d) Adsorption in purified fats

Steam distillation is the most widely used method. In this method macerated plant material is steam distilled to get essential oils into the distillate form these are extracted by using pure organic volatile solvents. If the compound decomposes during steam distillation, it may be extracted with petrol at 50°C. After extraction solvent is removed under reduced pressure.

ii) **Separation of Terpenoids from essential oil**: Several terpenoids are present in essentialoil obtained from the extraction. Definite physical and chemical methods can be used for the separation of terpenoids. They are separated by fractional distillation. The terpenoid hydrocarbons distill over first followed by the oxygenated derivatives.

More recently different chromatographic techniques have been used both for the isolation and separation of terpenoids.

General properties of Terpenoids

1. Most of the terpenoids are colourless, fragrant liquids which are lighter than water and volatile with steam. A few of them are solids e.g. camphor. All are soluble in organic solvent and usually insoluble in water. Most of them are optically active.

2. They are open chain or cyclic unsaturated compounds having one or more double bonds. Consequently they undergo addition reaction with hydrogen, halogen, acids, etc. A number of *addition* products *have* antiseptic properties.

3. They undergo polymerization and dehydrogenation.
4. They are easily oxidized nearly by all the oxidizing agents. On thermal decomposition, most of the terpenoids yields isoprene as one of the product.
5.

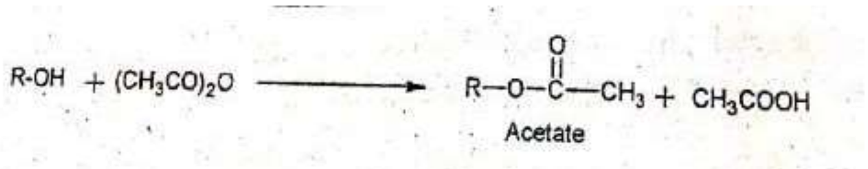
General Methods of structure elucidation

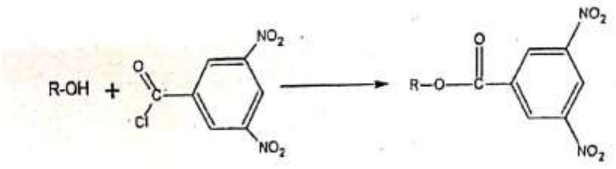
Terpenoids

1) Molecular formula: Molecular formula is determined by usual quantitative analysis and mol.wt determination methods and by means of mass spectrometry. If the terpenoid is optically active, its specific rotation can be measured.

2) Nature of oxygen atom present: If oxygen is present in terpenoids its functional nature is generally as alcohol aldehyde, ketone or carboxylic groups.

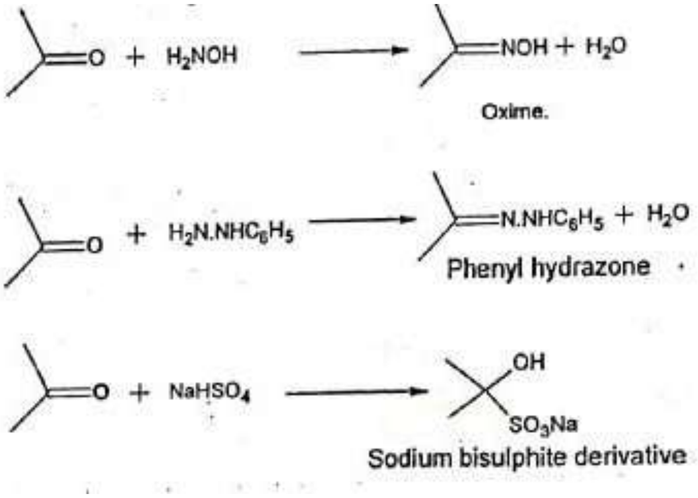
a) **Presence of oxygen atom present:** presence of —OH group can be determined by the formation of acetates with acetic anhydride and benzoyate with 3.5-dinitirobenzoyl chloride.





Primary alcoholic group undergo esterification more readily than secondary and tertiary alcohols.

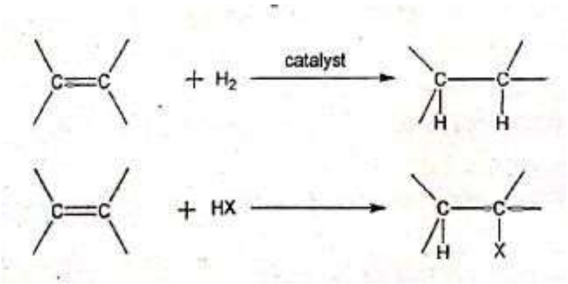
b) **Presence of >C=0 group**: Terpenoids containing carbonyl function form crystalline addition products like oxime, phenyl hydrazone and bisulphite etc.



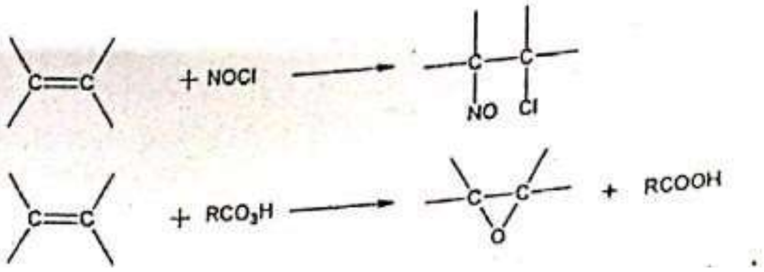
if carbonyl function is in the form of aldehyde it gives carboxylic acid on oxidation without loss of any carbon atom whereas the ketone on oxidation yields a mixture of lesser number of carbon atoms.

iii) **Unsaturation**: The presence of olefinic double bond is confirmed by means of bromine, and number of double bond determination by analysis of the bromide or by quantitative hydrogenation or by titration with monoperpthalic acid.

Presence of double bond also confirmed by means of catalytic hydrogenation or addition of halogen acids. Number of moles of HX absorbed by one molecule is equal to number of double bonds present.

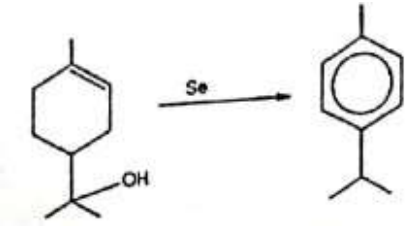


Addition of nitrosyl chloride(NOC1) **(Tilden's reagent)** and epoxide formation with peracid also gives idea about double bonds present in terpenoid molecule.

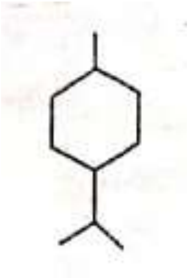


C=C
+ NOCl
C
C
NO
Cl
C=C
+ RCO3H
C
C
O
+ RCOOH

iv) **Dehydrogenation**: On dehydrogenation with sulphur, selenium, polonium or palladium terponoids converted to aromatic compounds. Examination of these products the skelton structure and position of side chain in the original terpenoids can be determined.
For example α-terpenol on Se-dehydrogenation_yields *p*-cymene.



Thus the carbon Skelton of terpenol is as follows.

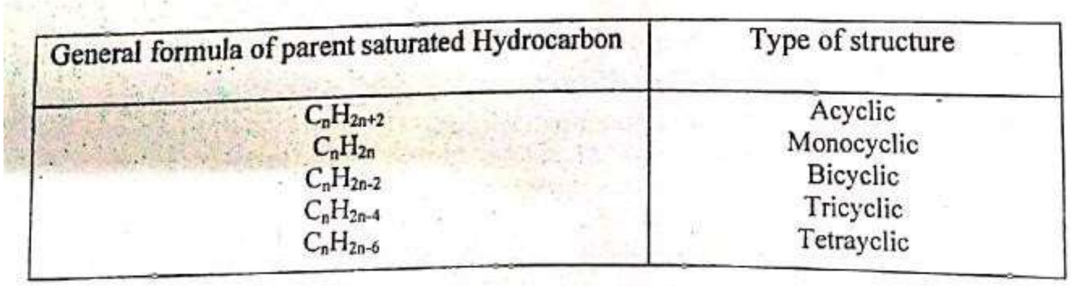


v) **Oxidative degradation**: Oxidative degradation has been the parallel tool for elucidating the structure of terpenoids. Reagents for degradative oxidation are ozone, acid, neutral or alkaline potassium permanganate, chromic acid, sodium hypobromide, osmium tetroxide, nitric acid, lead tetra acetate and peroxy acids. Since oxidizing agents are selective, depending on a particular group to be oxidized, the oxidizing agent is chosen with the help of stfucture of degradation product.

vi) **Number of the rings present**: With the help of general formula of corresponding parent saturated hydrocarbon, number of rings present in that molecule can be determined.

vii) **Relation between general formula of compound and type of compounds**: Table 2

Table-2: Relation between general formula of compound and type of compounds



General formula of parent saturated Hydrocarbon	Type of structure
C_nH_{2n+2}	Acyclic
C_nH_{2n}	Monocyclic
C_nH_{2n-2}	Bicyclic
C_nH_{2n-4}	Tricyclic
C_nH_{2n-6}	Tetrayclic

For example limonene (mol. formula. $C_{10}H_{16}$)absorbs 2 moles of hydrogen to give tetrahydro Iimonene (mol. Formula $C_{10}H_{20}$) corresponding to the general formula. C_nH_{2n}. It means limonoene has monocyclic structure.

viii) **Spectroscopic studies:** All the spectroscopic methods are very helpful for the confirmation of structure of natural terpenoids and also structure of degradation products. The various methods for elucidating the structure of terpenoids are:

a) **UV Spectroscopy**: In terpenes containing conjugated dienes or α,β–unsaturated ketones, UV spectroscopy is a very useful tool. The values of λ_{max} for various types of terpenoids have been calculated by applying Woodward's empirical rules. There is generally good agreement between calculation and observed values. Isolated double bonds, α,β-unsaturated esters, acids, lactones also have characteristic maxima.

b) **IR Spectroscopy**: IR spectroscopy is useful in detecting groups such as hydroxyl group (-3400cm^{-1}) or an oxo group (saturated 1750-1700cm^{-1}). Isopropyl group, cis and trans also have characteristic absorption peaks in IR region.

c) **NMR Spectroscopy**: This technique is useful to detect and identify double bonds, to determine the nature of end group and also the number of rings present, and also to reveal the orientation of methyl group in the relative position of double bonds.

d) **Mass Spectroscopy**: It is now being widely used as a means of elucidating structure of terpenoids. Used for determining mol. Wt., Mol. Formula, nature of functional groups present and relative positions of double bonds.

ix) **X-ray analysis**: This is very helpful technique for elucidating structure and stereochemistry of terpenoids.

x) **Synthesis**: Proposed structure is finally confirmed by synthesis. In terpenoid chemistry,many of the synthesis are ambiguous and in such cases analytical evidences are used in conjunction with the synthesis.

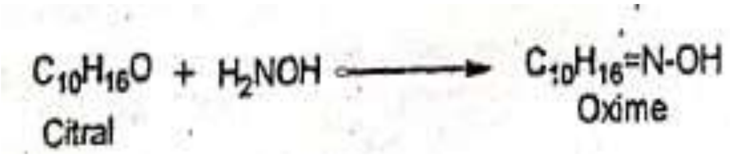
Citral

Citral is an acyclic monoterpenoid. It is a.major constituent of lemon grass oil in which it occurs to an extent of 60-80%. It is pale yellow liquid having strong lemon like odour and can be obtained by fractional distillation under reduced pressure from Lemongrass oil.

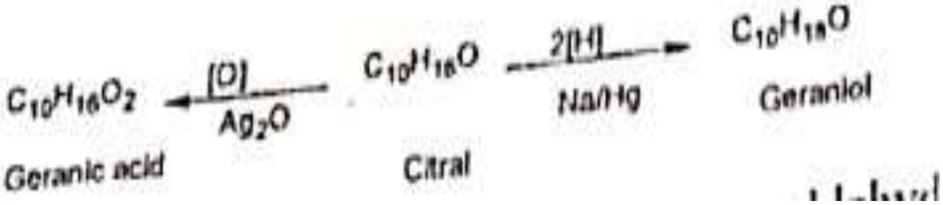
Constitution:

i) Mol. formula $C_{10}H_{16}O$, b.p-77°C

ii)Nature of Oxygen atom: Formation of oxime of citral indicates the presence of an oxo group in citral molecule.

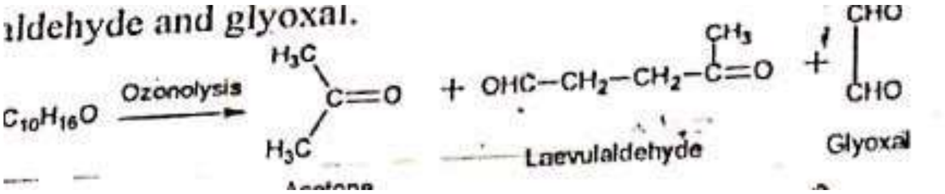


On reduction with Na/Hg it gives an alcohol called geraniol and on oxidation with silver oxide it give a monocarboxylic acid called Geranic acid without loss of any carbon atom.



Both these reactions reveal that oxo group in citral is therefore an aldehyde group. Citral reduces Fehling's solution. further confirming the presence of aldehydic group.

iii)It adds on two molecule of Br_2 showing the presence of two double bonds. On ozonlysis, it givesacetone, laevulaldehyde and glyoxal.



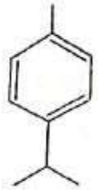
Formation of the above products shows that citral is an acyclic compound containing two double bonds. Corresponding saturated hydrocarbon of citral (mol. Formula $C_{10}H_{22}$) corresponds to the general formula C_nH_{2n+2} for acyclic compounds, indicating that citral must be an acyclic compound.

iv) Formation of *p*-cymcne and product obtained from the ozonolysis reveals that citral is formed by the joining of two isoprene units in the head to tail fashion

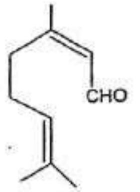
$$C_{10}H_{16}O \xrightarrow[-H_2O]{KHSO_4} \underset{\text{P-cymene}}{C_{10}H_{14}}$$

v) On the basis of above facts following structure was proposed for citral.

Carbon skelton of citral

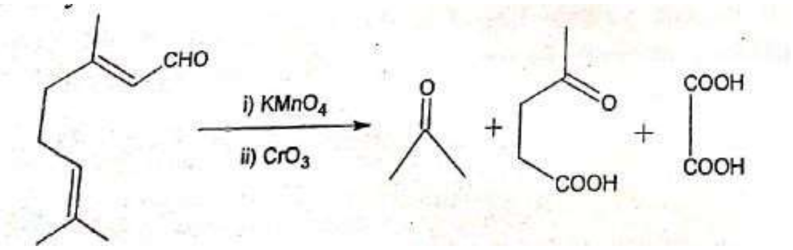


p-cymene

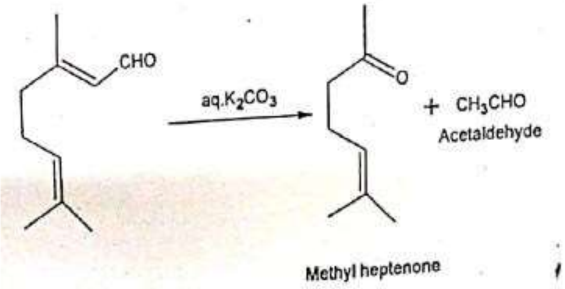


Citral

vi) Above structure was further supported by the degradation of citral on treatment with alkaline $KMnO_4$ followed by chromic acid.



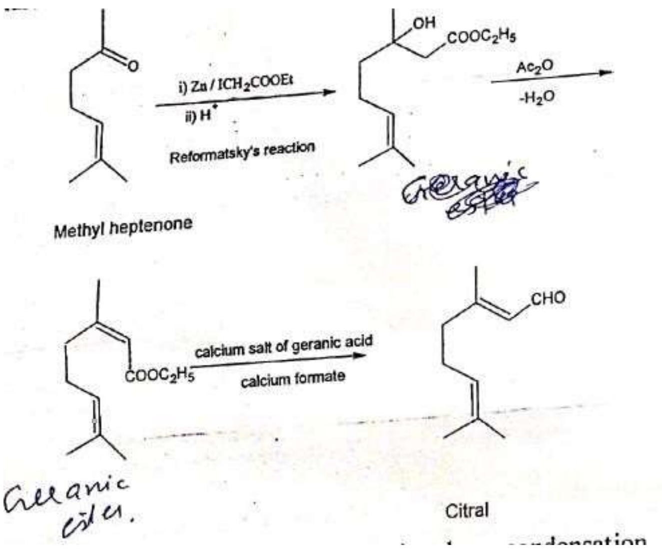
Verley found that citral on boiling with aqueous potassium carbonate yielded 6-methyl hept-5- ene-2-one and acetaldehyde. The formation of these can only be explained on the basis of proposed structure.



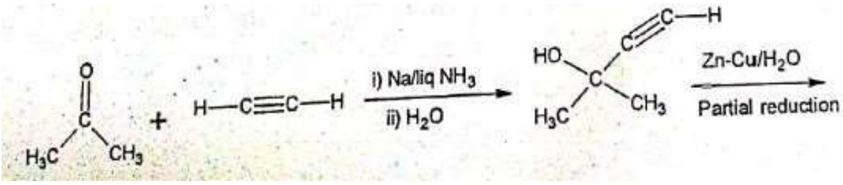
It Appears that citral is product of aldol condensation of these two.

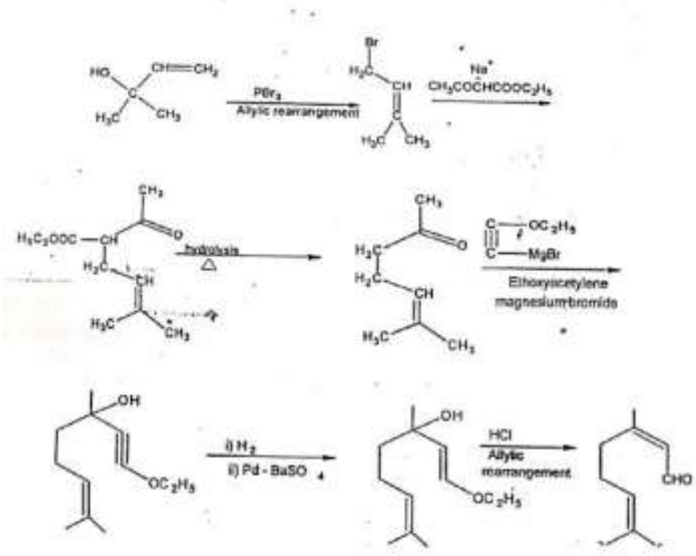
Synthesis: Finally the structure of citral was confirmed by its synthesis.

a) **Barbier-Bouveault-Tiemann's synthesis**: In this synthesis methyl heptenone is converted to geranic ester by using Reformatsky's reaction. Geranic ester is then converted to citral by distilling a mixture of calcium salts of geranic and formic acids.

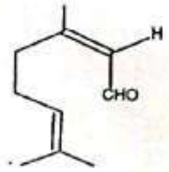


b)**Arens-Van Drop's Synthesis**: This synthesis involves condensation of acetone with acetylene in the presence of liquid ammonia. Condensation product is then reduced and treated with PBr_3, allylic rearrangement takes place. The rearranged product so obtained is treated with sodium salt of acetoacetic ester and then hydrolysed to yield methyl heptenone. The latter compound on condensation with ethoxy acetylene magnesium bromide, followed by the partial reduction and acidification yields citral by allylic rearrangement.

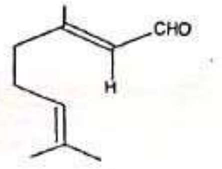




Isomerism of citral: two geometrical isomers occur in nature.



Cis-citral; citral-b: neral; Z-form

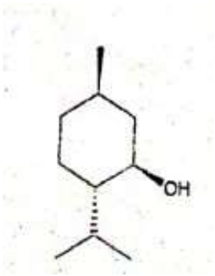


Trans-citral; Citral-a; geranial; E-form

The existence of the two isomeric Citrals in natural citral has been confirmed chemically by the formation of two different semicarbazones and formation of geraniol and nerol on reduction.

Menthol

Menthol is the major constituent of Mentha Piperi. It is used as an antiseptic and anesthetic. Menthol (also called peppermint camphor or mint camphor) is the major constituent of peppermint oil and is responsible for its odour and taste and the cooling sensation when applied to the skin. It is ingredient in cold balms. Menthol is optically active compound with mol. formula $C_{10}H_{20}O$.


OH

Alkaloids

Alkaloids are naturally-occurring organic compounds containing nitrogen moiety, and are usually heterocyclic in nature. They are nitrogen based organic compounds, with nitrogen enclosed in an heterocyclic ring.

The alkyl amines are referred to as proalkaloids.

Characteristics of alkaloids

(1) They are basic in nature due to the presence of nitrogen in their ring.

(2) They have complex structures.

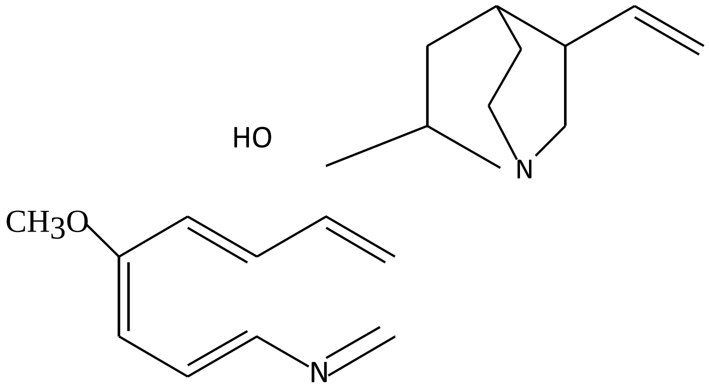
(3) They have bitter principles.

(4) They are mostly obtained from plant materials.

(5) They have high pharmacological and physiological activities.

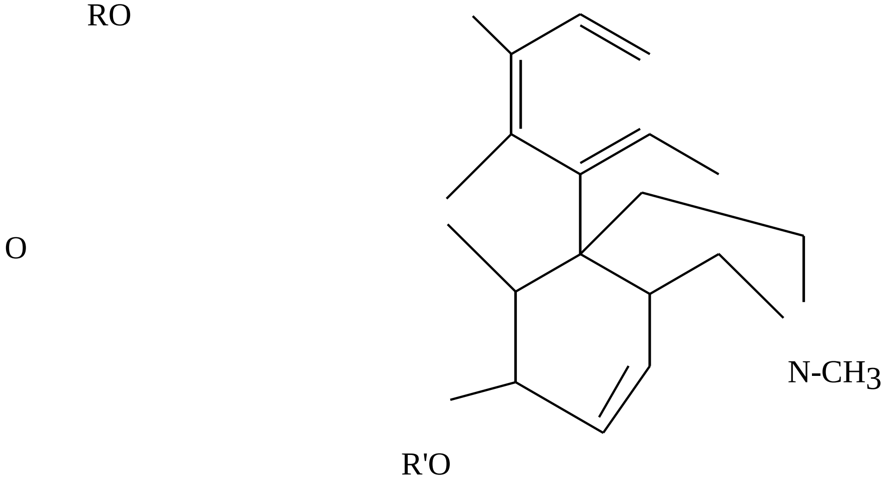
Examples of alkaloids are:

(1) **Quinine** — an antimalarial drug isolated from a plant called *Cinchonia officialis*



Quinine

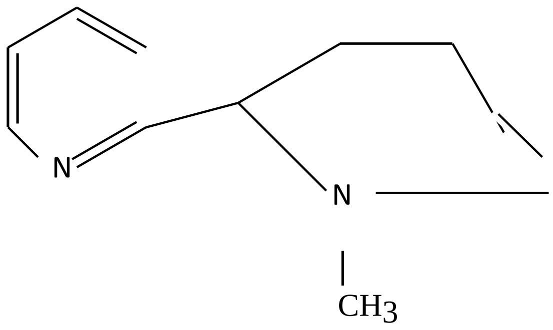
Quinine is an antipyretic alkaloid. Its molecular formular is $C_{20}H_{24}N_2O_2$..
Functional groups present in quinine are: methoxyl $–OCH_3$, hydroxyl –OH, tertiary amine group, etc.
Other examples of alkaloids are: morphine, cocaine, heroine, etc. Most are highly narcotic in nature.



R = R' = H Morphine alkaloid
R =H, R' = CH_3 Codeine
R = R' = $COCH_3$ Heroine

- Morphine is highly narcotic
- Morphine is analgesic
- Morphine is isolated from the plant *Papavera omniferous*
- Morphine is an opium alkaloid.

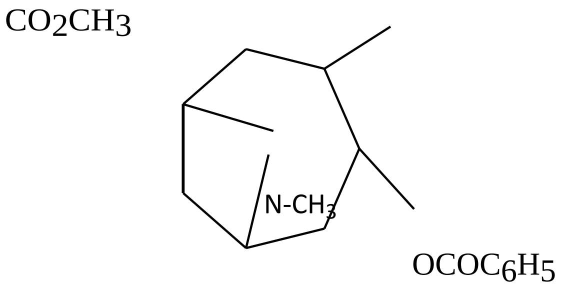
Nicotine is another example of alkaloid



Nicotine

Cocaine is an alkaloid.

- Cocaine is obtained from coca leaves,
- Cocaine is the first local unaesthetic ever discovered by man,
- Cocaine is highly narcotic,
- Cocaine stimulates the central nervous system i.e. CNS depressant,
- Cocaine can lead to psychiatric problem when taken in high dose or when addicted to it.

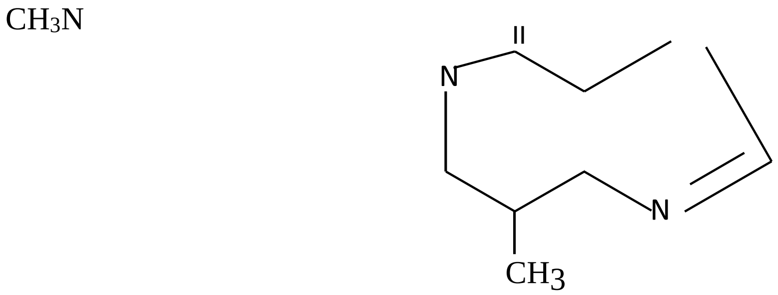


Cocaine

Caffeine is an alkaloid obtained from coffee tea. It is also a strong stimulant that can increase alertness, thereby causing insomnia when the body gets addicted. It is found in Nescafe drinks. Structure of caffeine is shown below:

H_3C

O



Caffeine

Occurrence of Alkaloids

Like other natural products, alkaloids are found in tissues of plants at point of intense cell activities, found majorly in stems, leaves and roots, seeds and barks of plants.

Alkaloids are common in some higher plants such as Rubiaceae, Rotaceae, Papaveraceae, etc. They perform certain functions in plants and animals as a result of their pharmacological activities. Pharmacologically, they acts as chelating agents, in which case they select one metal in preference to another from the soil while rejecting others.They are usually solid, though some exists in liquid form.

Nomenclature of Alkaloids

There is no systemic nomenclature for alkaloids due to the complexity in their structures; hence trivial names are often employed in the nomenclature of alkaloids. However, the names often end with **-ine** and this indicates the basic nature of the compound.

Sometimes, names of alkaloid depict the source of the alkaloid in question. An example is **Nicotine** isolated from the plant *Nicotina tobaccullum*. At times, names of alkaloids indicates the discoverer of such alkaloid or even the society or tradition where such plants originated; e.g. morphine alkaloid came from the name **Morphens** (the ancient god of Greek).

Classification of alkaloids

Alkaloids are classified into two broad classes:

(a) Classification based on the nature of the ring systems;

(b) Classification on the basis of plant sources.

Classification based on plant source

The classifications based on plant's source: in this case alkaloids are classified on the basis of the plant source such as the family and the genus. However, structural overlap may occur, using this classification. For instance, morphine alkaloid is from Apocynaceae family. Opium alkaloids such as morphine, codeine, nicotine and papaverine are derived from opium plant.

Rauwolfia alkaloids is reserpine, derived from Rauwolfia family. Reserpine is an antihypertensive alkaloid. It equally acts as a tranquilizer.Chintonia

Alkaloids: Quinine, from Cinchonine, etc.

Coca Alkaloods: from Erythroxyllum species

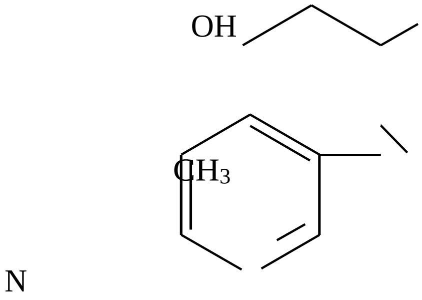
Solonaceae alkaloids: from Solanaceae family, etc.

Classification based on the Chemical structure of Alkaloids

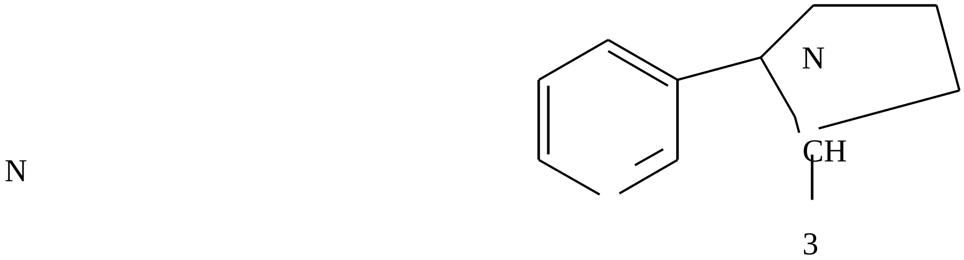
(a) **Heterocyclic** Alkaloids: pyrollidine nucleus

Pyridine nucleus,
Piperidine nucleus
Pyridine- Piperidine nucleus, etc

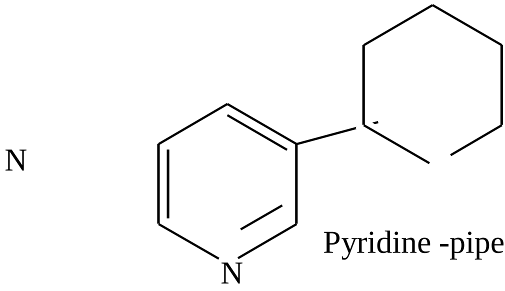
Structures of the various classification based on chemical structures are illustrated below



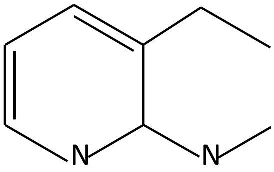
Pyridine nucleus



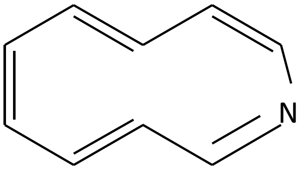
Pyridine -pyrrolidine nucleus



Pyridine -piperidine nucleus



Quinoline nucleus



IsoquinOline nucleus

Structural Elucidation of Alkaloids

Isolation of Alkaloids

In order to isolate pure alkaloids from a plant source, the procedures to be followed are as follows:

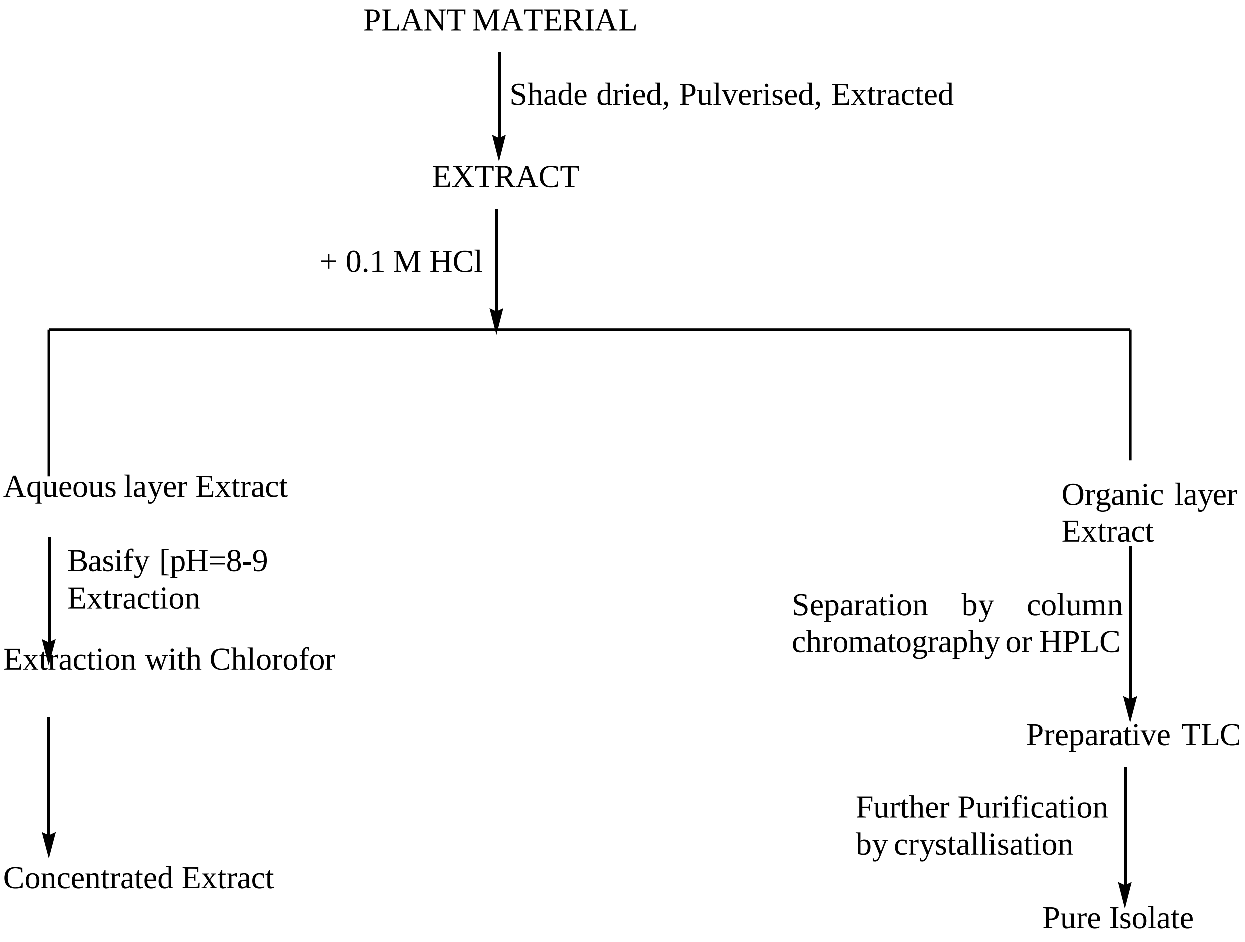
Step 1: Collection of plant material, air-drying the plant material to remove water, pulverizationof the air-dried plant material and solvent extraction protocol of the powdered plant materials.

Step 2: Purification of the alkaloid-This is done to separate alkaloid from the solution. This isdone by running chromatography on the syrupy form of the extract.

Step 3: involves the crystallization and further purification of the isolates.

Extraction method for Alkaloids

The extraction procedure often used in the isolation of alkaloids is summarized in the table below.



Physical Methods of Structural elucidation

Determination of melting point: this is to ascertain the purity of the isolate.

Elucidation of the structures: This is done by using spectroscopic technique such as Infra-Red, Ultra-violet, Nuclear Magnetic Resonance and Mass spectroscopic techniques.

Chemical Methods of Identification of Alkaloids

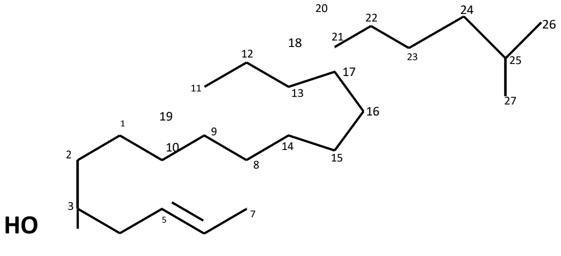
Several methods are involved.

(1) Test for functional group and Chemical transformation involving chemical tests for functional groups. Other method such as functional group interconversion is equally used for structural proof. Example is the conversion of phenol to an acetate derivative of phenol via acetylating step.

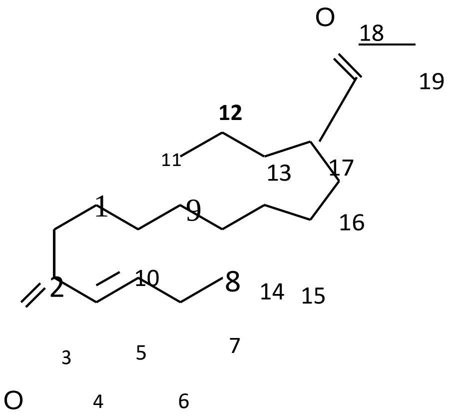
Detection of –OH group – this is done by acetylation or benzoylation or even methylation steps. Acetylation involves treatment with acetic anhydride in the presence of pyridine. Detection of phenolic compound is by treatment with Ferric chloride solution, in which a yellow precipitate confirms a phenolic compound.

Steroids

Steroids are groups of secondary metabolites found in all plants and animal tissues. They occur mostly in the brain and spinal cord in mammals. Examples of sterols are zoosterol, cholesterols, bile acids, and sex hormones such as estrogen and testosterone. Examples of phytosterols are ergosterol and stigmasterol. The most common sterol in the animal is cholesterol. Cholic acid is found in bile acid. Other common sterols are sex hormones such as oestrones and progesterone, responsible for secondary sexual characteristics.



CHOLESTEROL



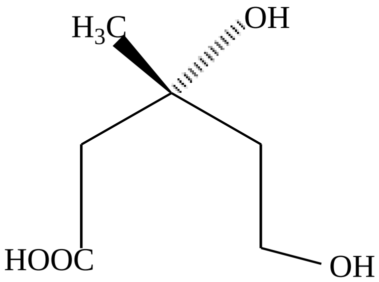
Progesterone

Cholesterol is the most common among sterols. It has a molecular formular $C_{27}H_{46}O$. An adult human being has 200-300 g of cholesterol in their body. It comprises 15% of the brain of a human. Several circulatory and heart diseases are closely associated with the level of cholesterol in the body. This is because cholesterol precipitates in the arteries thereby causing arteriosclerosis and other heart diseases. It is important because it forms the synthetic routes or starting material for other sterols.

There are eight chiral canters in cholesterol and 2^8 stereoisomers.

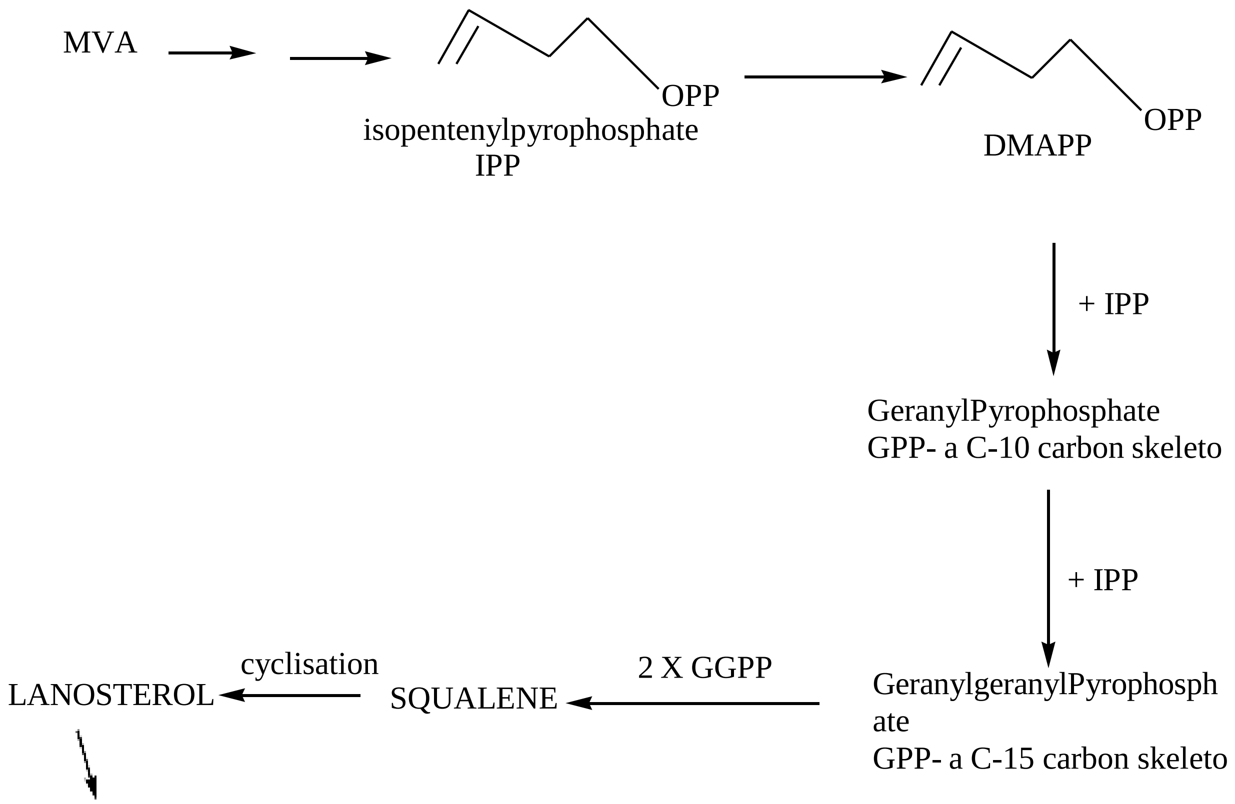
Synthesis of cholesterol

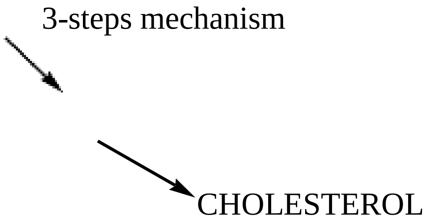
Cholesterol and other sterols are synthesized from primary metabolites such as Mevalonic acid (MVA). This is a biogenetic synthesis.



3-methyl-3,5-dihydroxylpentanoic acid [MVA]

The steps involved in the synthesis are illustrated below:




3-steps mechanism
CHOLESTEROL

Phenolics

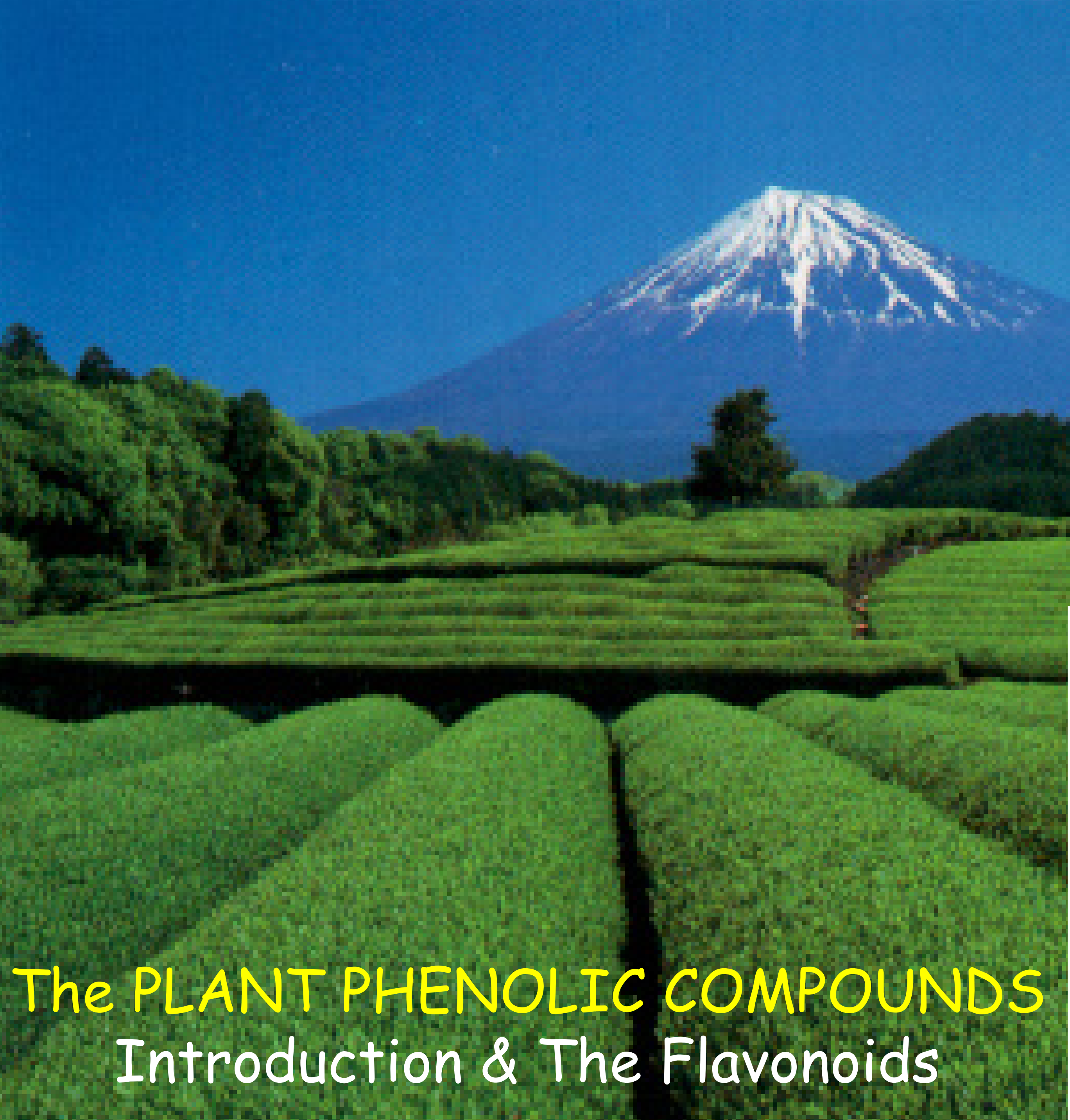
- Phenolics are a group of compounds characterized by at least **one aromatic ring bearing one or more hydroxyl groups.**
- Most of the thousands of phenolics known to date are of plant origin.
- These phenolic compounds are biosynthesized **Shikimate pathway**
- The shikimate pathway is important in plants through which many secondary plant products are synthesized.
- The **key starting materials** are **phosphoenolpyruvate (PEP) and erythrose 4P** derivedfrom glycolysis and pentose phosphate pathways, respectively.
- These two compounds condense to produce a six-carbon cyclic compound with one carbon(COOH) side chain namely **shikimate.**
- Then shikimate is phosphorylated and condensed with another molecule of PEP to produce a cyclic compound containing three carbon and one-carbon side chains.
- This is finally converted to aromatic amino acids **phenylalanine and tyrosine.**
- These amino acids are **deaminated** followed by **hydroxylation** at different carbon atoms inthe aromatic ring to form **cinnamic acid derivatives**.

- These cinnamic acid derivatives are utilized for the synthesis of different phenolic compounds.

Functions of phenolics

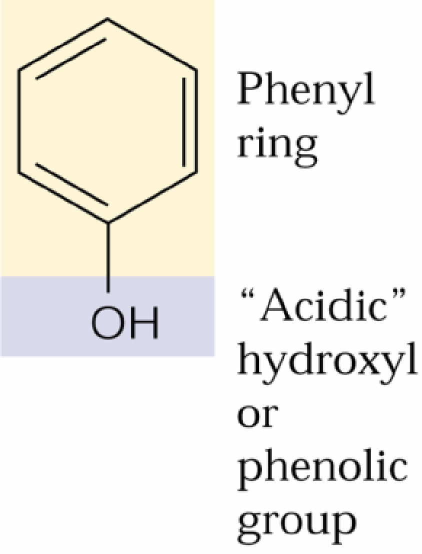
- Phenolics are of great importance as **cell wall components.**
- They form **part of cell wall structures** such as **lignins, cutins and suberins**, which provide **mechanical support and function as barriers against microbial attack**.
- F**lavonoids and anthocyanins** contribute to flower and fruit colours. This is important for attracting insects and animals to the plant for pollination and seed dispersal.
- Phenolics also play a **defensive role** in plants by protecting against predators.
- Simple phenolic acids, polyphenolics like tannins and phenolic resins at the plant surfaceare **effective feeding deterrents.**
- Phenolics are accumulated as **post-infectional low molecular compounds** called **phytoalexins** as a result of microbial attack.
- Among the phenolic phytoalexins, hydroxy coumarins and hydroxycinnamate conjugates contribute to the **disease resistance mechanism** in plants.
- Phenolic compounds also produce an **allelopathic effect**. A well- known compound from Juglans species is juglone which is highly toxic for a wide range of plants. It occurs in the plant as a non-toxic glucoside and is made active by deglucosylation and oxidation after leaching from the leaves into the soil.
- Phenolics also function as **signal molecules** in the interaction between **nitrogen-fixingbacteria and leguminous plants.**
- These plants exude **flavonoids** which act selectively in Rhizobia as **inducers of nodulation gene transcription.**
- Salicylic acid is strongly implicated as a **signal molecule** that induces active defense responses in several plant species against many types of pathogens.
- Recently, it has been shown that phenolic compounds function as **effective antioxidants.**
- Polyphenolics are important in **foodstuffs, wines and herbal teas** because of their **astringent taste.**

- Plants rich in polyphenolics were used as **tanning agents** in leather industries.
- **Phenolic pigments** (anthocyanins, flavones etc) of fruits are the most widespread food colours occurring in fruit juices, wines and jams.
- Anthocyanins have considerable potential in the food industry as **safe and effective foodadditives.**



The PLANT PHENOLIC COMPOUNDS

Introduction & The Flavonoids



Phenol

The plant phenolic compounds

- 8,000 Phenolic structures known

- Account for 40% of organic carbon circulating in the biosphere

- Evolution of vascular plants: in cell wall structures, plant defense, features of woods and barks, flower color, flavors

The plant phenolic compounds

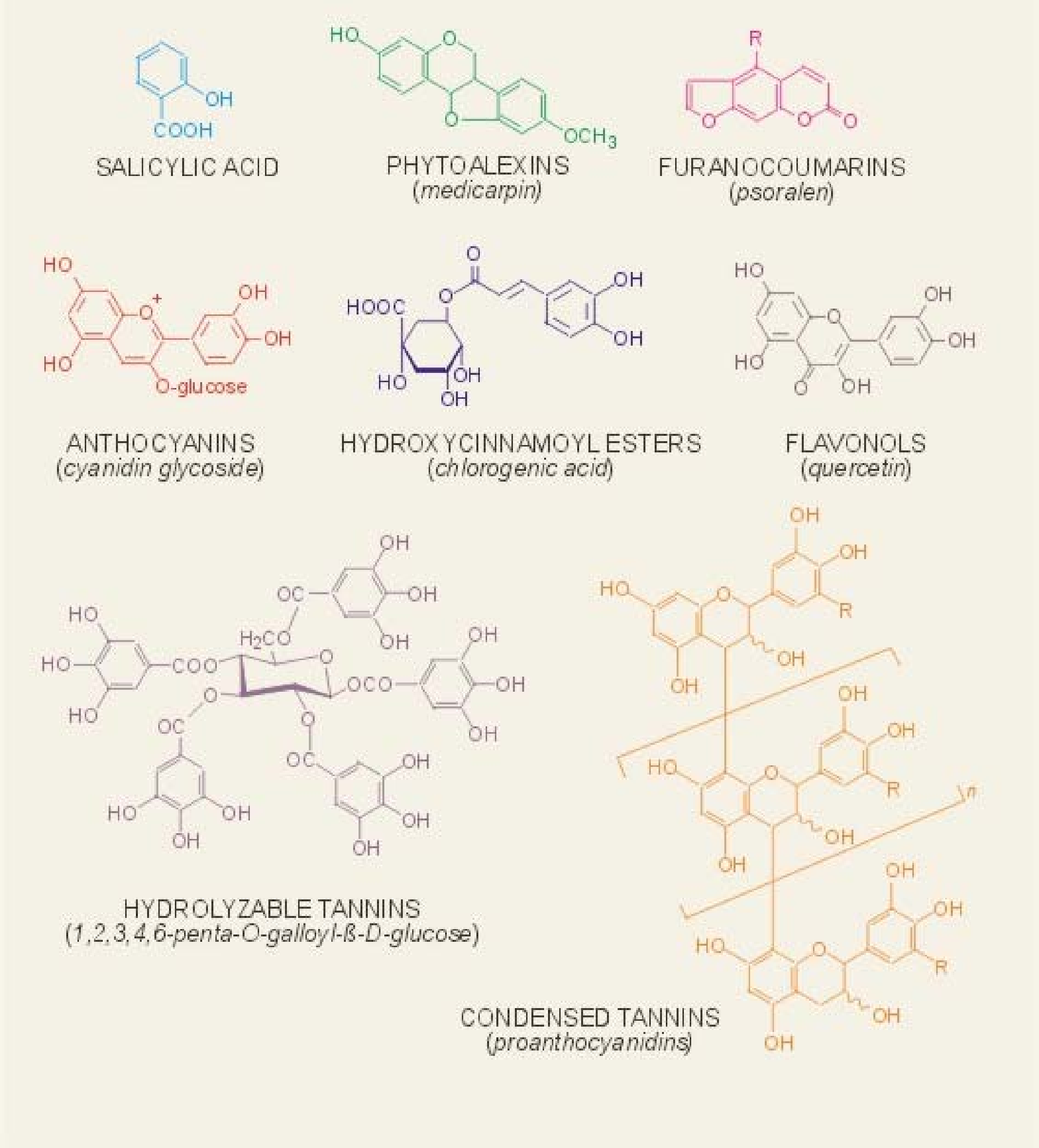
They can be:

Simple, low molecular weight, single aromatic ringed compounds

TO-

Large and complex- polyphenols

The Plant phenolic compounds



SALICYLIC ACID

PHYTOALEXINS
(*medicarpin*)

FURANOCOUMARINS
(*psoralen*)

ANTHOCYANINS
(*cyanidin glycoside*)

HYDROXYCINNAMOYL ESTERS
(*chlorogenic acid*)

FLAVONOLS
(*quercetin*)

HYDROLYZABLE TANNINS
(*1,2,3,4,6-penta-O-galloyl-ß-D-glucose*)

CONDENSED TANNINS
(*proanthocyanidins*)

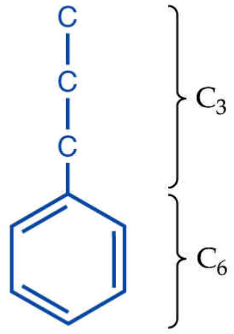
The plant phenolic compounds

- Primarily derived from the:

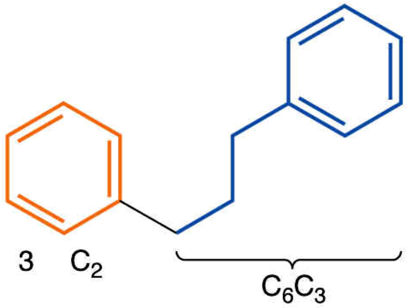
Phenylpropanoid pathway and acetate pathway

(and related pathways)

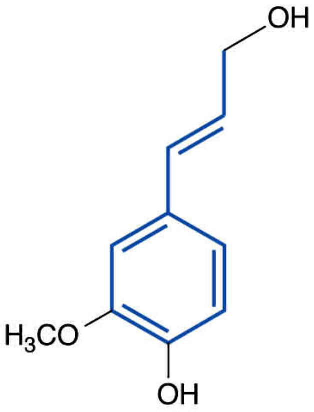
Phenylpropanoid pathway and phenylpropanoid-acetate pathway



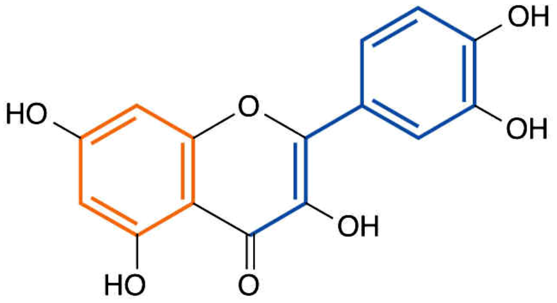
Phenylpropanoid
skeleton (C_6C_3)



Phenylpropanoid–acetate skeleton (C_6C_3–C_6),
with phenylpropanoid-derived (C_6C_3) and
acetate-derived (3 C_2) rings



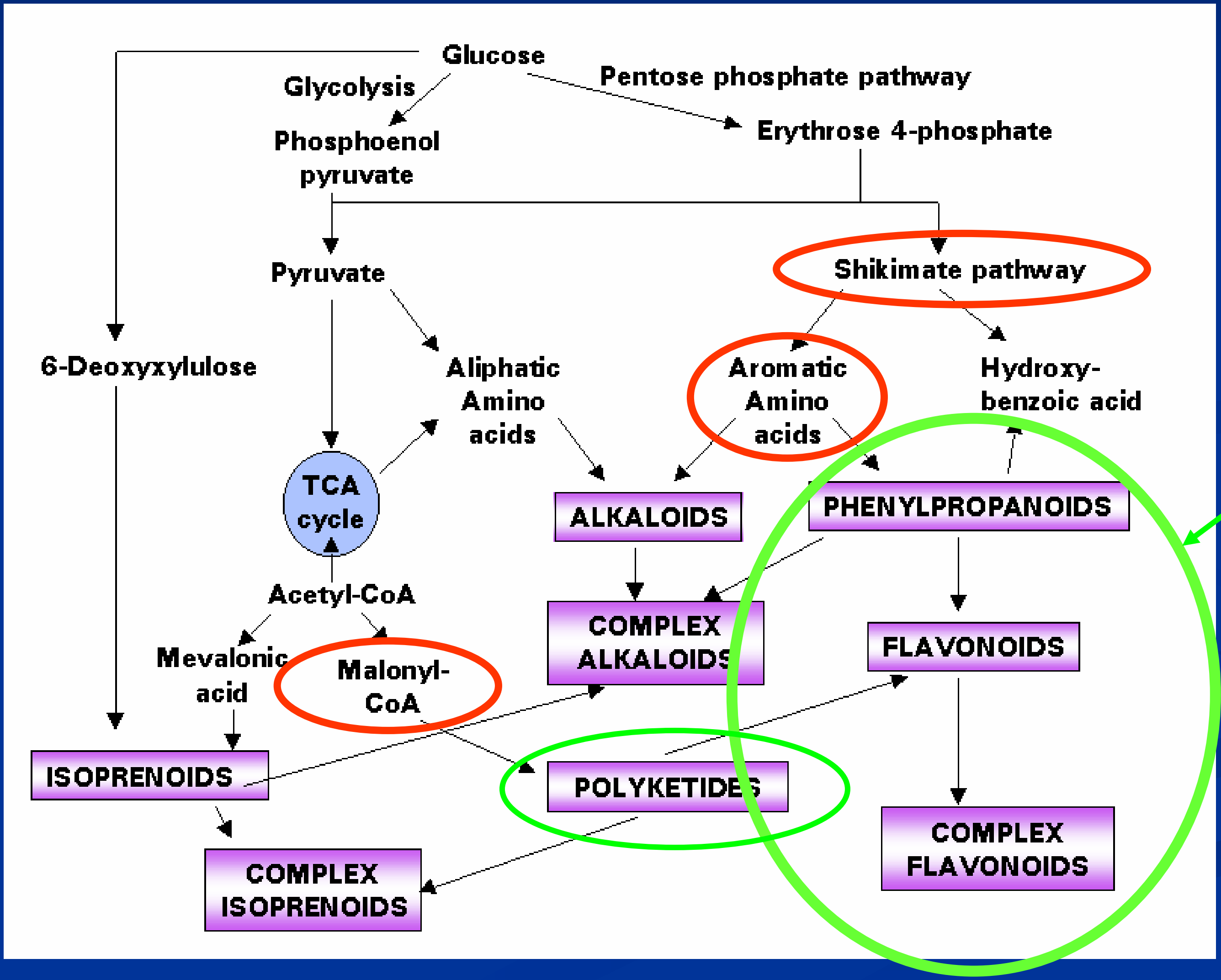
Coniferyl alcohol,
a component of
lignins and many lignans



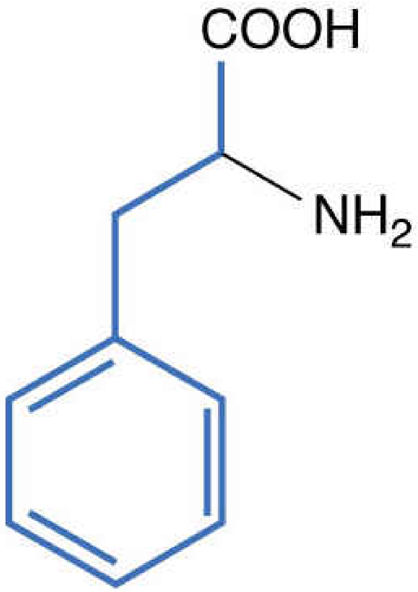
Quercetin, a flavonoid
(C_6C_3–C_6)

Phenylpropanoid skeleton
Acetate-derived rings

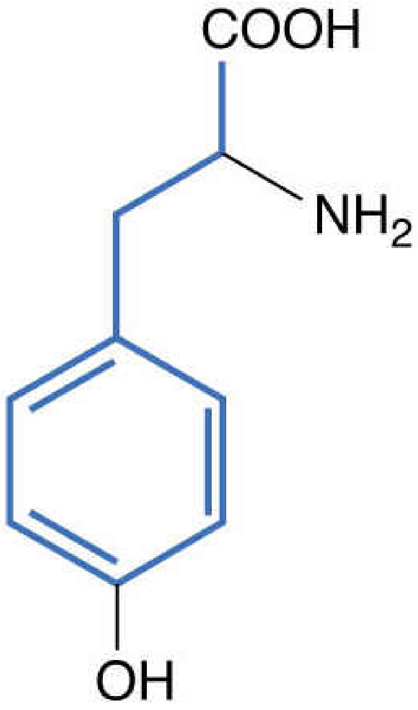
Precursors for plant phenolic compounds



The phenylpropanoids: products of the shikimic acid pathway

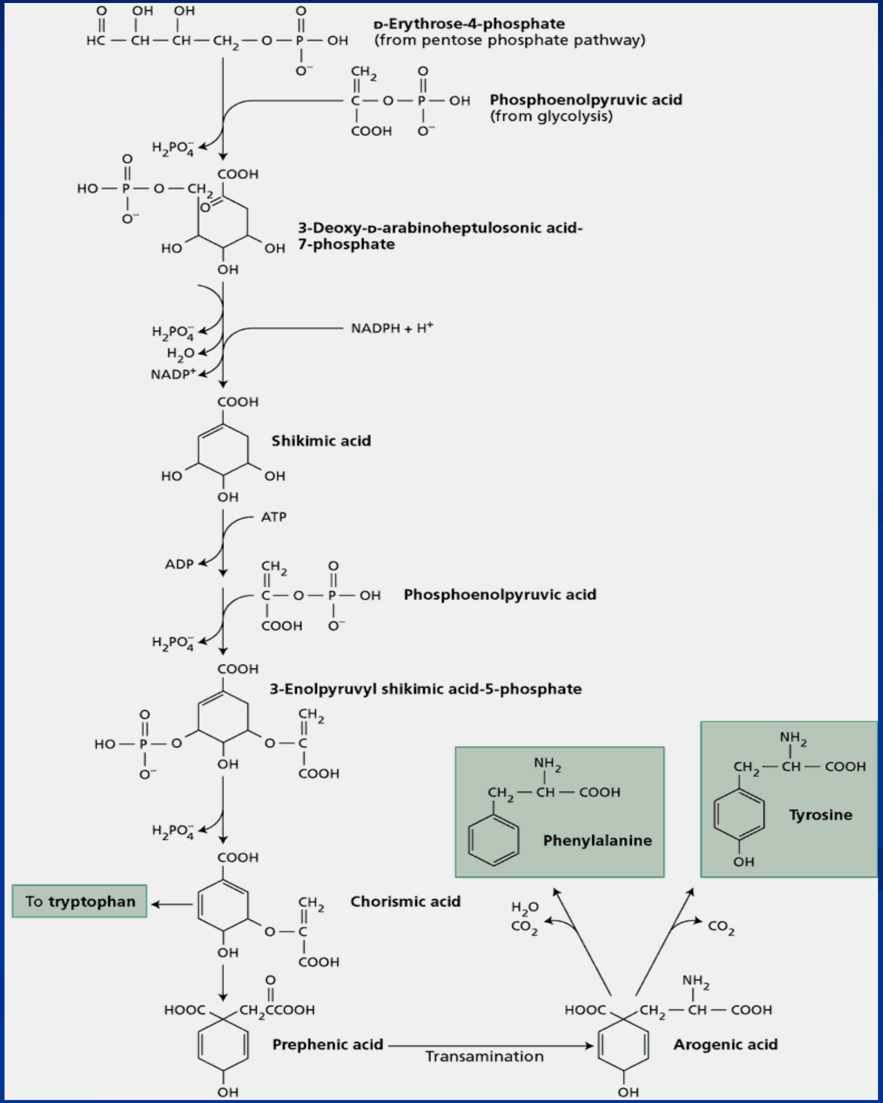


Phenylalanine



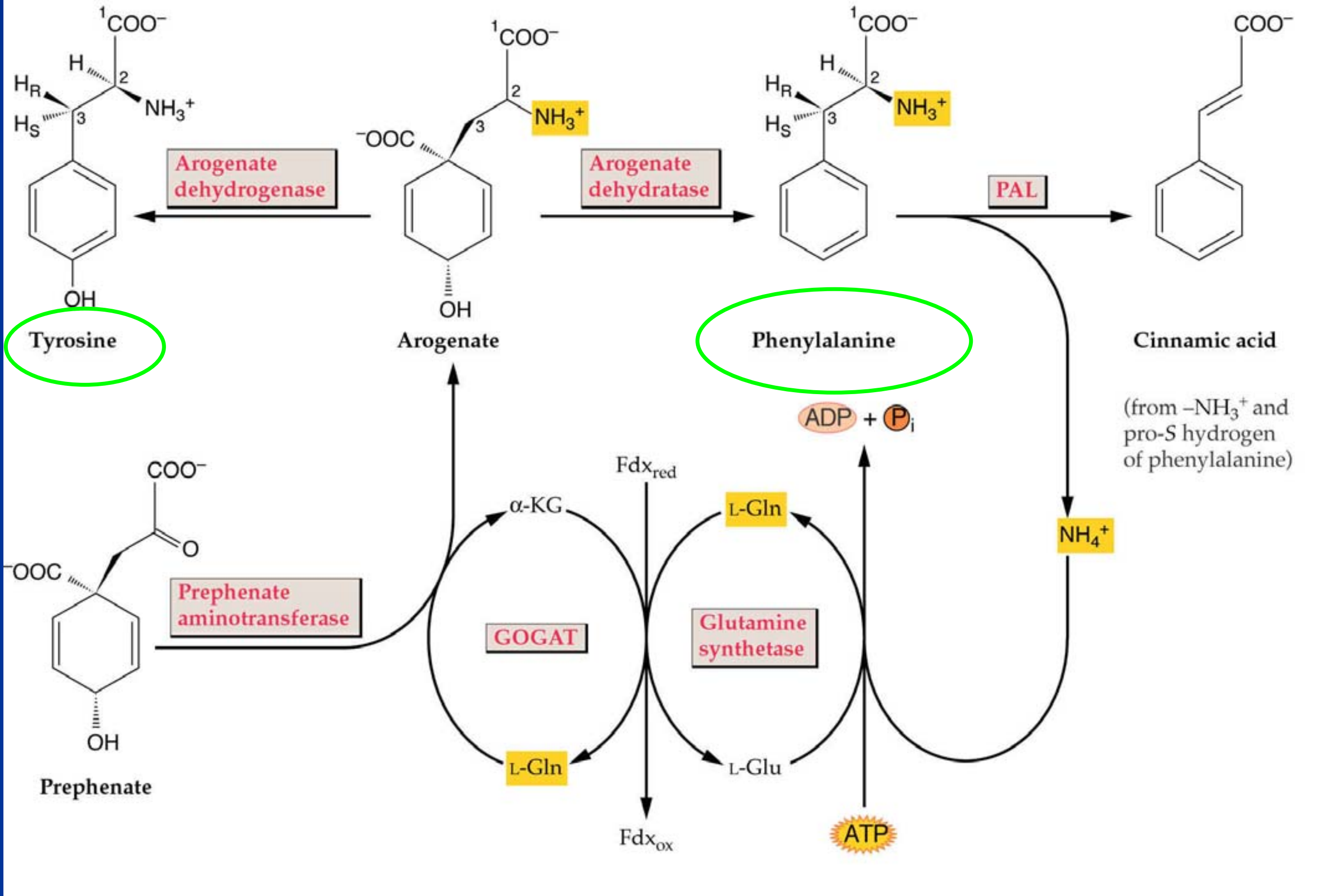
Tyrosine

The phenylpropanoids: products of the shikimic acid pathway (phe and tyr)



THE PHENYLPROPANOIDS: PRODUCTS OF THE SHIKIMIC ACID PATHWAY (phe & tyr)

The shikimate pathway



The plant phenolic compounds

- As in other cases of SMs, branches of pathway leading to biosynthesis of phenols are found or amplified only in specific plant families

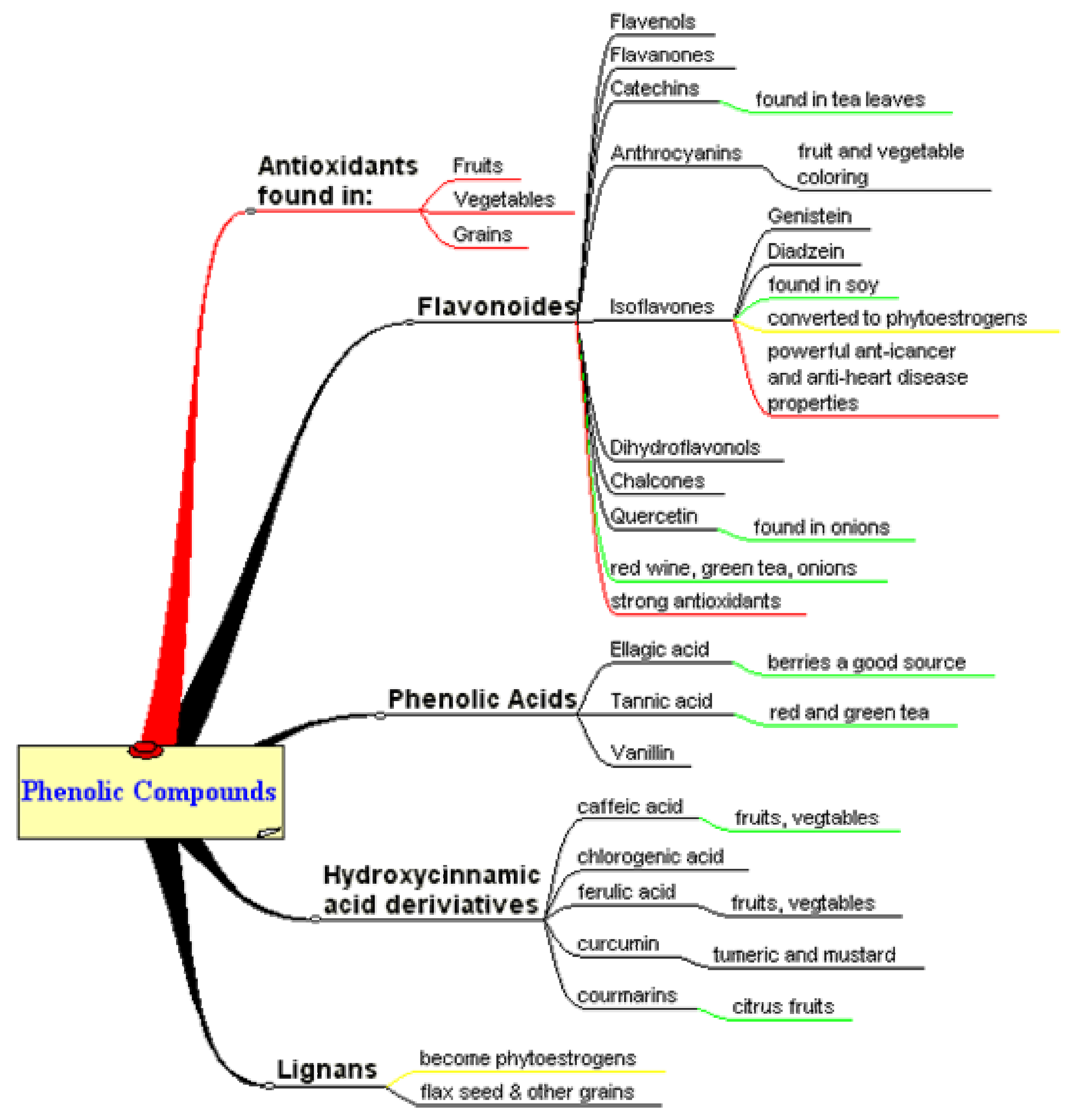
- Commonly found conjugated to sugars and organic acids

The plant phenolic compounds

Phenolics can be classified into 2 groups:

1. The FLAVONOIDS

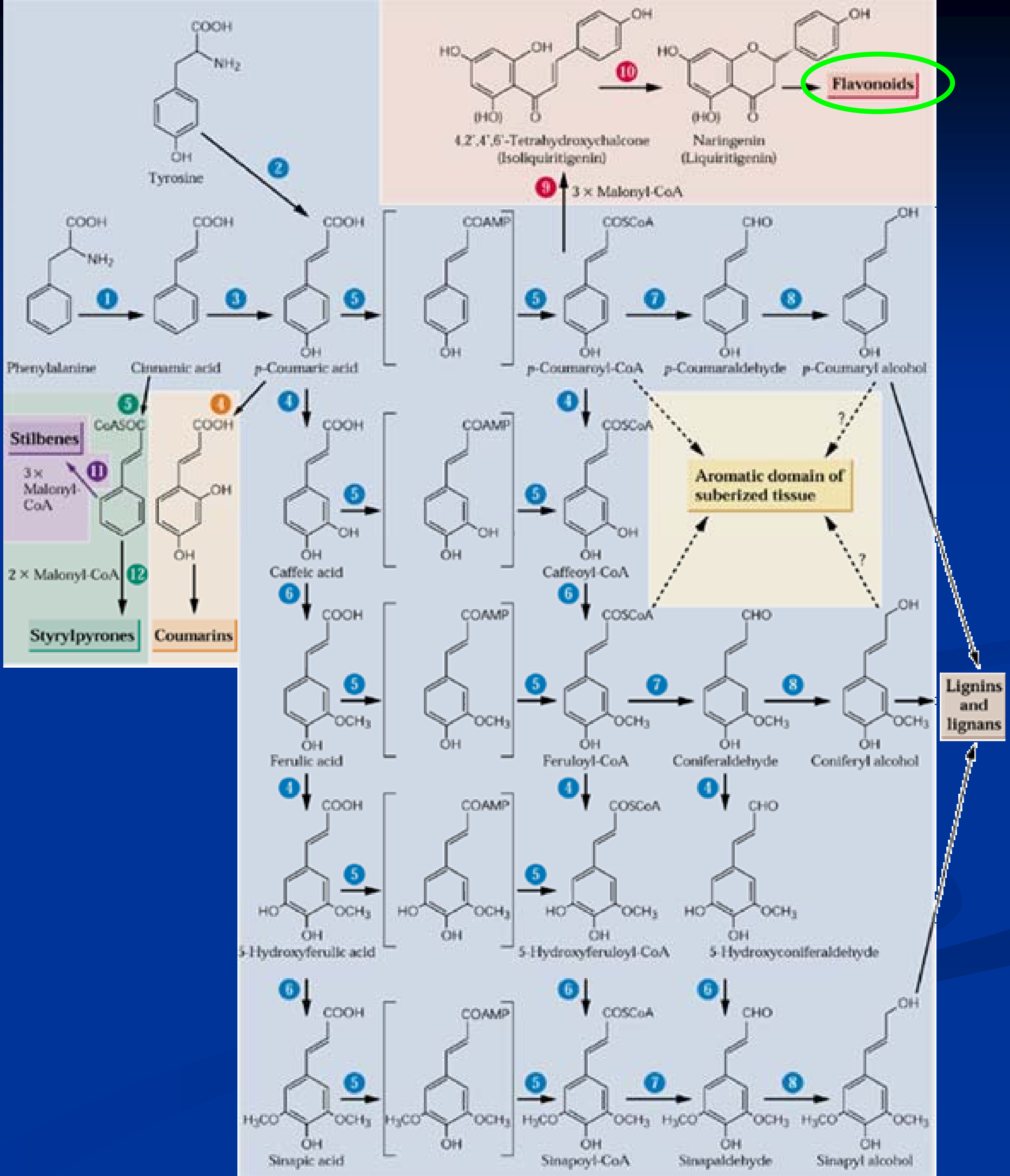
2. The NON-FLAVONOIDS



Phenolic Compounds

- **Antioxidants found in:**
  - Fruits
  - Vegetables
  - Grains
- **Flavonoides**
  - Flavenols
  - Flavanones
  - Catechins
    - found in tea leaves
  - Anthrocyanins
    - fruit and vegetable coloring
  - Isoflavones
    - Genistein
    - Diadzein
    - found in soy
    - converted to phytoestrogens
    - powerful ant-icancer and anti-heart disease properties
  - Dihydroflavonols
  - Chalcones
  - Quercetin
    - found in onions
  - red wine, green tea, onions
  - strong antioxidants
- **Phenolic Acids**
  - Ellagic acid
    - berries a good source
  - Tannic acid
    - red and green tea
  - Vanillin
- **Hydroxycinnamic acid deriviatives**
  - caffeic acid
    - fruits, vegtables
  - chlorogenic acid
  - ferulic acid
    - fruits, vegtables
  - curcumin
    - tumeric and mustard
  - coumarins
    - citrus fruits
- **Lignans**
  - become phytoestrogens
  - flax seed & other grains

The plant phenolic compounds

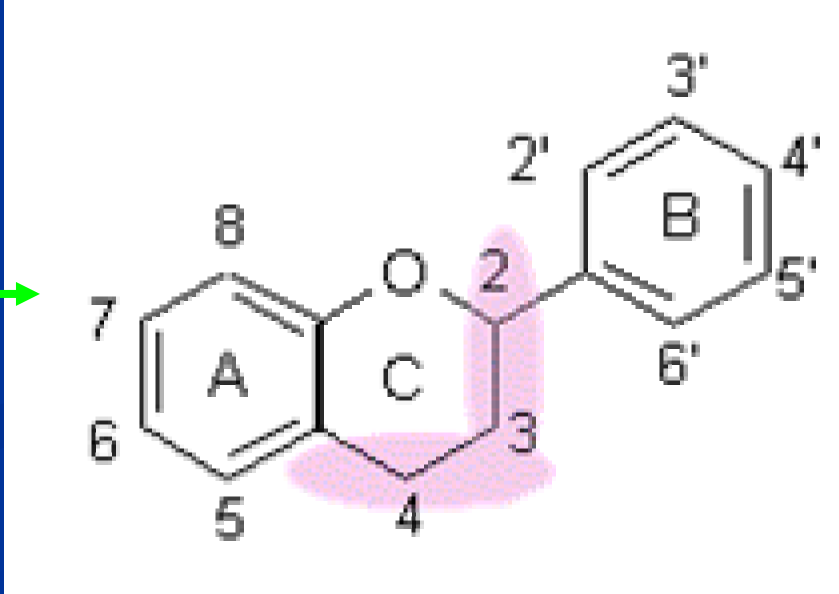


THE FLAVONOIDS

- Polyphenolic compounds
- Comprise:

15 carbons + 2 aromatic rings connected with a 3 carbon bridge

The Flavane Nucleus



THE FLAVONOIDS

- Largest group of phenols: 4500

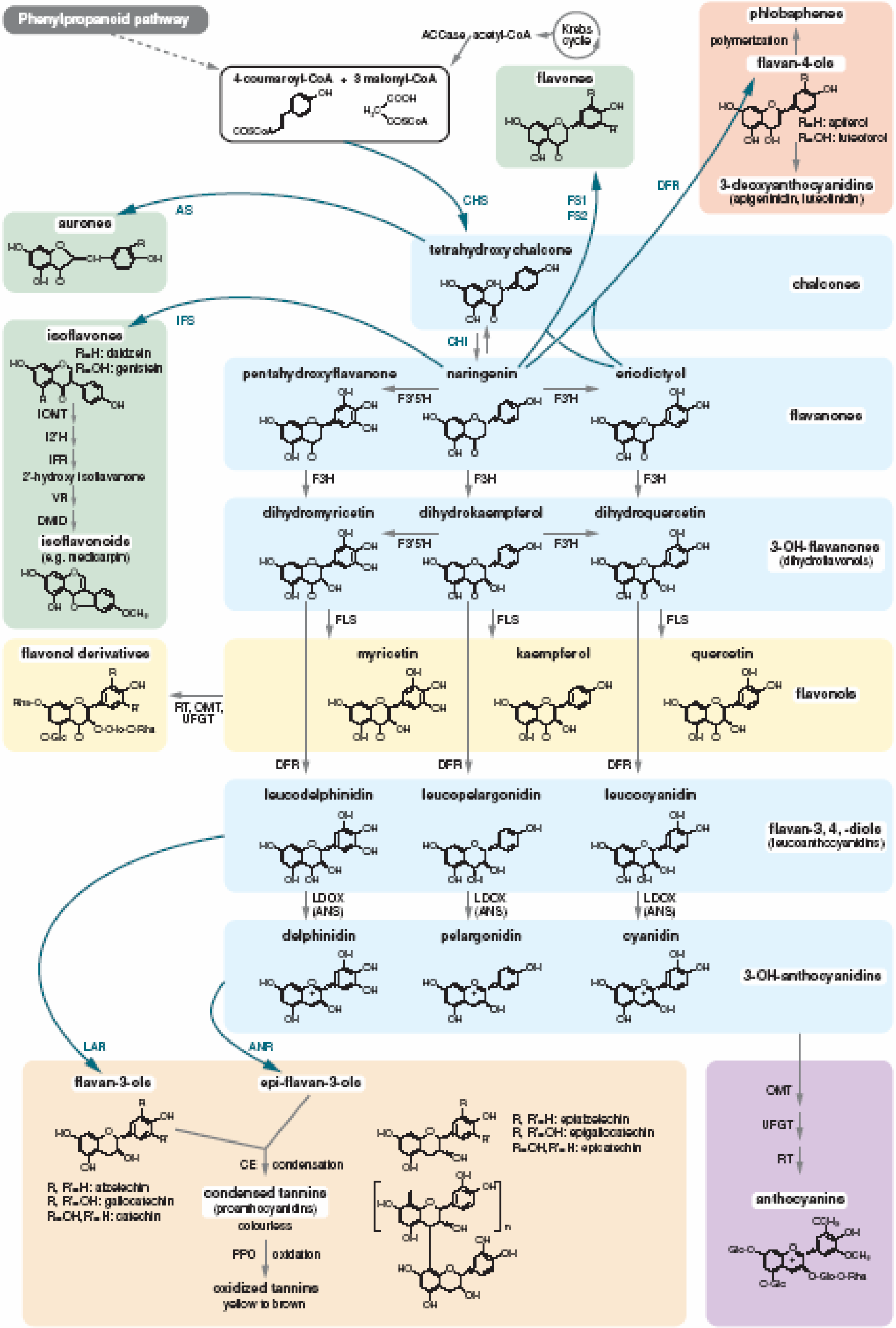
- Major role in plants: color, pathogens, light stress

- Very often in epidermis of leaves and fruit skin

- Potential health promoting compounds- antioxidants

- A large number of genes known

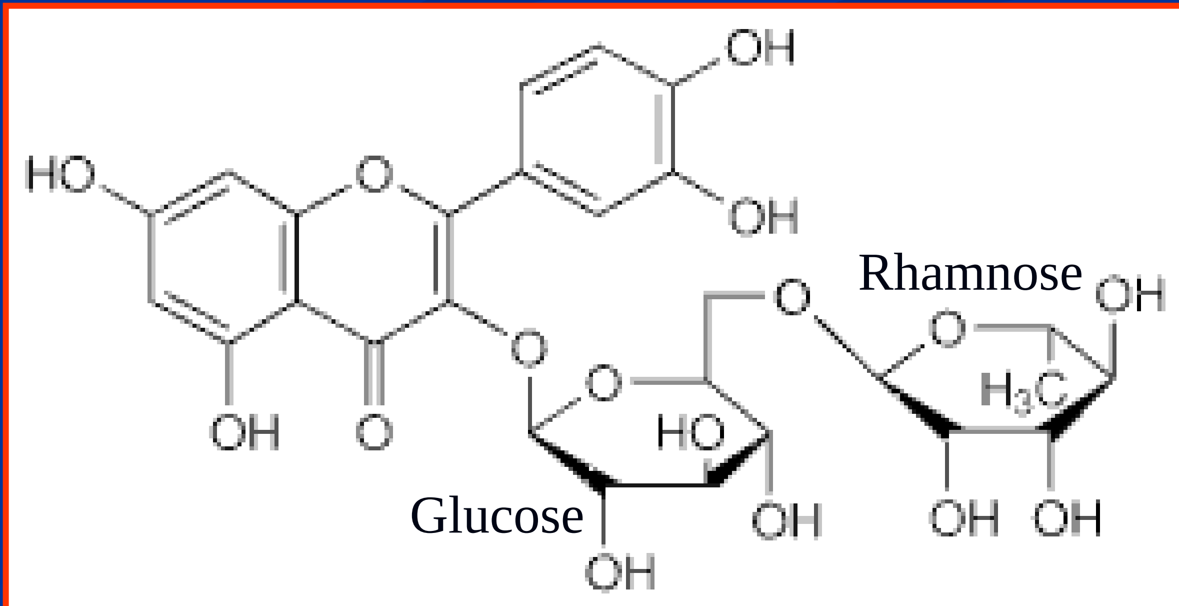
THE Flavonoids-classes



Lepiniec et al., 2006

THE Flavonoids

- The basic flavonoid skeleton can have a large number of substitutions on it:
- Hydroxyl groups
- Sugars - e.g. glucose, galactose, rhamnose. most structures are glycosylated
- Methylated
- Prenylated (farnesylated)
- Acylated



THE Flavonoids

- Sugars and hydroxyl groups increase the water solubility of flavonoids

- Methyl and isopentyl groups make flavonoids lipophilc

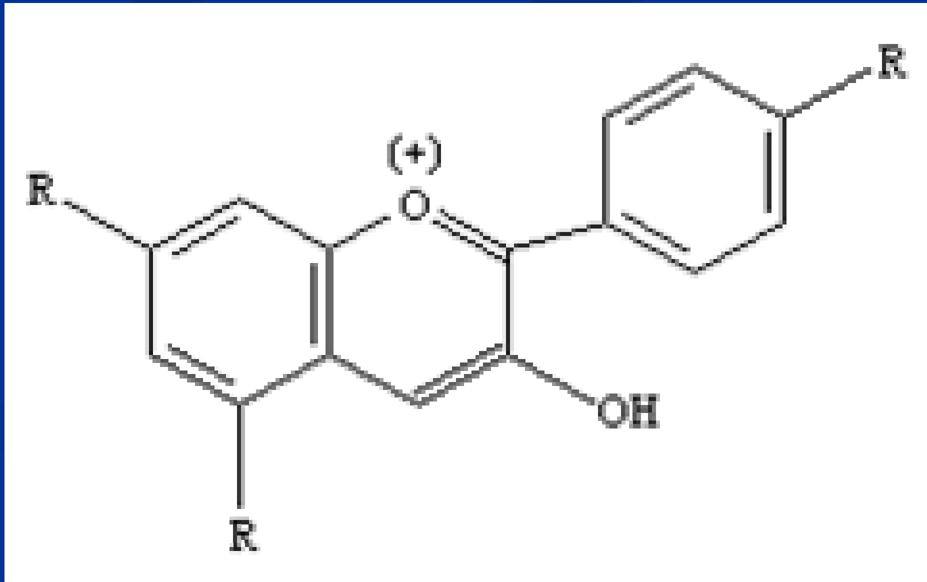
- If no sugar- AGLYCONE
- With sugar- GLYCOSIDE

Anthocyanins, Carotenoids, Chlorophylls

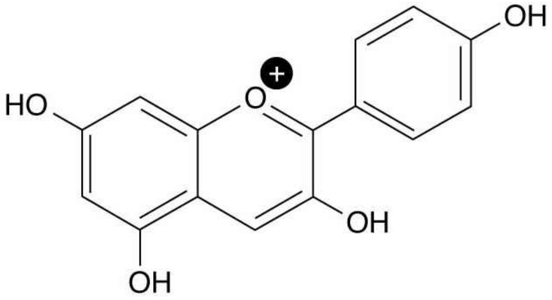
Anthocyanidins

- A positive charge the C ring

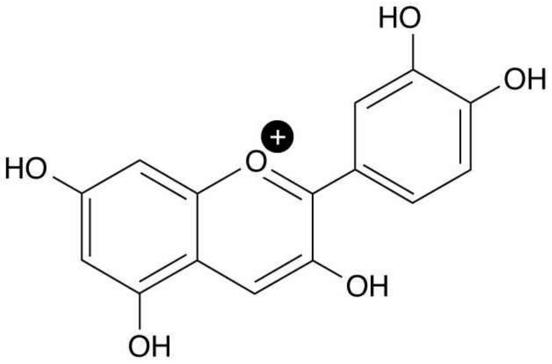
- Two double bonds in the C ring



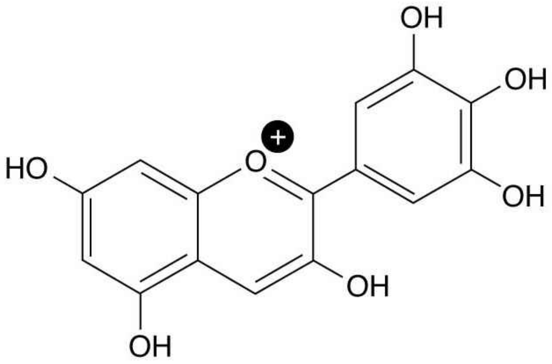
Anthocyanidins



Pelargonidin



Cyanidin



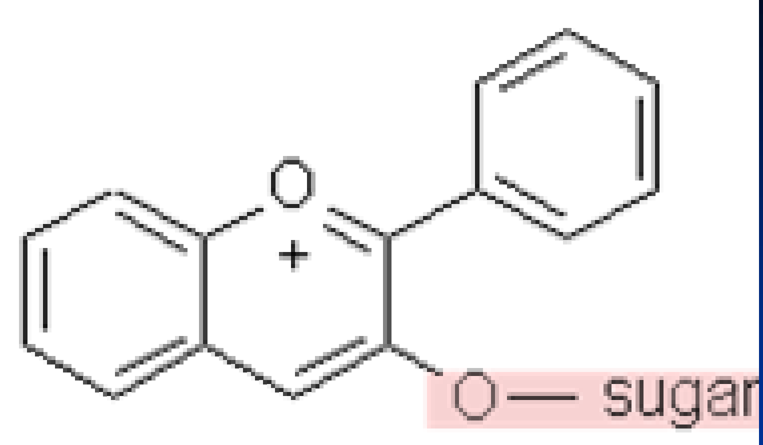
Delphinidin

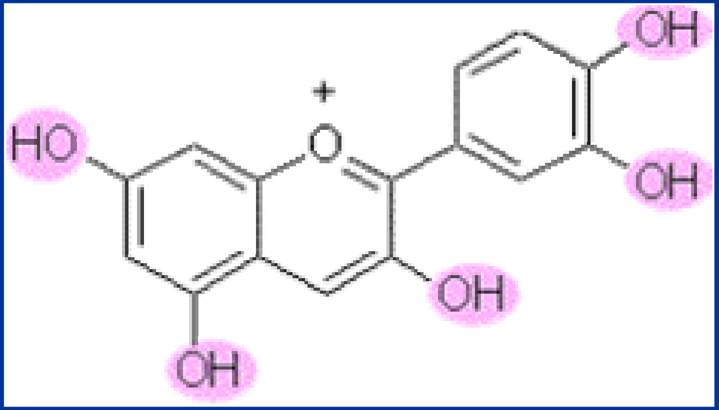
Pelargonium
(Geranium)

Rosa
(Rose)

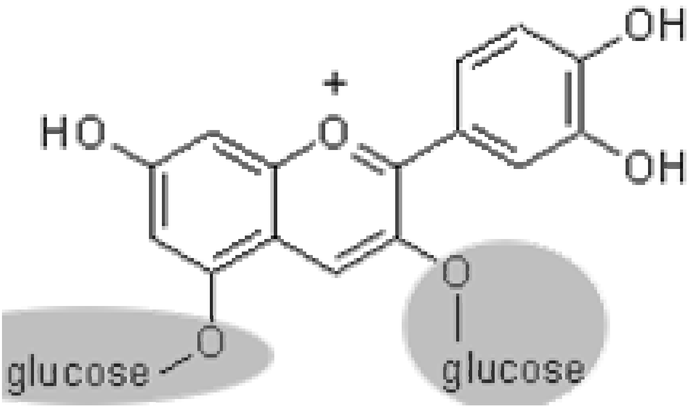
Delphinium
(Larkspur)

Anthocyanines





Cyanidin



Cyanin

Anthocyanidins

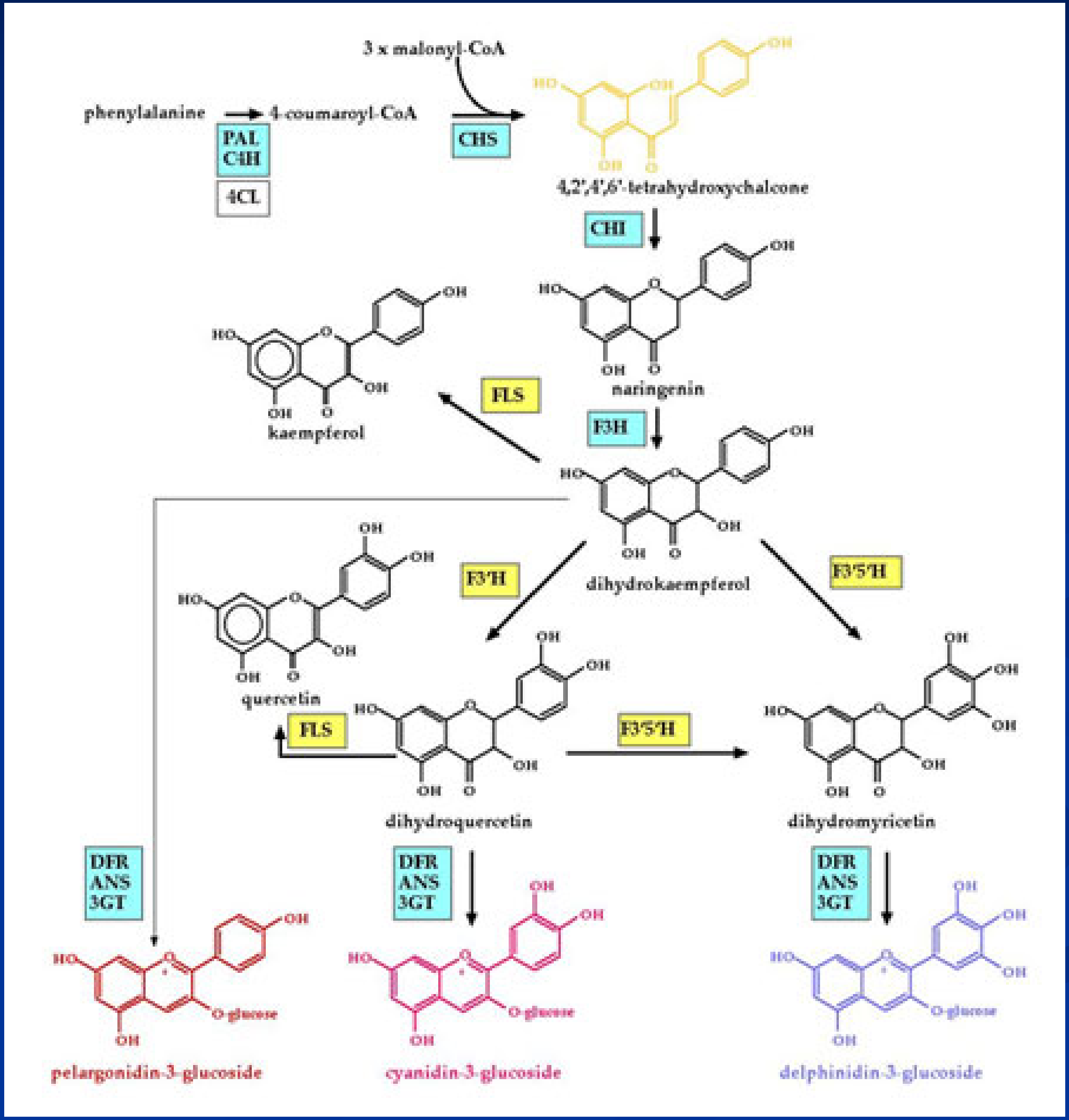
Anthocyanidin	**R_1**	**R_2**	**R_3**	**R_4**	**R_5**	**R_6**	**R_7**	**main colour**
Apigeninidin	-H	-OH	-H	-H	-OH	-H	-OH	orange
Aurantinidin	-H	-OH	-H	-OH	-OH	-OH	-OH	orange
Capensinidin	$-OCH_3$	-OH	$-OCH_3$	-OH	$-OCH_3$	-H	-OH	bluish-red
Cyanidin	-OH	-OH	-H	-OH	-OH	-H	-OH	magenta
Delphinidin	-OH	-OH	-OH	-OH	-OH	-H	-OH	purple, blue
Europinidin	$-OCH_3$	-OH	-OH	-OH	$-OCH_3$	-H	-OH	bluish red
Hirsutidin	$-OCH_3$	-OH	$-OCH_3$	-OH	-OH	-H	$-OCH_3$	bluish-red
Luteolinidin	-OH	-OH	-H	-H	-OH	-H	-OH	orange
Pelargonidin	-H	-OH	-H	-OH	-OH	-H	-OH	orange, salmon
Malvidin	$-OCH_3$	-OH	$-OCH_3$	-OH	-OH	-H	-OH	purple
Peonidin	$-OCH_3$	-OH	-H	-OH	-OH	-H	-OH	magenta
Petunidin	-OH	-OH	$-OCH_3$	-OH	-OH	-H	-OH	purple
Pulchellidin	-OH	-OH	-OH	-OH	$-OCH_3$	-H	-OH	bluish-red
Rosinidin	$-OCH_3$	-OH	-H	-OH	-OH	-H	$-OCH_3$	red
Triacetidin	-OH	-OH	-OH	-H	-OH	-H	-OH	red

Anthocyanins- Fruit color

Anthocyanins- Flower color

Anthocyanins- leaves and root color

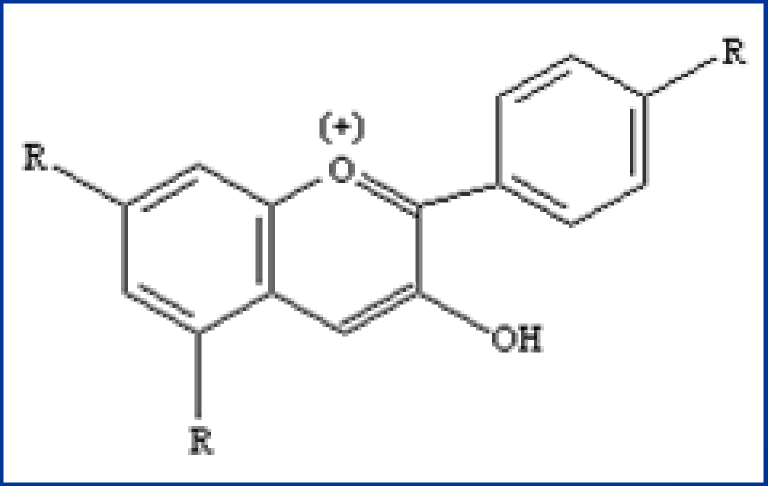
Anthocyanins biosynthesis



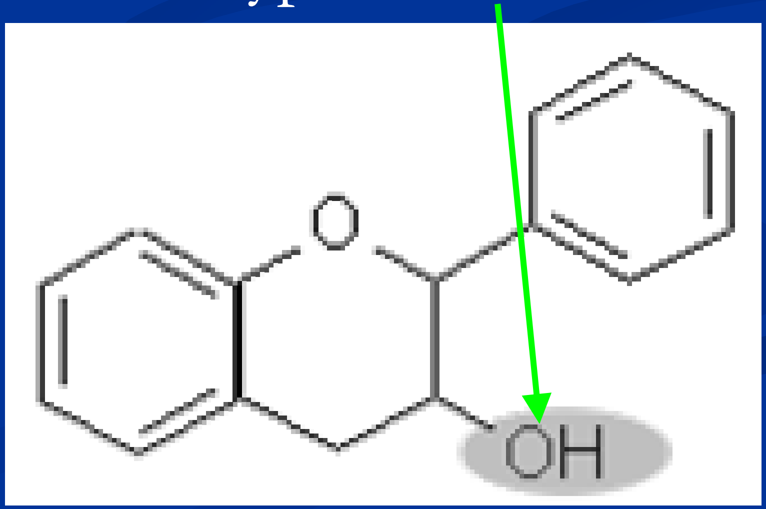
The flavAnols

- Structures are very similar to those of anthocyanidins: But no positive charge on the oxygen atom and no double bonds in the C ring.

Anthocyanidin



One type: Flavan-3-ol

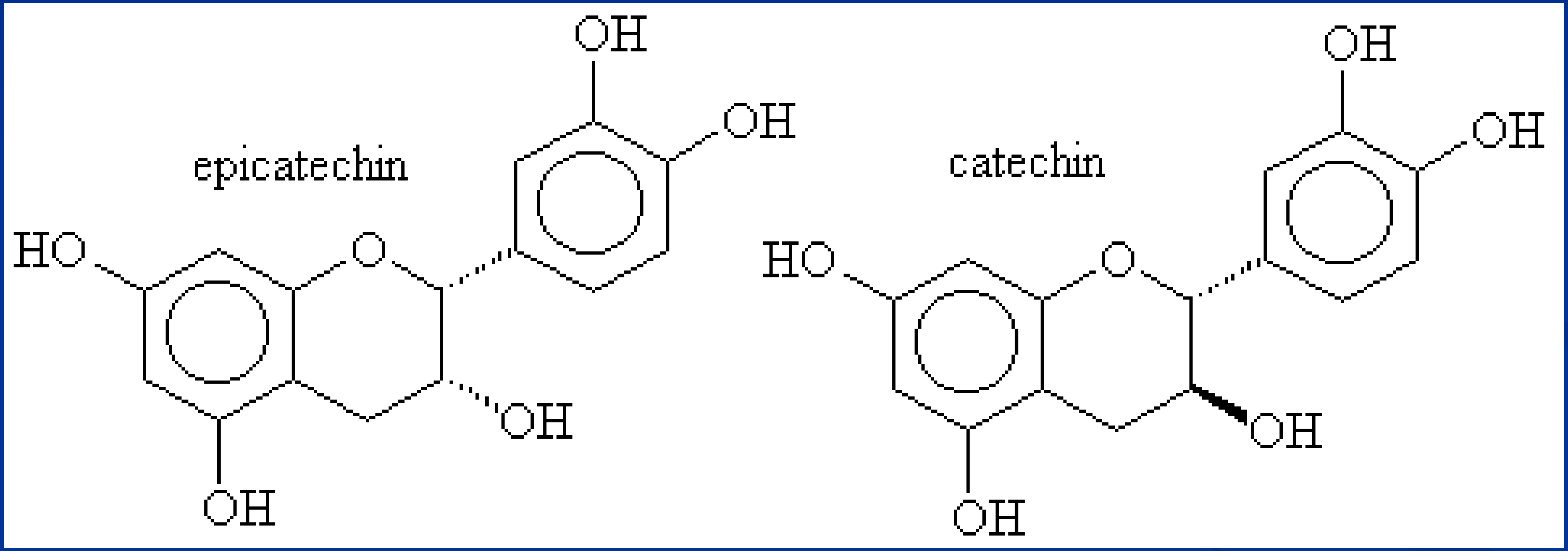


The flavAnols: Catechin & Epicatechin

Catechin-

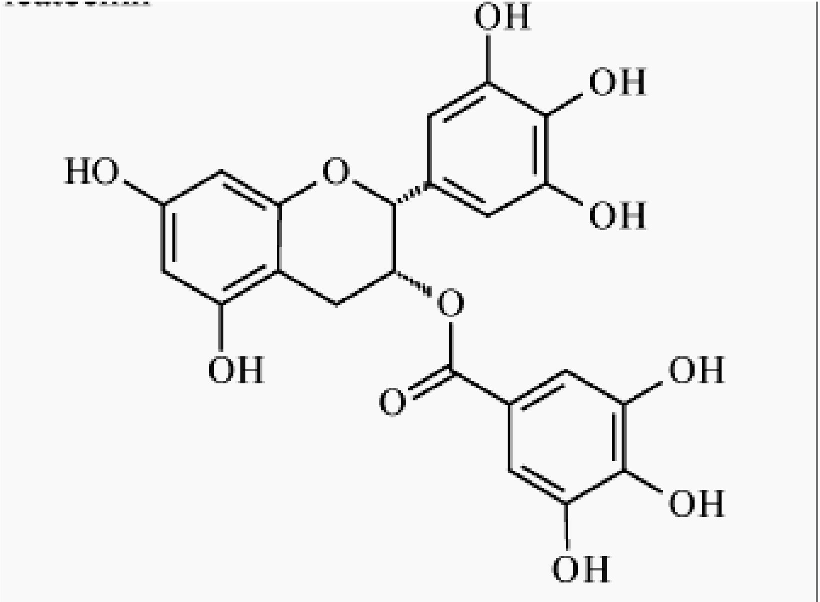
1. A common flavan-3-ol that occurs in many plants.
2. It's found in Green tea, Cocoa powder, Red wine
3. It is also a common subunit of proanthocyanidin polymers such as Procyanidin C2.
4. Epicatechin is another common example; it differs from Catechin only in the spatial orientation of its -OH group.

The flavAnols: Catechin & Epicatechin



The flavAnols in green tea

-Green tea contains high levels of flav-3-ols such as (-) Epigallocatechin gallate



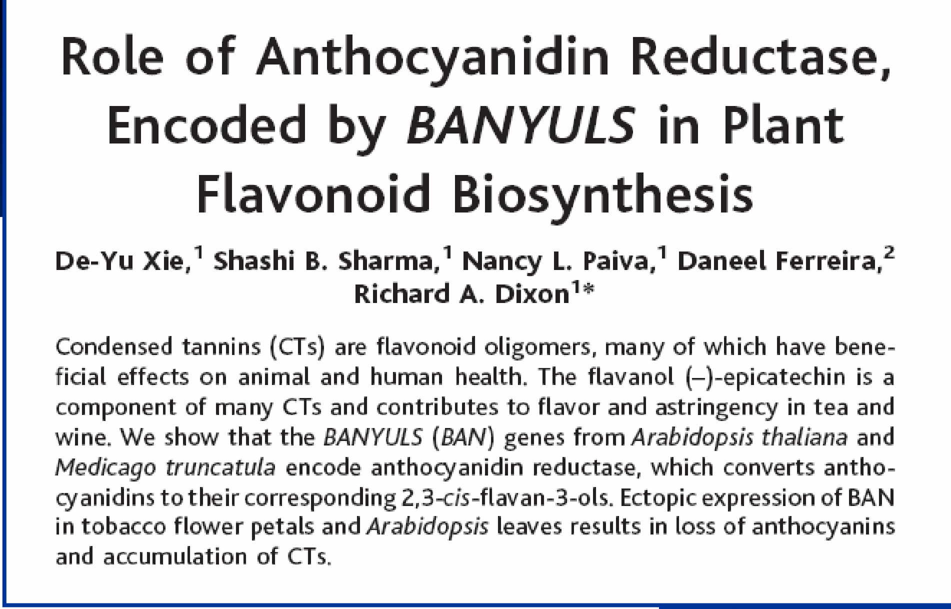
(-) – Epigallocatechin gallate

The flavAnols in green tea

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

1. Powerful antioxidants
2. Have beneficial effects on cardiac health, immunity and longevity
3. Levels of flav-3-ols decline in roasting

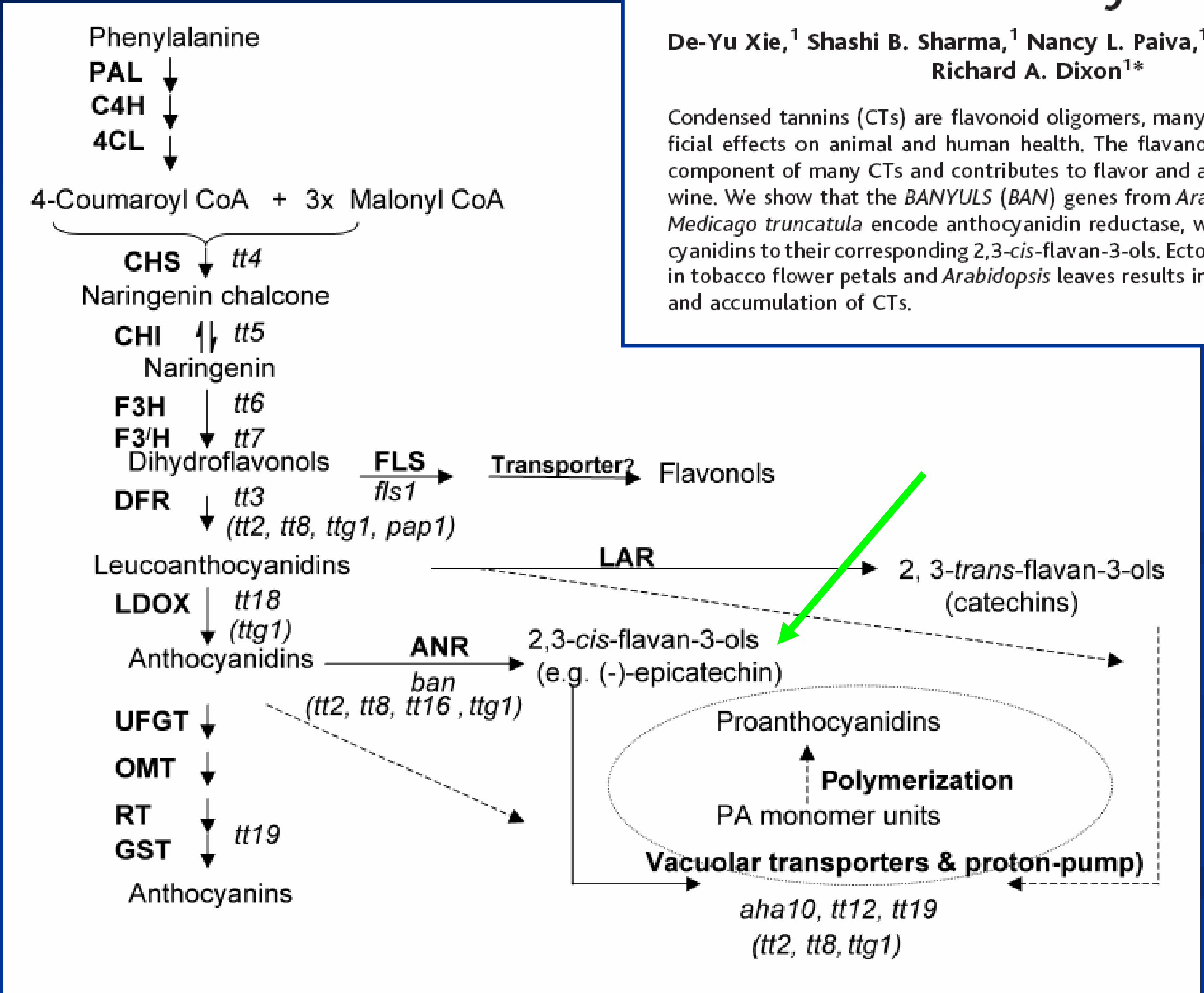
FlavAnols Biosynthesis



Role of Anthocyanidin Reductase, Encoded by *BANYULS* in Plant Flavonoid Biosynthesis

De-Yu Xie,[1] Shashi B. Sharma,[1] Nancy L. Paiva,[1] Daneel Ferreira,[2] Richard A. Dixon[1]*

Condensed tannins (CTs) are flavonoid oligomers, many of which have beneficial effects on animal and human health. The flavanol (–)-epicatechin is a component of many CTs and contributes to flavor and astringency in tea and wine. We show that the *BANYULS* (*BAN*) genes from *Arabidopsis thaliana* and *Medicago truncatula* encode anthocyanidin reductase, which converts anthocyanidins to their corresponding 2,3-*cis*-flavan-3-ols. Ectopic expression of BAN in tobacco flower petals and *Arabidopsis* leaves results in loss of anthocyanins and accumulation of CTs.



Phenylalanine
PAL
C4H
4CL
4-Coumaroyl CoA + 3x Malonyl CoA
CHS *tt4*
Naringenin chalcone
CHI *tt5*
Naringenin
F3H *tt6*
F3'H *tt7*
Dihydroflavonols
FLS *fls1*
Transporter?
Flavonols
DFR *tt3*
(tt2, tt8, ttg1, pap1)
Leucoanthocyanidins
LAR
2, 3-*trans*-flavan-3-ols
(catechins)
LDOX *tt18*
(ttg1)
Anthocyanidins
ANR
ban
(tt2, tt8, tt16 , ttg1)
2,3-*cis*-flavan-3-ols
(e.g. (-)-epicatechin)
Proanthocyanidins
Polymerization
PA monomer units
UFGT
OMT
RT
GST
tt19
Anthocyanins
Vacuolar transporters & proton-pump)
aha10, tt12, tt19
(tt2, tt8, ttg1)

(Sharma & Dixon, 2005)

Proanthocyanidines or Condensed Tannins

- Polymers made from multiple flavAnols

- They are called proanthocyanidins because, if broken apart with acid treatment, they yield anthocyanidins such as Cyanidin

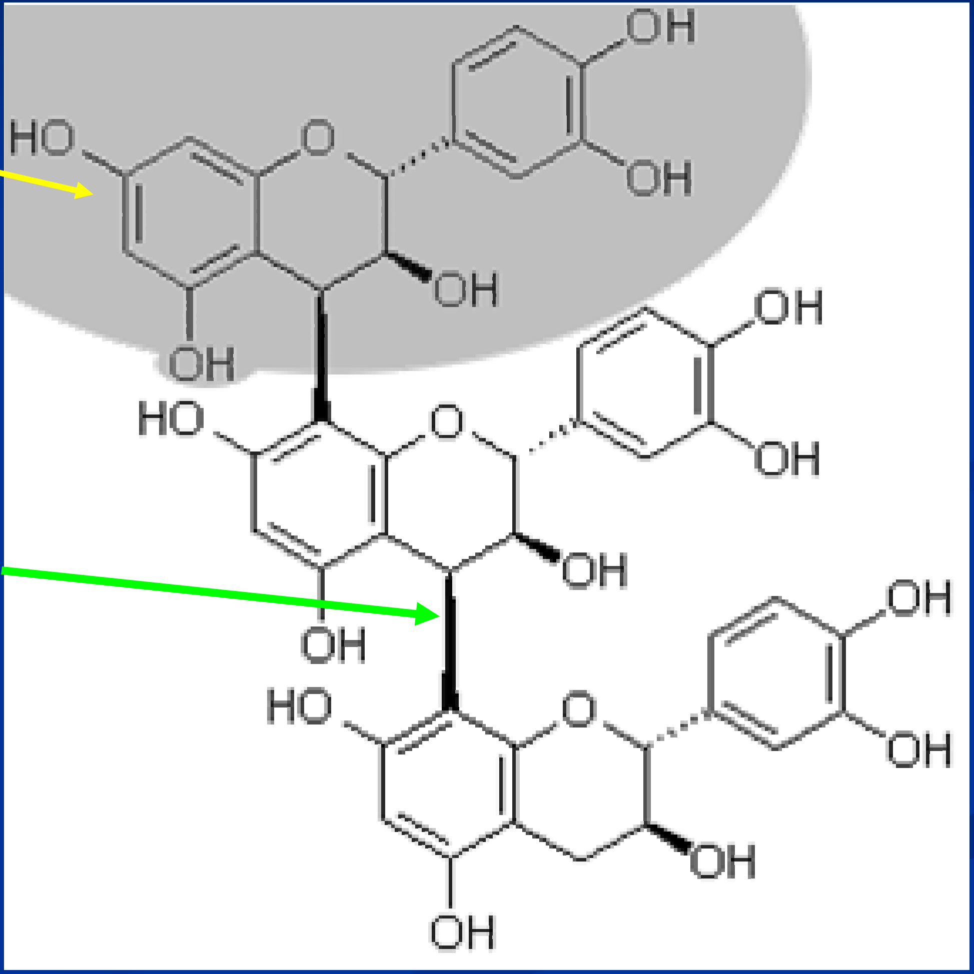
- Proanthocyanidin polymers consisting of up to 50 subunits

- Oligomeric proanthocyanidins (OPCs) are the water-soluble, short-chain polymers

Proanthocyanidines or Condensed Tannins

- Sometimes referred to as "condensed tannins"
- Responsible for astringency in many foods and medicinal herbs
- Red wine contains many complex proanthocyanidins (extracted from grape skins and seeds); so do blueberries, blackberries, strawberries, elderberries, and other red/blue/purple colored plant parts

Type-B proanthocyanidins (formed from - epicatechin and + catechin)



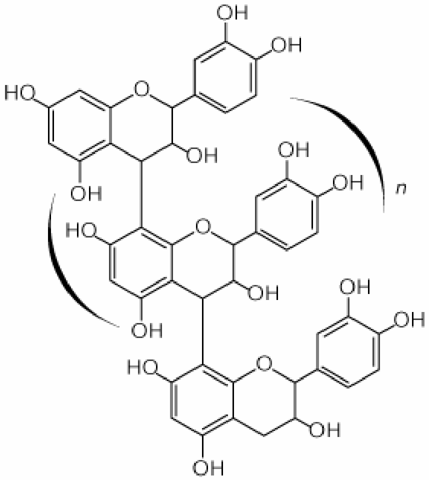
Catechin subunit

Oxidative coupling between C-4 of the heterocycle and the C-6 or C-8 positions of the adjacent unit

Type-B proanthocyanidins (formed from -epicatechin and + catechin)

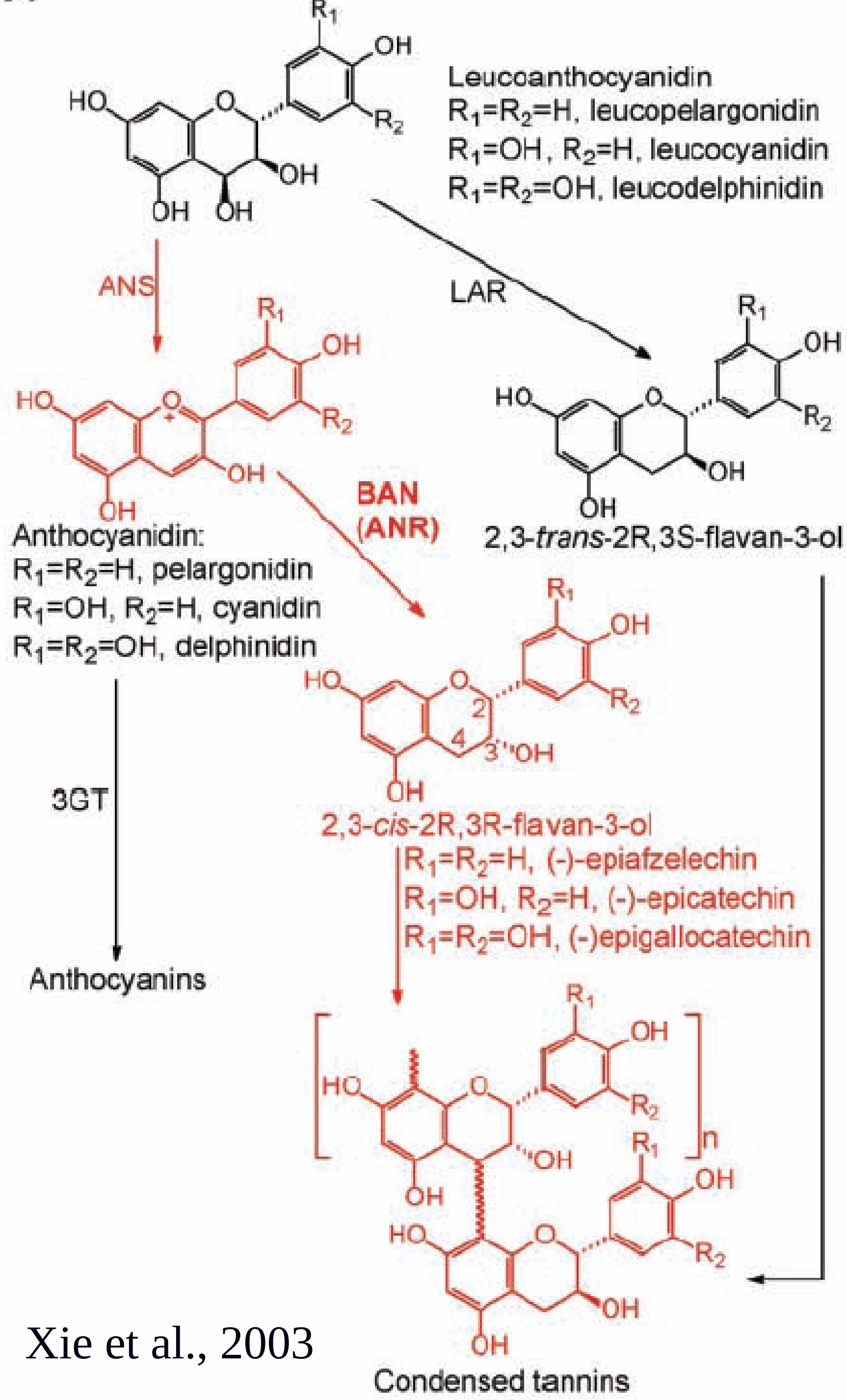
- Antifeedant proanthocyanidin in red sorghum
- These condensed tannins deter birds from feeding on the seed
- White sorghum deficient in these compounds is eaten by birds


Red sorghum



Proanthocyanidin ($n = 1$–30)

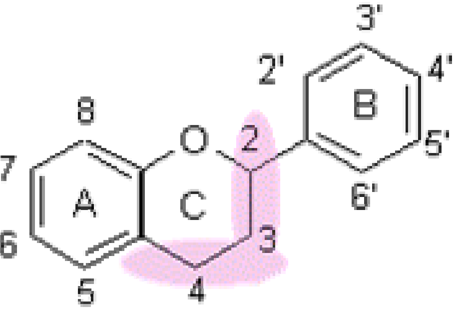
Proanthocyanidins (or condensed tannins) Biosynthesis

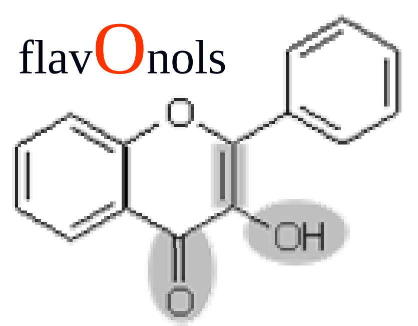


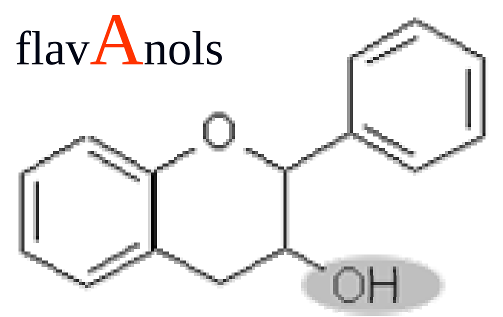
Xie et al., 2003

The FlavOnols

- The molecule has a double-bonded oxygen atom attached to position 4 (that’s why flavOnols).
- They're still "-ols" because they retain the -OH group at position 3 like the flavAnols
- The double-bonded oxygen atom, makes them like another class of flavonoids known as "flavones" (next)
- Double bond in between C2 and C3 (C ring)
- Involved in UV screening, due to their strong absorbance in UV-A (325-400nm) and UV-B (280-325 nm) wavelengths





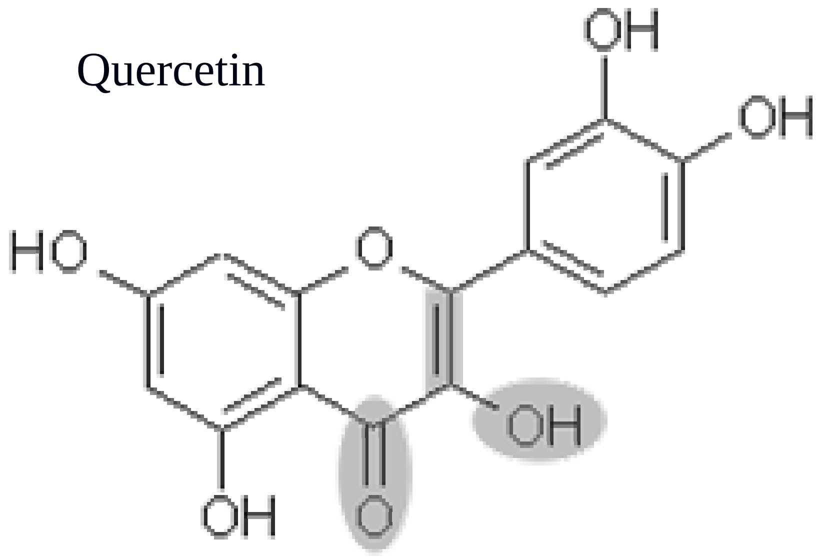


The FlavOnols- Quercetin

- The most abundant flavonol in the diet and is found in hundreds of herbs and foods.

- Onions are especially rich in *Quercetin*.

- It has proven antioxidant effects



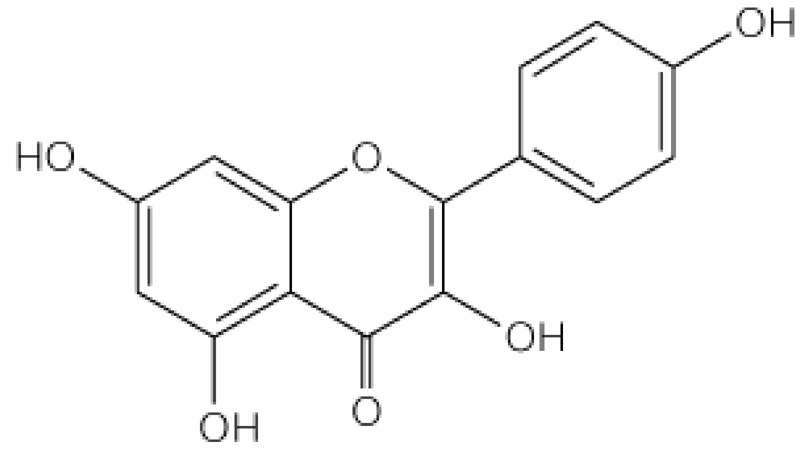
The FlavOnols- Quercetin

- FalvOnols are mostly found as *O*-glycosides

- Aglycons- 300

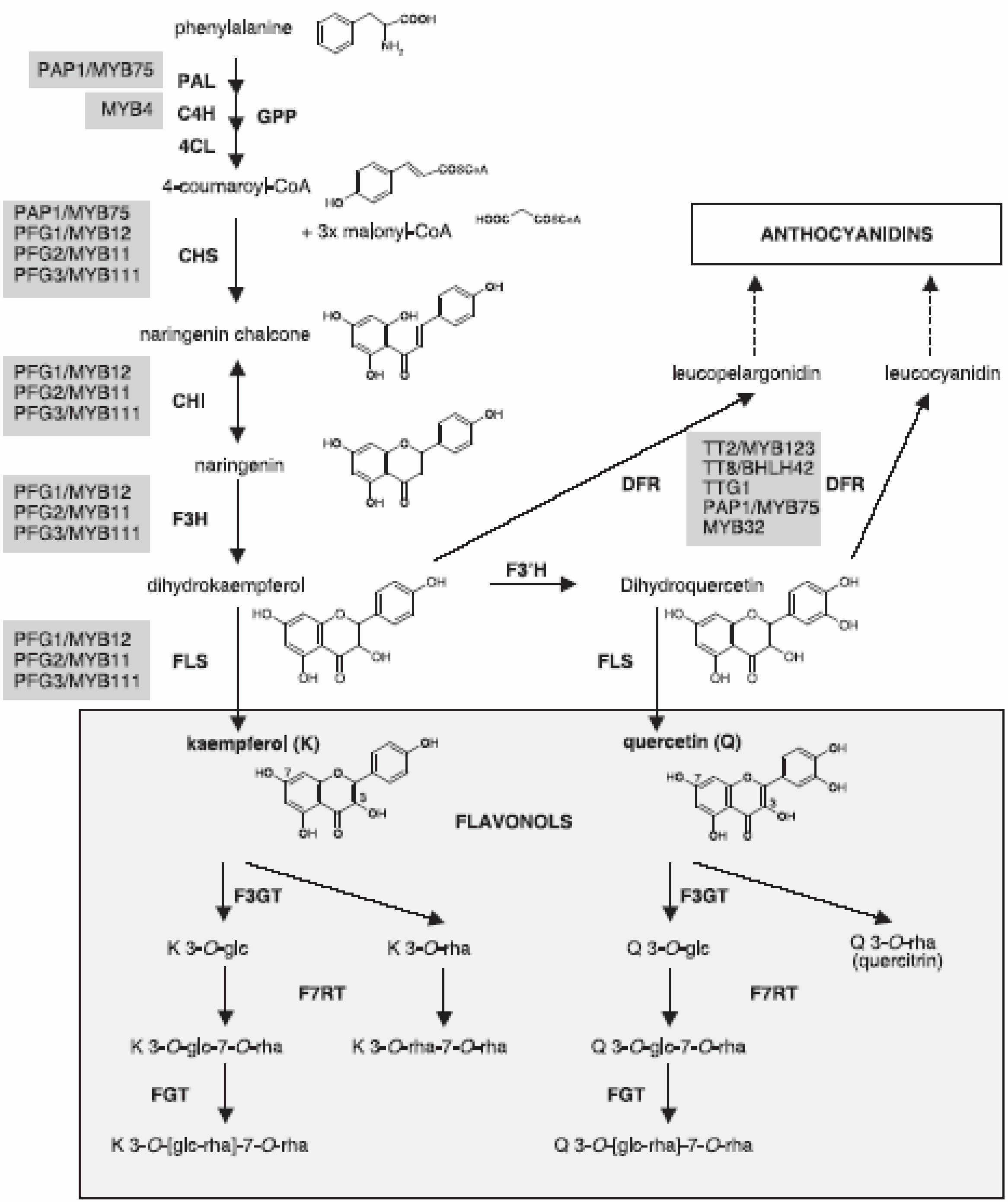
- Total- 1030

- More than 200 different sugar conjugates of Kaempferol !!



Kaempferol

FlavOnols Biosynthesis

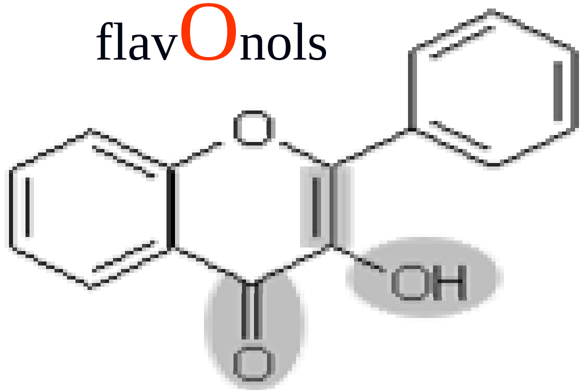


The Flavones

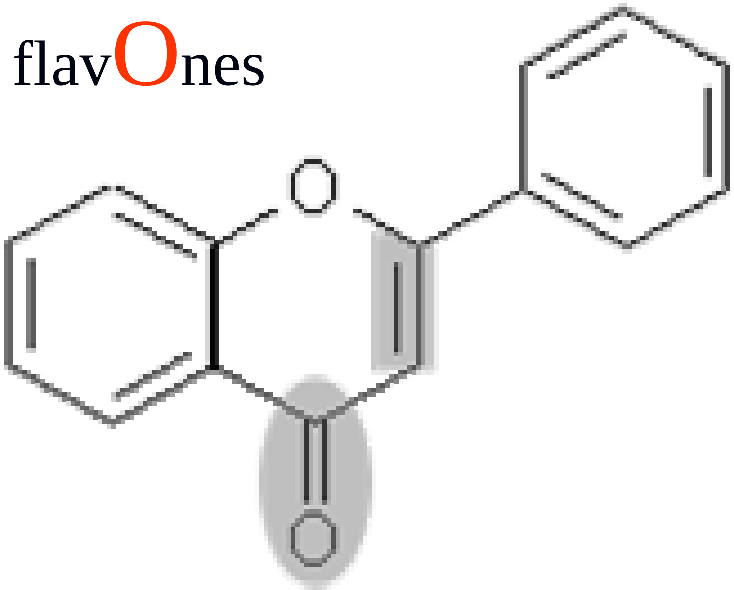
- Close to the flavOnols but not so widespread (celery, parsley and some herbs)

- BUT Without the "-ol." there is no longer an -OH group at position 3 on the central ring

flavOnols

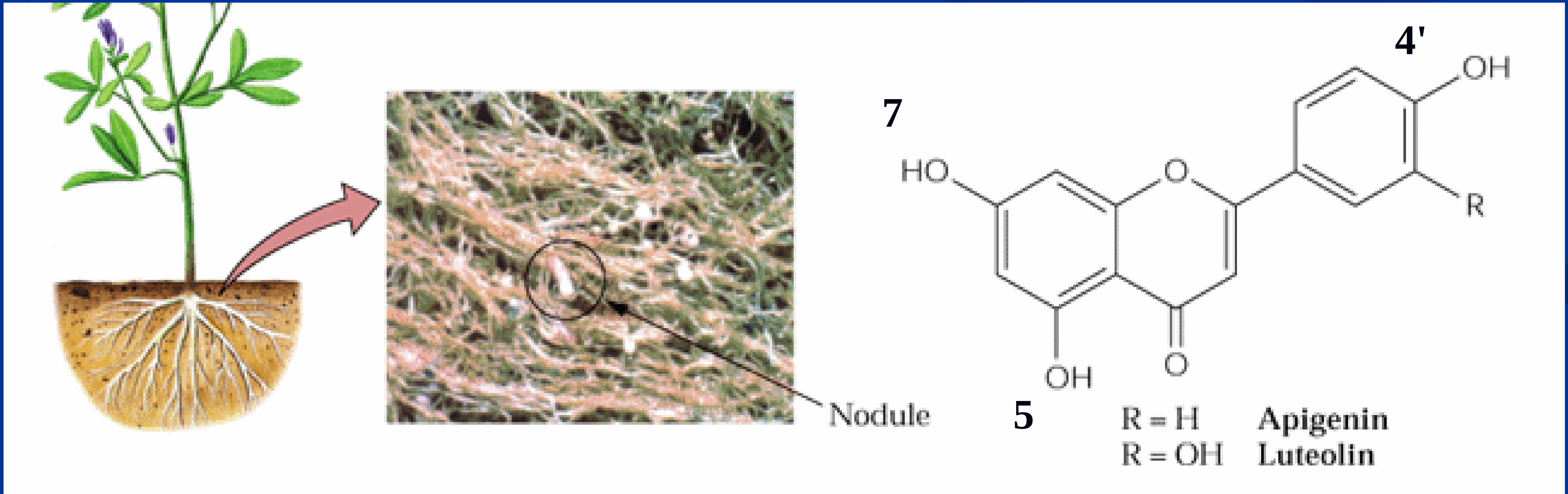


flavOnes



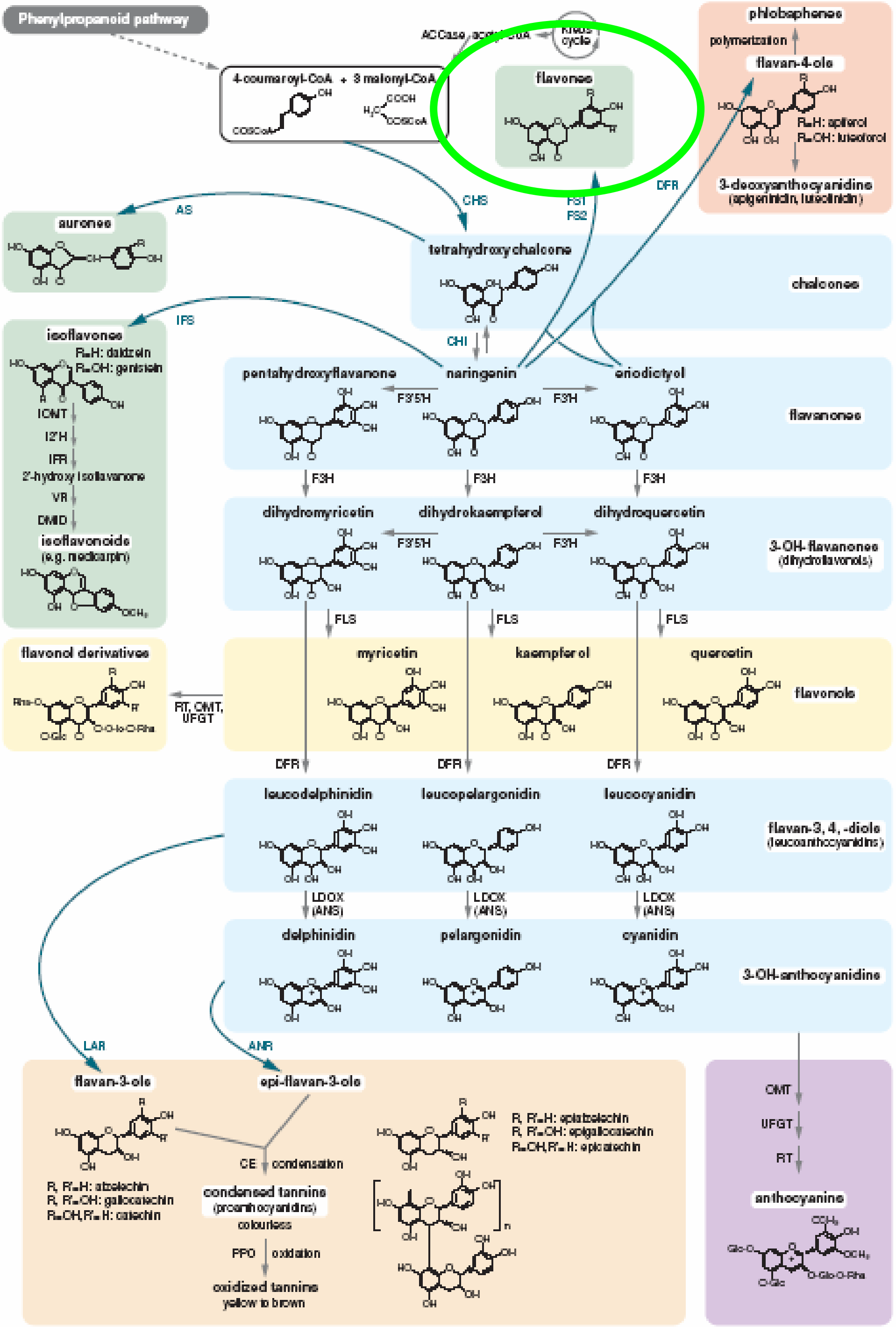
The Flavones- Apigenin

- Apigenin, a flavone with - OH groups added to positions 5, 7, and 4'
- Another flavone is luteolin, found in sweet red peppers
- Both act as signaling molecules that induce NOD factors in compatible interaction with Rhizobium bacteria (nitrogen fixing root nodules) in legumes (e.g. alfalfa)



THE Flavones

Origenate from the Flavanones (naringenin)

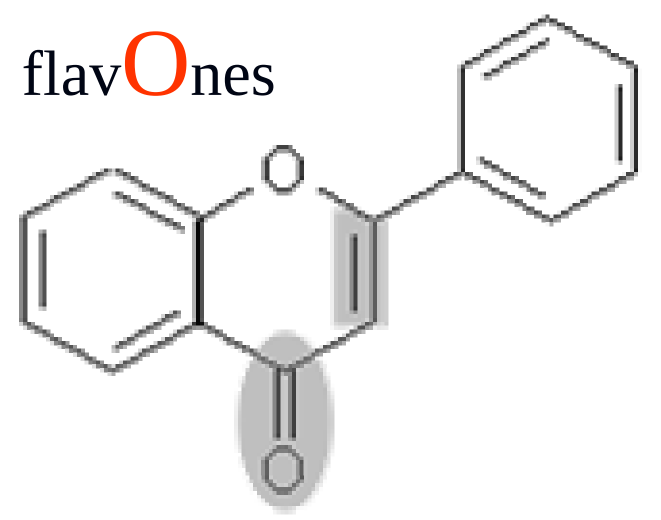


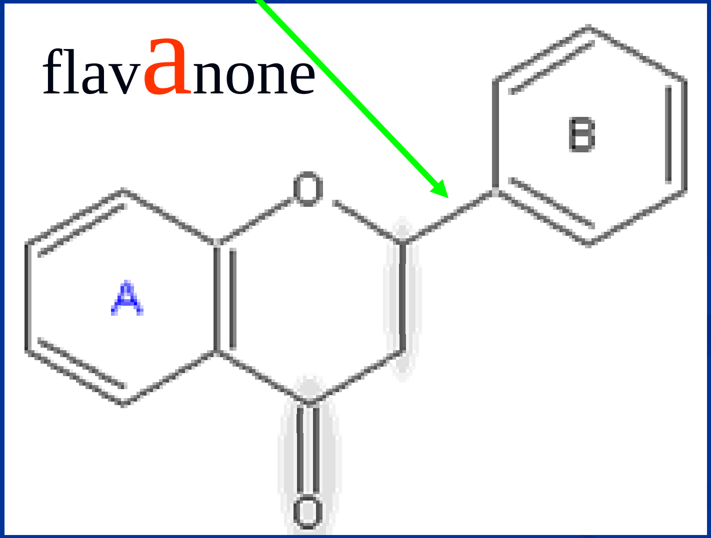
Lepiniec et al., 2006

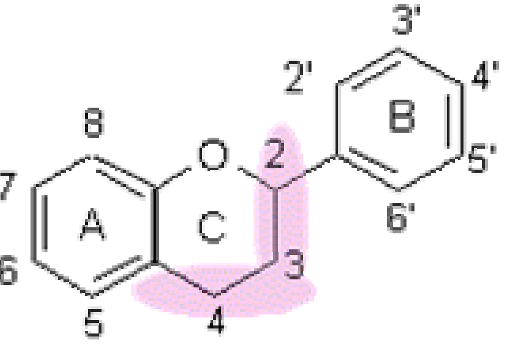
The Flavanones

- No double bond between carbons 2 and 3 of the flavone structure, and chiral center (C2)

- A highly reactive structure (a lot of substitutions)





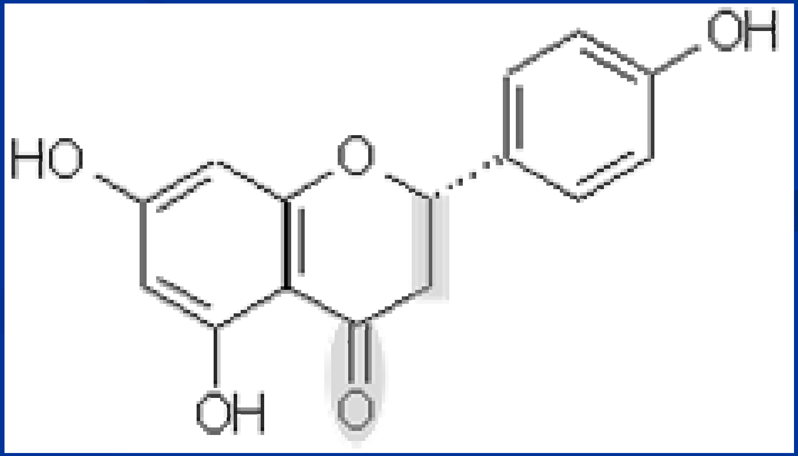


The Flavanones- Naringenin

- An antioxidant flavanone from citrus species

- Has - OH groups attached at positions 5, 7, and 4'

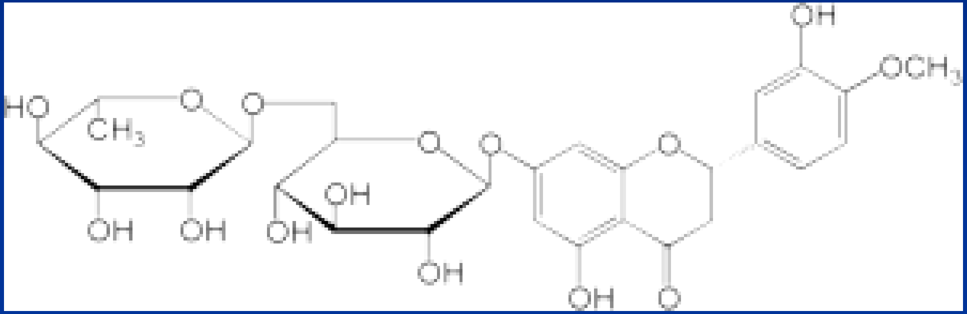
- Studies have indicated that it has anti-inflammatory, anti-cancer, and liver protective effects



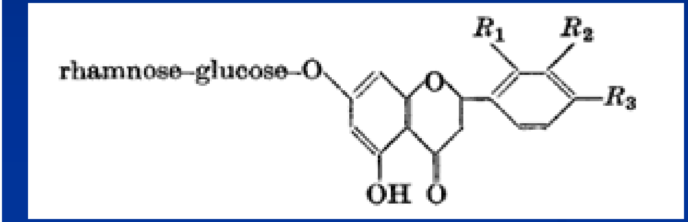
The Flavanones of citrus

The Flavanones- in citrus

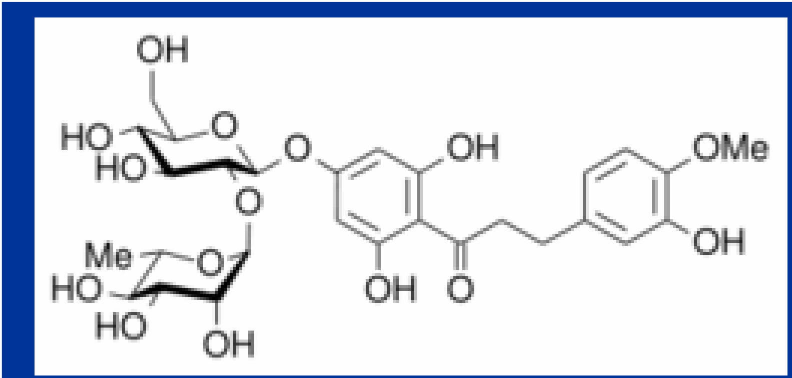
- High concentrations in citrus fruit



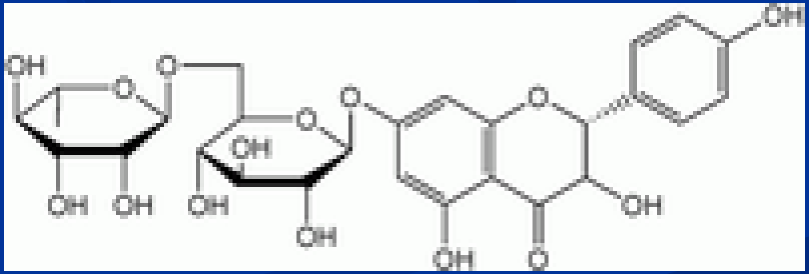
Hesperidin, citrus peel, tasteless



Neohesperidin, bitter orange, intense bitter taste

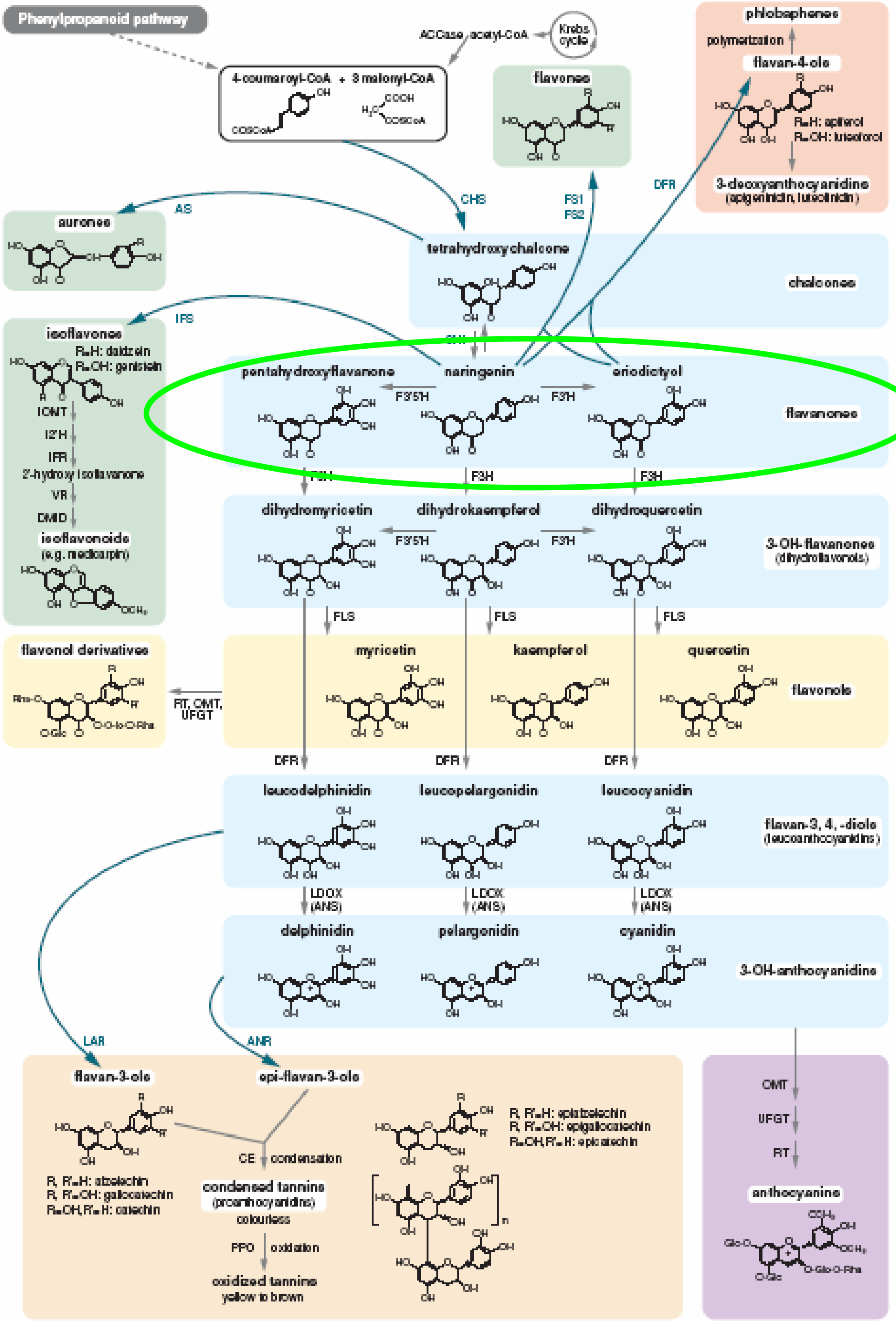


Neohesperidin dihydrochalcon, citrus, arteficial sweetner, in non-alcoholic beers



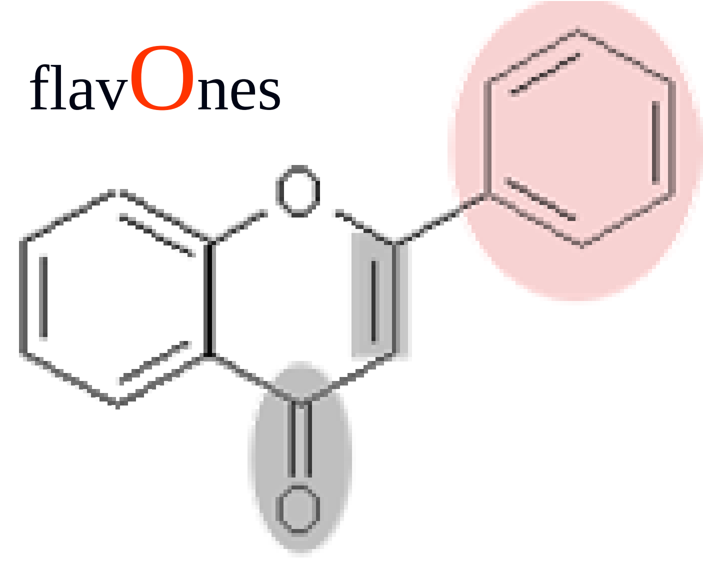
Naringin, grapefruit peel, intense bitter taste

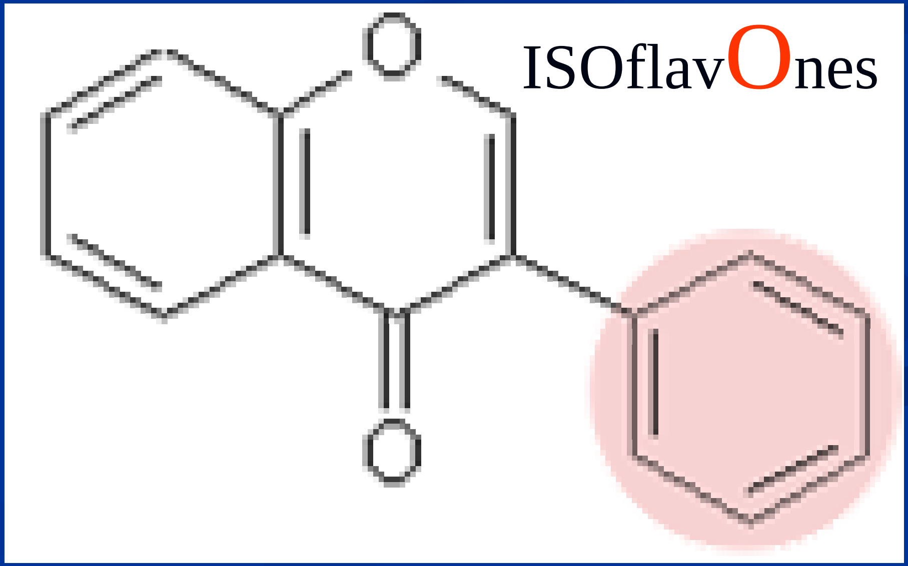
Flavanones Biosynthesis



The Isoflavones (Isoflavonoids)

- Isoflavones are also known as isoflavonoids
- Very similar to flavones, except that the B ring is attached to position 3 of the C ring, rather than to position 2 as in the flavones





The Isoflavones (Isoflavonoids)

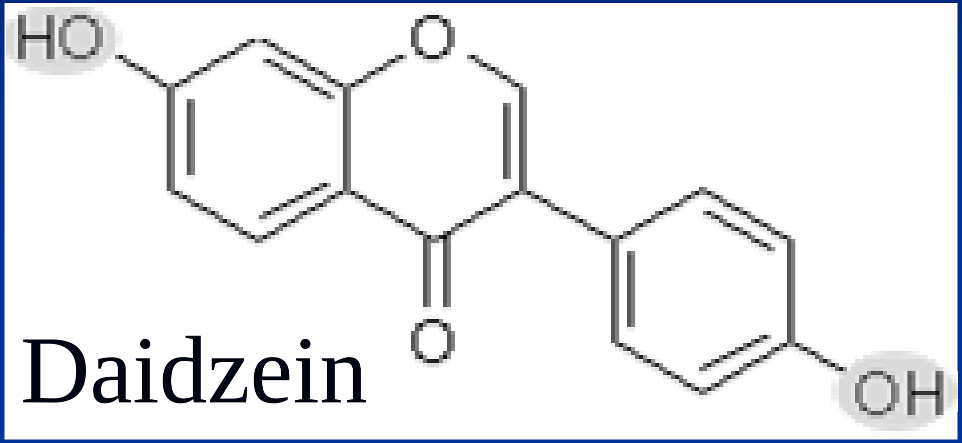
- Found almost exclusively in leguminous plants with highest concentrations in soybean

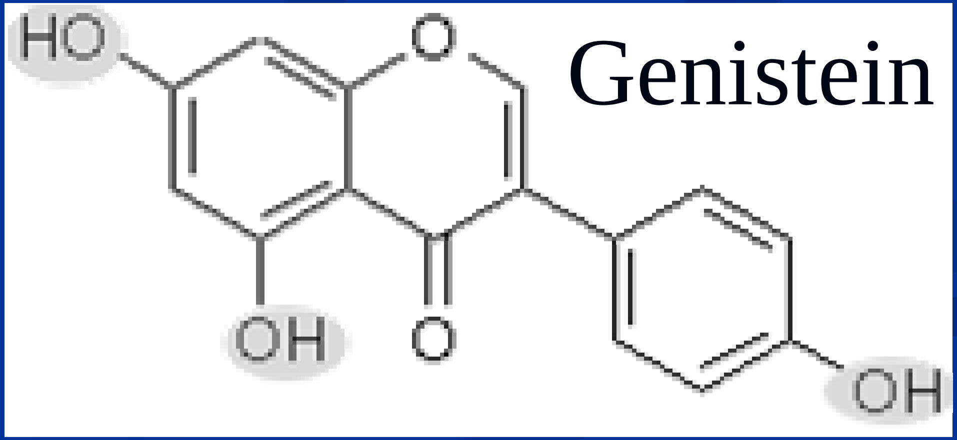
- Genistein, daidzein- phyto-oestrogens (can effect reproduction of grazing animals)

- Structure similar to the steroidal hormone oestradiol which blocks ovulation

- Low isoflavonoid producing varieties are being fed to animals

The Isoflavones (Isoflavonoids)


Daidzein


Genistein

The Isoflavones (Isoflavonoids)

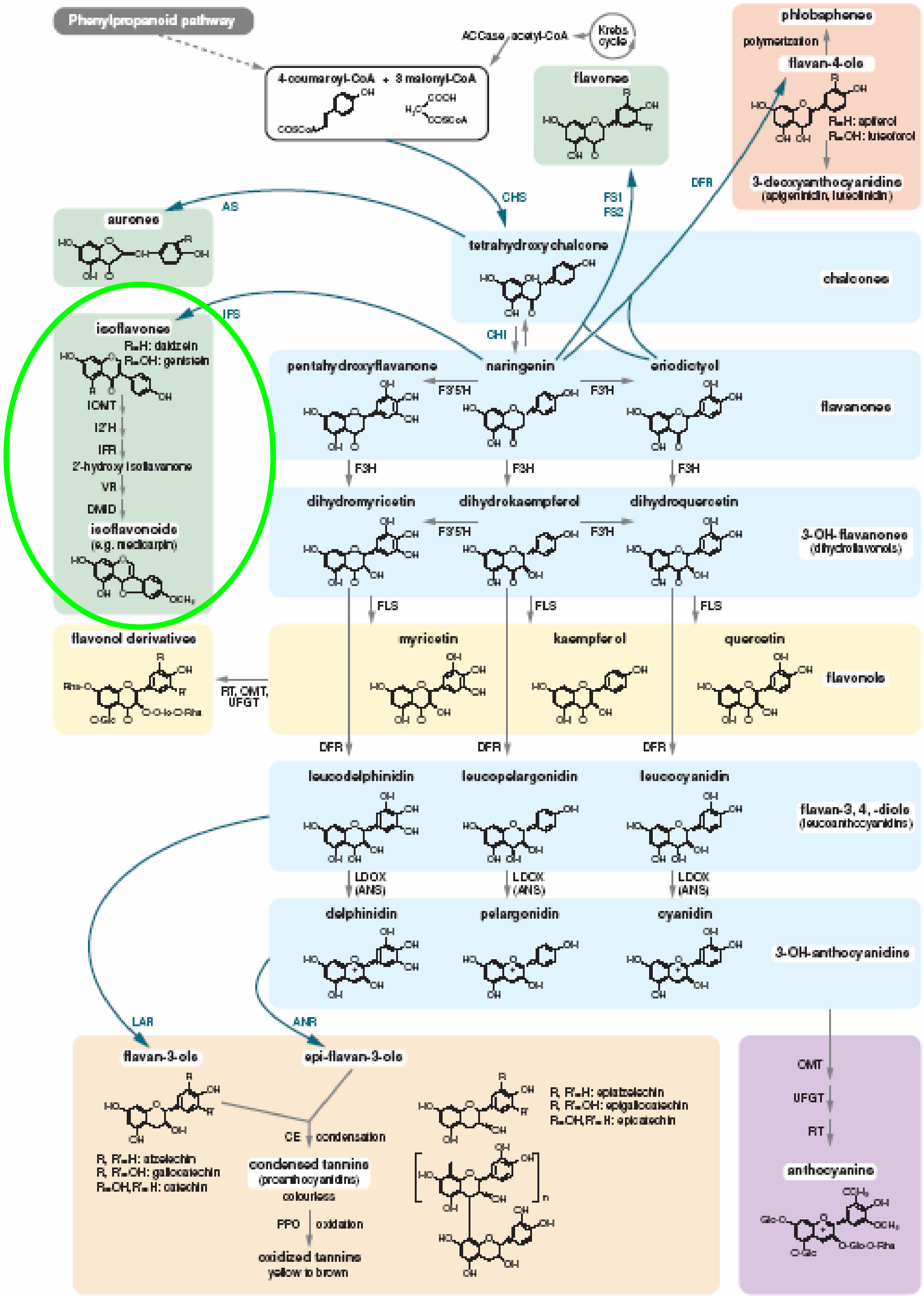
Important for human health:

- Reduce prostate and breast cancer

- In prostate cancer- growth of cancer cells by testosterone but suppressed by oestrradiol. - Isoflavonoids can suppress testosterone when oestradiol is not sufficient

- Anti-inflammatory and show cardioprotective

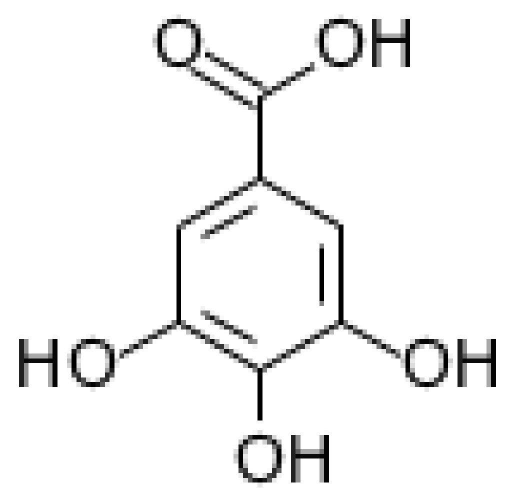
Isoflavones Biosynthesis



Non-Flavonoids- Phenolic acids

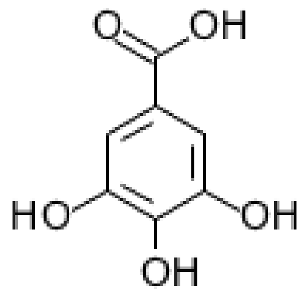
- Also known as hydroxybenzoates

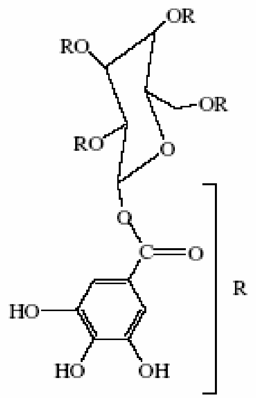
- Principle component is Gallic acid (derived from the shikimate pathway)



Non-Flavonoids- Gallotanines

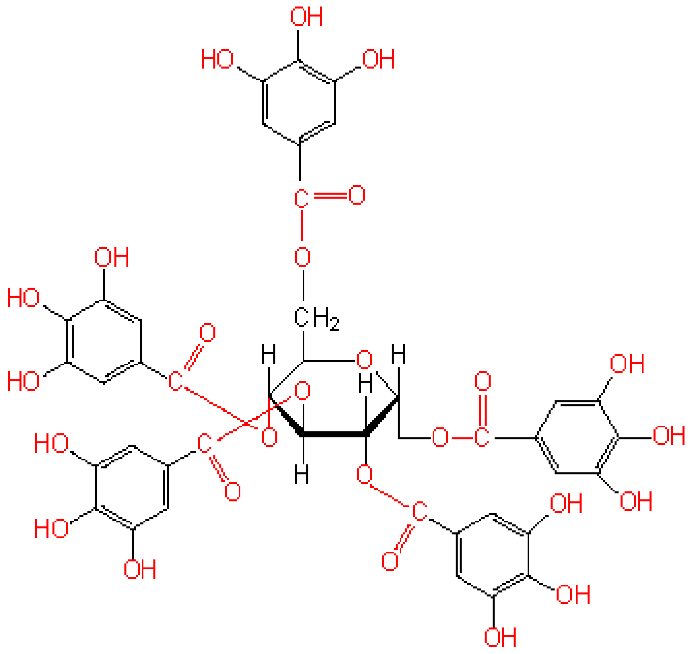
- Gallic acid is the base unit of Gallotanines
- Gallic acid residues linked to Glucose (often) via glycosidic bond (galloyl moiety)
- Gallotanines are hydrolysable tannins, treatment with dilute acids release gallic acid residues





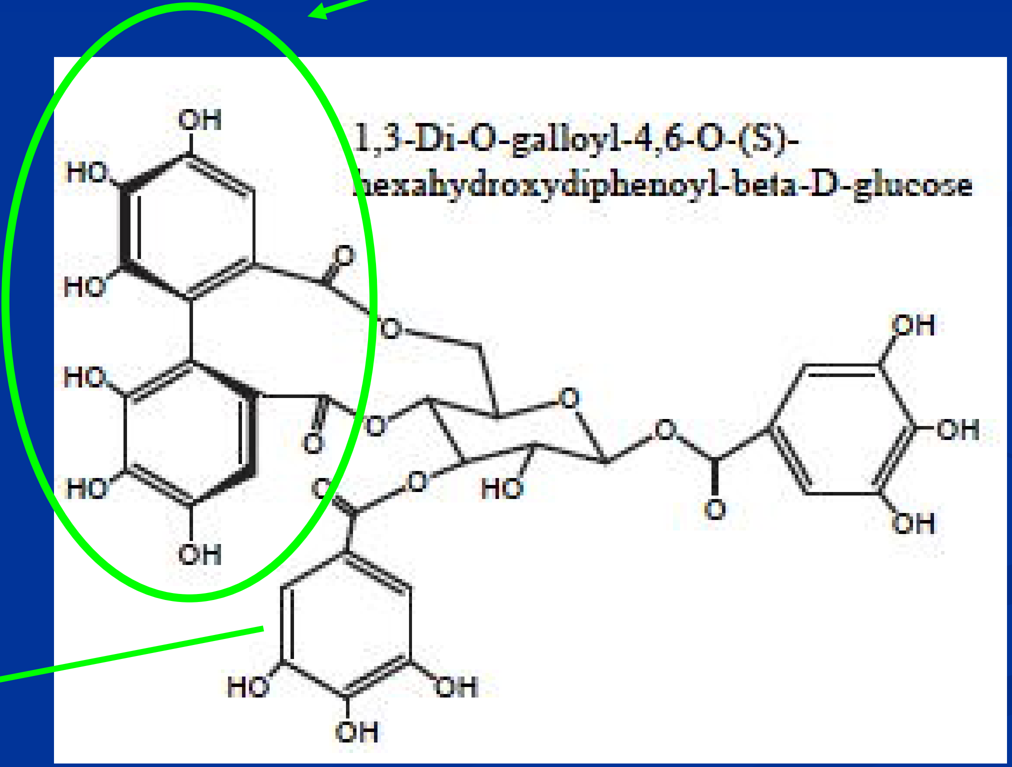
R=Galloyl moiety (G)

Gallotannins

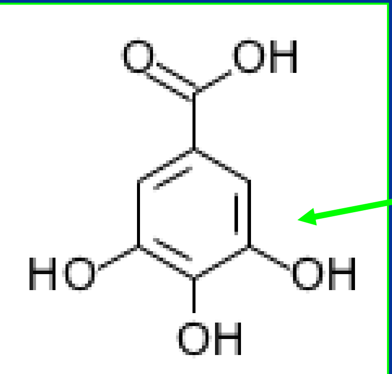


Non-Flavonoids- Ellagitanines

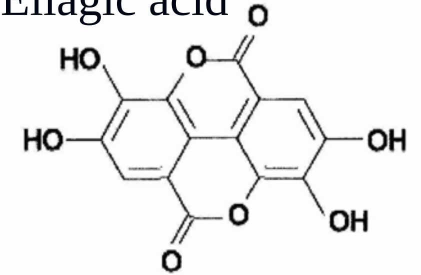
- Called ellagitannins since ellagic acid is released acid
- Composed of Gallic acid and hexahydroxydiphenoyl moieties


1,3-Di-O-galloyl-4,6-O-(S)-hexahydroxydiphenoyl-beta-D-glucose

Gallic acid

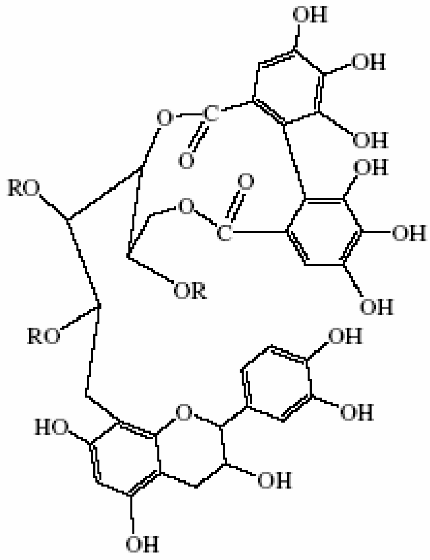


Ellagic acid



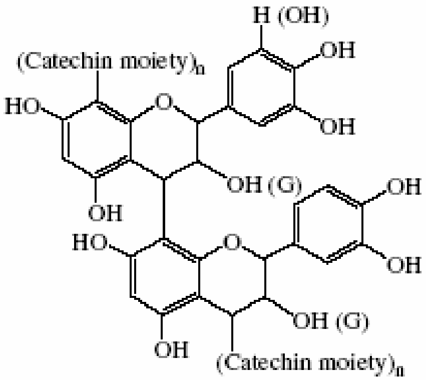
Non-Flavonoids- Type of Tannins

- Complex tannins- catechin or epicatechin bound bound to a galltannin or ellagitannin unit



R=Galloyl moiety (G) or other substituents

Complex Tannins



Condensed Tannins

Non-Flavonoids- General

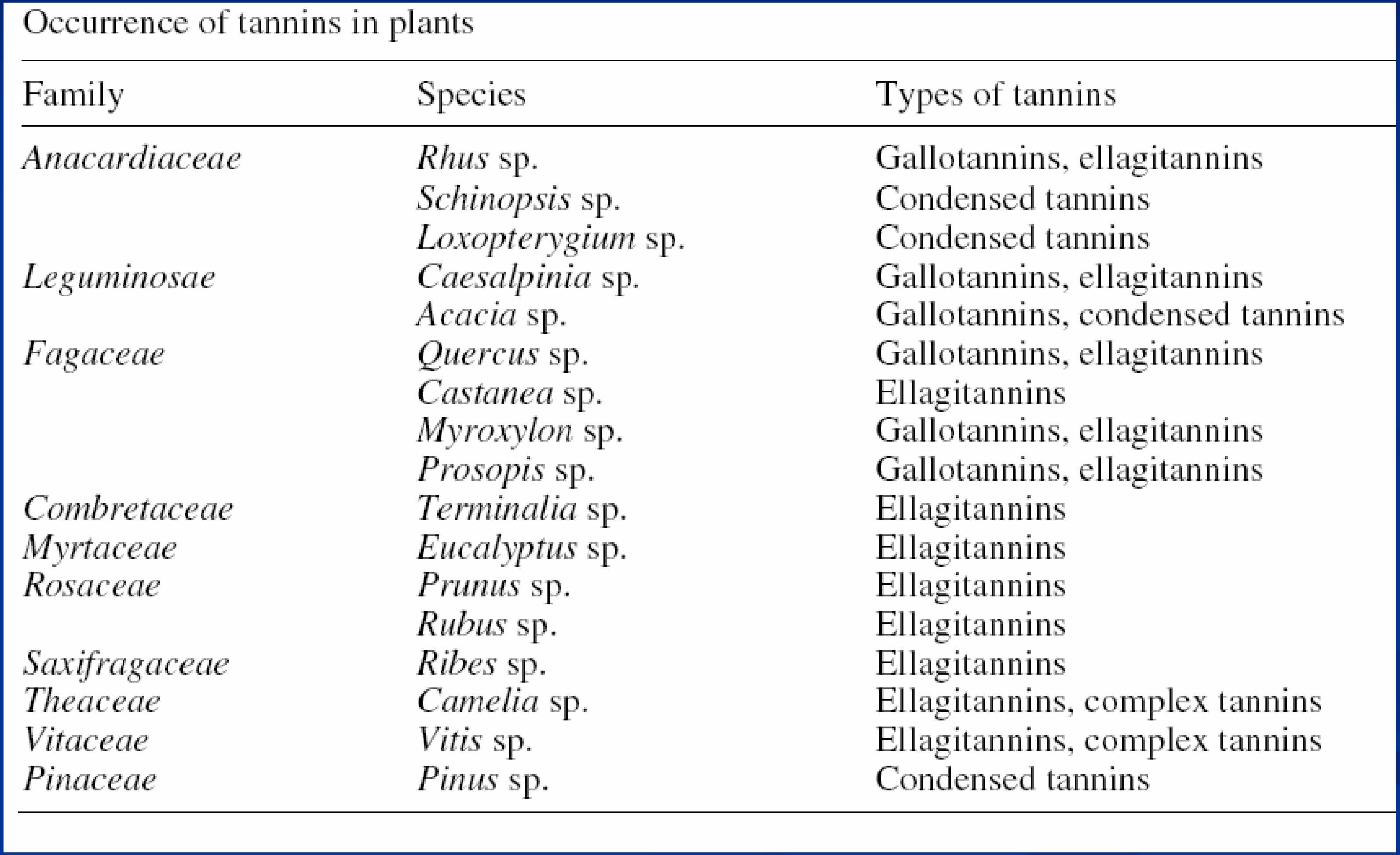
Hydrolysable & Condensed Tannines

- Plant polyphenols that have the ability to precipitate protein- generally called tannins

- Used for a 1000 years to convert raw animal hides into leather

- In this process, tannin molecules cross-link the protein and make it more resistant to bacterial and fungal attack

Non-Flavonoids- Tannines



Occurrence of tannins in plants

Family	Species	Types of tannins
Anacardiaceae	*Rhus* sp.	Gallotannins, ellagitannins
	Schinopsis sp.	Condensed tannins
	Loxopterygium sp.	Condensed tannins
Leguminosae	*Caesalpinia* sp.	Gallotannins, ellagitannins
	Acacia sp.	Gallotannins, condensed tannins
Fagaceae	*Quercus* sp.	Gallotannins, ellagitannins
	Castanea sp.	Ellagitannins
	Myroxylon sp.	Gallotannins, ellagitannins
	Prosopis sp.	Gallotannins, ellagitannins
Combretaceae	*Terminalia* sp.	Ellagitannins
Myrtaceae	*Eucalyptus* sp.	Ellagitannins
Rosaceae	*Prunus* sp.	Ellagitannins
	Rubus sp.	Ellagitannins
Saxifragaceae	*Ribes* sp.	Ellagitannins
Theaceae	*Camelia* sp.	Ellagitannins, complex tannins
Vitaceae	*Vitis* sp.	Ellagitannins, complex tannins
Pinaceae	*Pinus* sp.	Condensed tannins

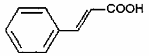
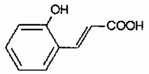
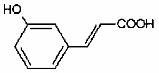
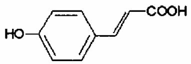
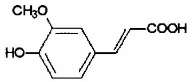
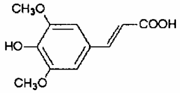
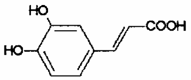
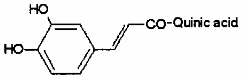
Non-Flavonoids- General

Hydrolysable & Condensed Tannines

- Tannins bind to salivary proteins and making the astringency taste
- In fruit- Astringency in Persimmon, strawberry (boser)
- Astringency (mild) enhances the taste of wine and tea
- Animals such as apes and dear will not eat fruit with high tannins
- In fruit- tannins decline in ripening-evolution for seed dispersal

Non-Flavonoids- Hydroxycinnamates or cinnamic acids

- Generated from cinnamic acid
- They are phenylpropanoids
- Most common: p-coumaric acid, caffeic and ferulic acids

Name	Structure	Source
cinnamic acid		oil of cinnamon, coca leaves
o-coumaric acid		cherry, plum
m-coumaric acid		cherry, plum
p-coumaric acid		most fruits (esp. blueberry, raspberry and pineapple) apple, tomato, grape, olive
ferulic acid		grains, nuts, tumeric, peppers, citrus fruit, tomato, cabbage, asparagus
sinapic acid		brussel sprouts, potatoes, rapeseed; trace amounts in citrus, pineapple, tomato
caffeic acid		grape, apple, plum, tomato, eggplant, cabbage, asparagus, endives, potatoes (the most abundant hydroxycinnamic acid)
chlorogenic acid		apple, pear, peach (and most fruits), tomato, coffee

Non-Flavonoids- Stilbens

- Members of the stilbene family have the C6-C2-C6 structure
- Polyphenolic like flavonoids

- They are phytoalexines, produced in response to fungal, bacterial, viral attack

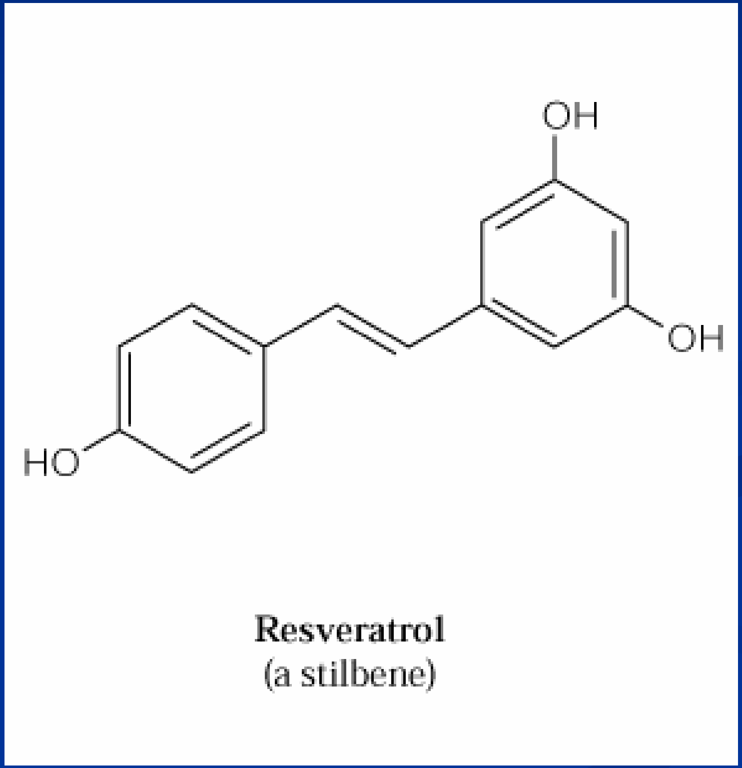
- Resveratrol, the most common stilbene

- Major source: grape, wine, peanut products and soya

- trans-resveratrol and its glucoside are the active agents in the famous Itadori root ("well being" in Japanese)

- Cardio protective effects of red wine, can inhibit LDL oxidation which is the initial stage of atherosclerosis

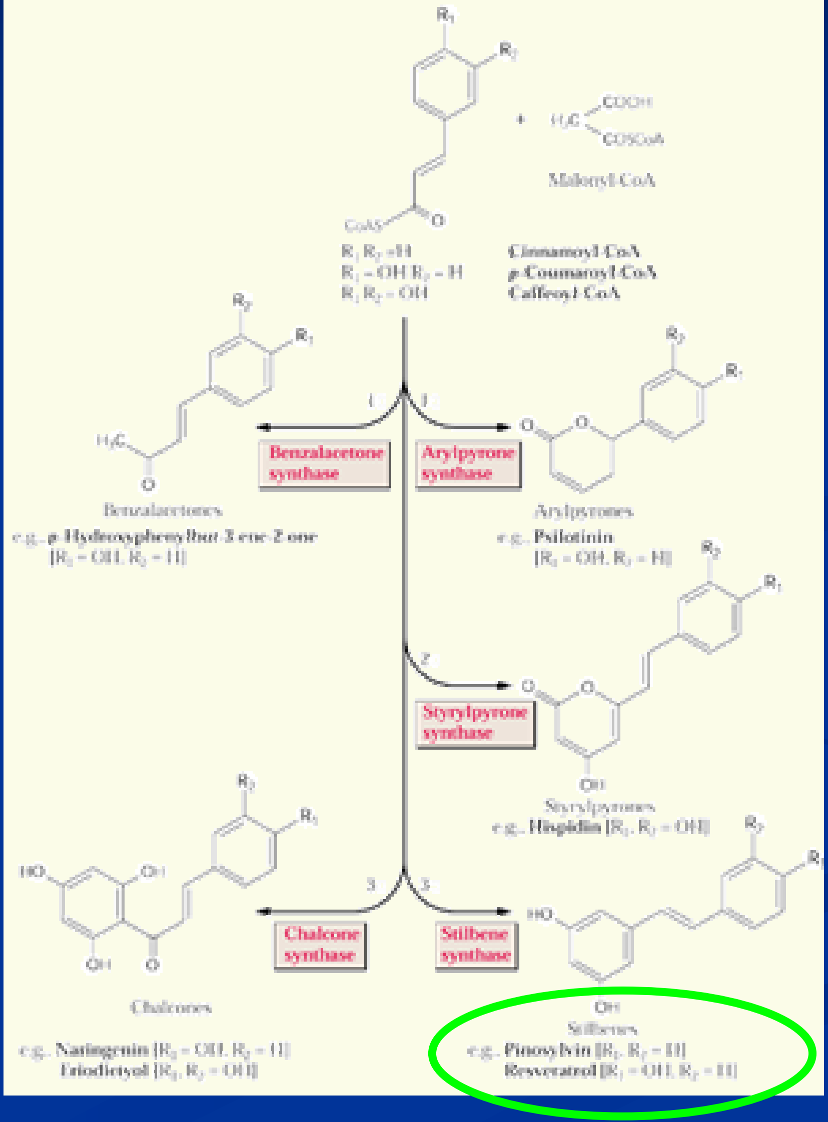
Non-Flavonoids- Stilbens- Resveratrol



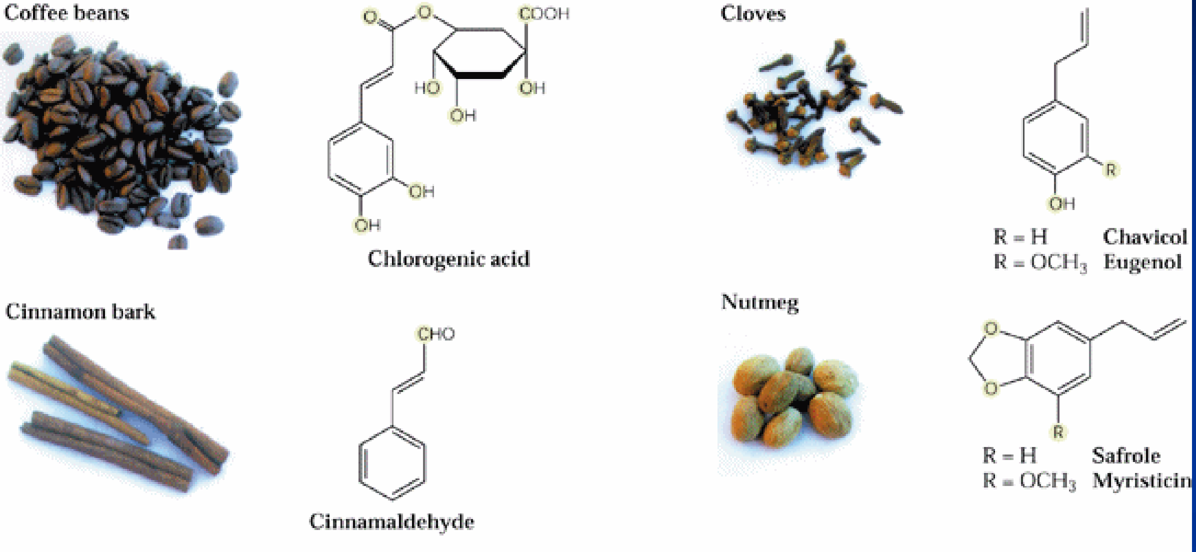
Resveratrol
(a stilbene)

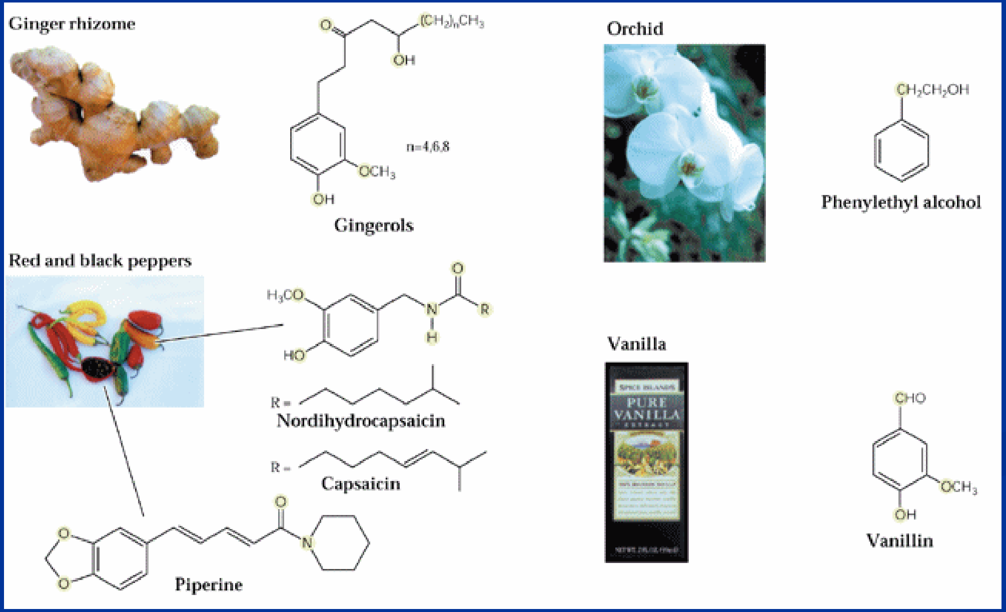
Also has potent anti-tumer activity

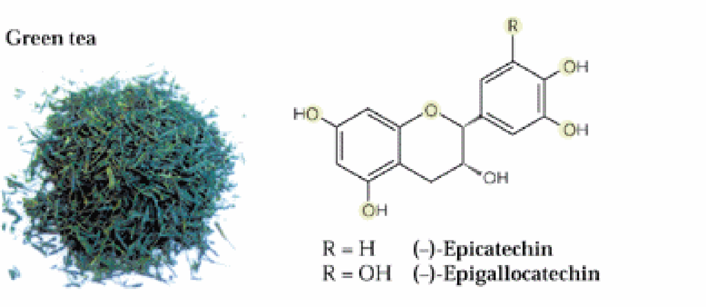
Non-Flavonoids- Stilbens- Resveratrol



Phenylypropanoids & flavour/fragrance







Nucleic acids

Nucleic acid, naturally occurring chemical compound that is capable of being broken down to yield phosphoric acid, sugars, and a mixture of organic bases (purines and pyrimidines). Nucleic acids are the main information-carrying molecules of the cell, and, by directing the process of protein synthesis, they determine the inherited characteristics of every living thing. The two main classes of nucleic acids are deoxyribonucleic acid (DNA) and ribonucleic acid (RNA). DNA is the master blueprint for life and constitutes the genetic material in all free-living organisms and most viruses. RNA is the genetic material of certain viruses, but it is also found in all living cells, where it plays an important role in certain processes such as the making of proteins.

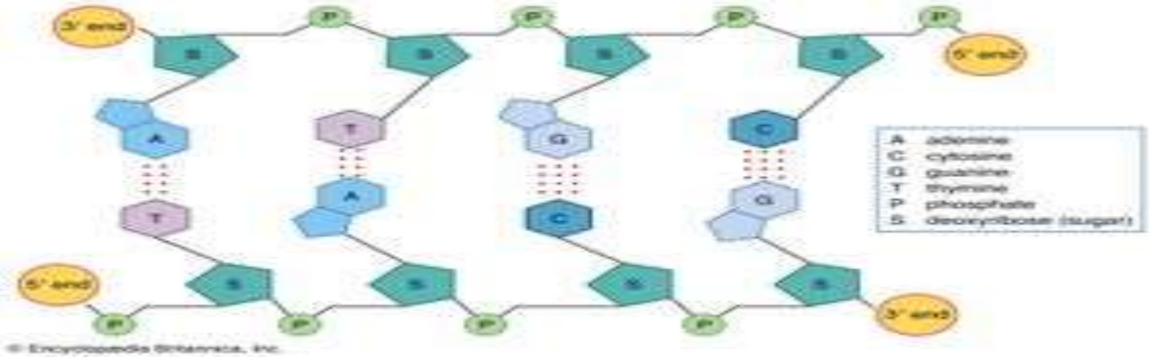
Nucleic acids are polynucleotides—that is, long chainlike molecules composed of a series of nearly identical building blocks called nucleotides. Each nucleotide consists of a nitrogen-containing aromatic base attached to a pentose (five-carbon) sugar, which is in turn attached to a phosphate group. Each nucleic acid contains four of five possible nitrogen-containing bases: adenine (A), guanine (G), cytosine (C), thymine (T), and uracil (U). A and G are categorized as purines, and C, T, and U are collectively called pyrimidines. All nucleic acids contain the bases A, C, and G; T, however, is found only in DNA, while U is found in RNA. The pentose sugar in DNA (2′-deoxyribose) differs from the sugar in RNA (ribose) by the absence of a hydroxyl group (—OH) on the 2′ carbon of the sugar ring. Without an attached phosphate group, the sugar attached to one of the bases is known as a nucleoside. The phosphate group connects successive sugar residues by bridging the 5′-hydroxyl group on one sugar to the 3′-hydroxyl group of the next sugar in the chain. These nucleoside linkages are called phosphodiester bonds and are the same in RNA and DNA.

Biosynthesis and degradation

Nucleotides are synthesized from readily available precursors in the cell. The ribose phosphate portion of both purine and pyrimidine nucleotides is synthesized from glucose via the pentose phosphate pathway. The six-atom pyrimidine ring is synthesized first and subsequently attached to the ribose phosphate. The two rings in purines are synthesized while attached to the ribose phosphate during the assembly of adenine or guanine nucleosides. In both cases the end product is a nucleotide carrying a phosphate attached to the 5′ carbon on the sugar. Finally, a specialized enzyme called a kinase adds two phosphate groups using adenosine triphosphate (ATP) as the phosphate donor to form ribonucleoside triphosphate, the immediate precursor of RNA. For DNA, the 2′-hydroxyl group is removed from the ribonucleoside diphosphate to give deoxyribonucleoside diphosphate. An additional phosphate group from ATP is then added by another kinase to form a deoxyribonucleoside triphosphate, the immediate precursor of DNA.

During normal cell metabolism, RNA is constantly being made and broken down. The purine and pyrimidine residues are reused by several salvage pathways to make more genetic material. Purine is salvaged in the form of the corresponding nucleotide, whereas pyrimidine is salvaged as the nucleoside.

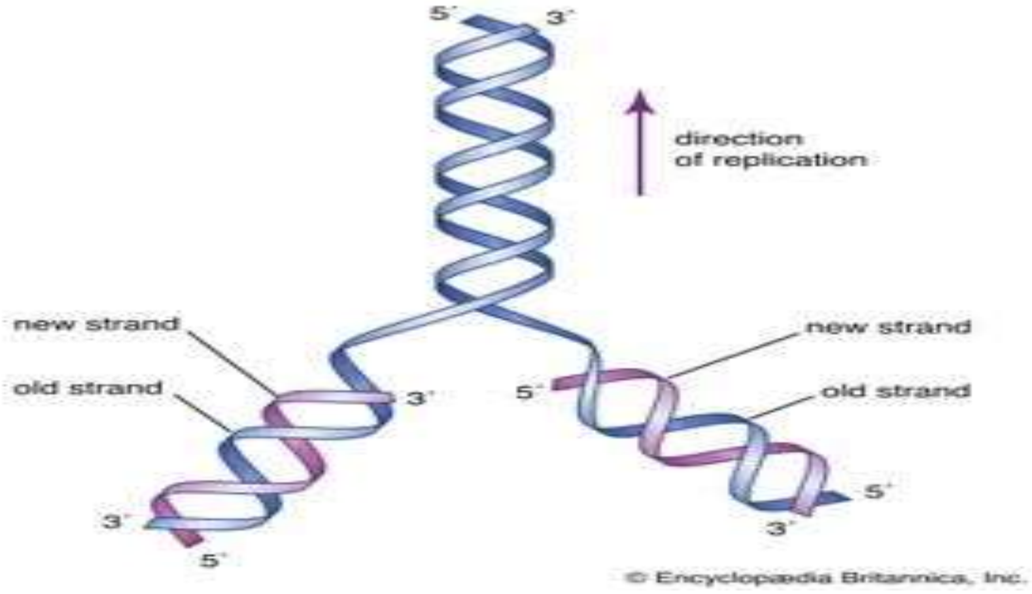
Deoxyribonucleic acid (DNA)



DNA structure

DNA is a polymer of the four nucleotides A, C, G, and T, which are joined through a backbone of alternating phosphate and deoxyribose sugar residues. These nitrogen-containing bases occur in complementary pairs as determined by their ability to form hydrogen bonds between them. A always pairs with T through two hydrogen bonds, and G always pairs with C through three hydrogen bonds. The spans of A:T and G:C hydrogen-bonded pairs are nearly identical, allowing them to bridge the sugar-phosphate chains uniformly. This structure, along with the molecule’s chemical stability, makes DNA the ideal genetic material. The bonding between complementary bases also provides a mechanism for the replication of DNA and the transmission of genetic information.

Chemical structure



Initial proposal of DNA structure

In 1953 James D. Watson and Francis H.C. Crick proposed a three-dimensional structure for DNA based on low-resolution X-ray crystallographic data and on Erwin Chargaff's observation that, in naturally occurring DNA, the amount of T equals the amount of A and the amount of G equals the amount of C. Watson and Crick, who shared a Nobel Prize in 1962 for their efforts, postulated that two strands of polynucleotides coil around each other, forming a double helix. The two strands, though identical, run in opposite directions as determined by the orientation of the 5′ to 3′ phosphodiester bond. The sugar-phosphate chains run along the outside of the helix, and the bases lie on the inside, where they are linked to complementary bases on the other strand through hydrogen bonds.

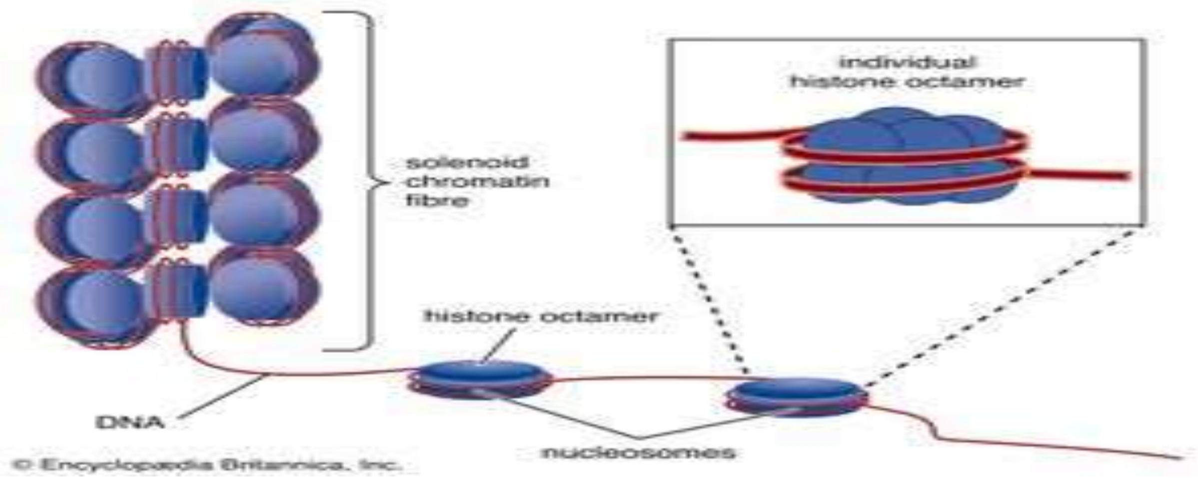
The double helical structure of normal DNA takes a right-handed form called the B-helix. The helix makes one complete turn approximately every 10 base pairs. B-DNA has two principal grooves, a wide major groove and a narrow minor groove. Many proteins interact in the space of the major groove, where they make sequence-specific contacts with the bases. In addition, a few proteins are known to make contacts via the minor groove.

Several structural variants of DNA are known. In A-DNA, which forms under conditions of high salt concentration and minimal water, the base pairs are tilted and displaced toward the minor groove. Left-handed Z-DNA forms most readily in strands that contain sequences with alternating purines and pyrimidines. DNA can form triple helices when two strands containing runs of pyrimidines interact with a third strand containing a run of purines.

B-DNA is generally depicted as a smooth helix; however, specific sequences of bases can distort the otherwise regular structure. For example, short tracts of A residues interspersed with short sections of general sequence result in a bent DNA molecule. Inverted base sequences, on the other hand, produce cruciform structures with four-way junctions that are similar to recombination intermediates. Most of these alternative DNA structures have only been characterized in the laboratory, and their cellular significance is unknown.

Biological structures

Naturally occurring DNA molecules can be circular or linear. The genomes of single-celled bacteria and archaea (the prokaryotes), as well as the genomes of mitochondria and chloroplasts (certain functional structures within the cell), are circular molecules. In addition, some bacteria and archaea have smaller circular DNA molecules called plasmids that typically contain only a few genes. Many plasmids are readily transmitted from one cell to another. For a typical bacterium, the genome that encodes all of the genes of the organism is a single contiguous circular molecule that contains a half million to five million base pairs. The genomes of most eukaryotes and some prokaryotes contain linear DNA molecules called chromosomes. Human DNA, for example, consists of 23 pairs of linear chromosomes containing three billion base pairs.



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In all cells, DNA does not exist free in solution but rather as a protein-coated complex called chromatin. In prokaryotes, the loose coat of proteins on the DNA helps to shield the negative charge of the phosphodiester backbone. Chromatin also contains proteins that control gene expression and determine the characteristic shapes of chromosomes. In eukaryotes, a section of DNA between 140 and 200 base pairs long winds around a discrete set of eight positively charged proteins called a histone, forming a spherical structure called the nucleosome. Additional histones are wrapped by successive sections of DNA, forming a series of nucleosomes like beads on a string. Transcription and replication of DNA is more complicated in eukaryotes because the nucleosome complexes have to be at least partially disassembled for the processes to proceed effectively.

Most prokaryote viruses contain linear genomes that typically are much shorter and contain only the genes necessary for viral propagation. Bacterial viruses called bacteriophages (or phages) may contain both linear and circular forms of DNA. For instance, the genome of bacteriophage λ (lambda), which infects the bacterium *Escherichia coli*, contains 48,502 base pairs and can exist as a linear molecule packaged in a protein coat. The DNA of phage λ can also exist in a circular form (as described in the section Site-specific recombination) that is able to integrate into the circular genome of the host bacterial cell. Both circular and linear genomes are found among eukaryotic viruses, but they more commonly use RNA as the genetic material.

Biochemical properties

Denaturation

The strands of the DNA double helix are held together by hydrogen bonding interactions between the complementary base pairs. Heating DNA in solution easily breaks these hydrogen bonds, allowing the two strands to separate—a process called denaturation or melting. The two strands may reassociate when the solution cools, reforming the starting DNA duplex—a process called renaturation or hybridization. These processes form the basis of many important techniques for manipulating DNA. For example, a short piece of DNA called an oligonucleotide can be used to test whether a very long DNA sequence has the complementary sequence of the oligonucleotide embedded within it. Using hybridization, a single-stranded DNA molecule can capture complementary sequences from any source. Single strands from RNA can also reassociate. DNA and RNA single strands can form hybrid molecules that are even more stable than double-stranded DNA. These molecules form the basis of a technique that is used to purify and characterize messenger RNA (mRNA) molecules corresponding to single genes.

Ultraviolet absorption

DNA melting and reassociation can be monitored by measuring the absorption of ultraviolet (UV) light at a wavelength of 260 nanometres (billionths of a metre). When DNA is in a double-stranded conformation, absorption is fairly weak, but when DNA is single-stranded, the unstacking of the bases leads to an enhancement of absorption called hyperchromicity. Therefore, the extent to which DNA is single-stranded or double-stranded can be determined by monitoring UV absorption.

Chemical modification

After a DNA molecule has been assembled, it may be chemically modified—sometimes deliberately by special enzymes called DNA methyltransferases and sometimes accidentally by oxidation, ionizing radiation, or the action of chemical carcinogens. DNA can also be cleaved and degraded by enzymes called nucleases.

Methylation

Three types of natural methylation have been reported in DNA. Cytosine can be modified either on the ring to form 5-methylcytosine or on the exocyclic amino group to form N^4-methylcytosine. Adenine may be modified to form N^6-methyladenine. N^4-methylcytosine and N^6-methyladenine are found only in bacteria and archaea, whereas 5-methylcytosine is widely distributed. Special enzymes called DNA methyltransferases are responsible for this methylation; they recognize specific sequences within the DNA molecule so that only a subset of the bases is modified. Other methylations of the bases or of the deoxyribose are sometimes induced by carcinogens. These usually lead to mispairing of the bases during replication and have to be removed if they are not to become mutagenic.

Natural methylation has many cellular functions. In bacteria and archaea, methylation forms an essential part of the immune system by protecting DNA molecules from fragmentation by restriction endonucleases. In some organisms, methylation helps to eliminate incorrect base sequences introduced during DNA replication. By marking the parental strand with a methyl group, a cellular mechanism known as the mismatch repair system distinguishes between the newly replicated strand where the errors occur and the correct sequence on the template strand. In higher eukaryotes, 5-methylcytosine controls many cellular phenomena by preventing DNA transcription. Methylation is also believed to signal imprinting, a process whereby some genes inherited from one parent are selectively inactivated. Correct methylation may also repress or activate key genes that control embryonic development. On the other hand, 5-methylcytosine is potentially mutagenic because thymine produced during the methylation process converts C:G pairs to T:A pairs. In mammals, methylation takes place selectively within the dinucleotide sequence CG—a rare sequence, presumably because it has been lost by mutation. In many cancers, mutations are found in key genes at CG dinucleotides.

Nucleases

Nucleases are enzymes that hydrolytically cleave the phosphodiester backbone of DNA. Endonucleases cleave in the middle of chains, while exonucleases operate selectively by degrading from the end of the chain. Nucleases that act on both single- and double-stranded DNA are known. Restriction endonucleases are a special class that recognize and cleave specific sequences in DNA. Type II restriction endonucleases always cleave at or near their recognition sites. They produce small, well-defined fragments of DNA that help to characterize genes and genomes and that produce recombinant DNAs. Fragments of DNA produced by restriction endonucleases can be moved from one organism to another. In this way it has been possible to express proteins such as human insulin in bacteria.

Mutation

Chemical modification of DNA can lead to mutations in the genetic material. Anions such as bisulfite can deaminate cytosine to form uracil, changing the genetic message by causing C-to-T transitions. Exposure to acid causes the loss of purine residues, though specific enzymes exist in cells to repair these lesions. Exposure to UV light can cause adjacent pyrimidines to dimerize, while oxidative damage from free radicals or strong oxidizing agents can cause a variety of lesions that are mutagenic if not repaired. Halogens such as chlorine and bromine react directly with uracil, adenine, and guanine, giving substituted bases that are often mutagenic. Similarly, nitrous acid reacts with primary amine groups—for example, converting adenosine into inosine—which then leads to changes in base pairing and mutation. Many chemical mutagens, such as chlorinated hydrocarbons and nitrites, owe their toxicity to the production of halides and nitrous acid during their metabolism in the body.

Supercoiling

Circular DNA molecules such as those found in plasmids or bacterial chromosomes can adopt many different topologies. One is active supercoiling, which involves the cleavage of one DNA strand, its winding one or more turns around the complementary strand, and then the resealing of the molecule. Each complete rotation leads to the introduction of one supercoiled turn in the DNA, a process that can continue until the DNA is fully wound and collapses on itself in a tight ball. Reversal is also possible. Special enzymes called gyrases and topoisomerases catalyze the winding and relaxation of supercoiled DNA. In the linear chromosomes of eukaryotes, the DNA is usually tightly constrained at various points by proteins, allowing the intervening stretches to be supercoiled. This property is partially responsible for the great compaction of DNA that is necessary to fit it within the confines of the cell. The DNA in one human cell would have an extended length of between two and three metres, but it is packed very tightly so that it can fit within a human cell nucleus that is 10 micrometres in diameter.

Sequence determination

Methods to determine the sequences of bases in DNA were pioneered in the 1970s by Frederick Sanger and Walter Gilbert, whose efforts won them a Nobel Prize in 1980. The Gilbert-Maxam method relies on the different chemical reactivities of the bases, while the Sanger method is based on enzymatic synthesis of DNA in vitro. Both methods measure the distance from a fixed point on DNA to each occurrence of a particular base—A, C, G, or T. DNA fragments obtained from a series of reactions are separated according to length in four “lanes” by gel electrophoresis. Each lane corresponds to a unique base, and the sequence is read directly from the gel. The Sanger method has now been automated using fluorescent dyes to label the DNA, and a single machine can produce tens of thousands of DNA base sequences in a single run.

Ribonucleic acid (RNA)

RNA is a single-stranded nucleic acid polymer of the four nucleotides A, C, G, and U joined through a backbone of alternating phosphate and ribose sugar residues. It is the first intermediate in converting the information from DNA into proteins essential for the working of a cell. Some RNAs also serve direct roles in cellular metabolism. RNA is made by copying the base sequence of a section of double-stranded DNA, called a gene, into a piece of single-stranded nucleic acid. This process, called transcription (*see below* RNA metabolism), is catalyzed by an enzyme called RNA polymerase.

Chemical structure

Whereas DNA provides the genetic information for the cell and is inherently quite stable, RNA has many roles and is much more reactive chemically. RNA is sensitive to oxidizing agents such as periodate that lead to opening of the 3′-terminal ribose ring. The 2′-hydroxyl group on the ribose ring is a major cause of instability in RNA, because the presence of alkali leads to rapid cleavage of the phosphodiester bond linking ribose and phosphate groups. In general, this instability is not a significant problem for the cell, because RNA is constantly being synthesized and degraded.

Interactions between the nitrogen-containing bases differ in DNA and RNA. In DNA, which is usually double-stranded, the bases in one strand pair with complementary bases in a second DNA strand. In RNA, which is usually single-stranded, the bases pair with other bases within the same molecule, leading to complex three-dimensional structures. Occasionally, intermolecular RNA/RNA duplexes do form, but they form a right-handed A-type helix rather than the B-type DNA helix. Depending on the amount of salt present, either 11 or 12 base pairs are found in each turn of the helix. Helices between RNA and DNA molecules also form; these adopt the A-type conformation and are more stable than either RNA/RNA or DNA/DNA duplexes. Such hybrid duplexes are important species in biology, being formed when RNA polymerase transcribes DNA into mRNA for protein synthesis and when reverse transcriptase copies a viral RNA genome such as that of the human immunodeficiency virus (HIV).

Single-stranded RNAs are flexible molecules that form a variety of structures through internal base pairing and additional non-base pair interactions. They can form hairpin loops such as those found in transfer RNA (tRNA), as well as longer-range interactions involving both the bases and the phosphate residues of two or more nucleotides. This leads to compact three-dimensional structures.

Most of these structures have been inferred from biochemical data, since few crystallographic images are available for RNA molecules. In some types of RNA, a large number of bases are modified after the RNA is transcribed. More than 90 different modifications have been documented, including extensive methylations and a wide variety of substitutions around the ring. In some cases these modifications are known to affect structure and are essential for function.

Types of RNA

Messenger RNA (mRNA)

Messenger RNA (mRNA) delivers the information encoded in one or more genes from the DNA to the ribosome, a specialized structure, or organelle, where that information is decoded into a protein. In prokaryotes, mRNAs contain an exact transcribed copy of the original DNA sequence with a terminal 5′-triphosphate group and a 3′-hydroxyl residue. In eukaryotes the mRNA molecules are more elaborate. The 5′-triphosphate residue is further esterified, forming a structure called a cap. At the 3′ ends, eukaryotic mRNAs typically contain long runs of adenosine residues (polyA) that are not encoded in the DNA but are added enzymatically after transcription. Eukaryotic mRNA molecules are usually composed of small segments of the original gene and are generated by a process of cleavage and rejoining from an original precursor RNA (pre-mRNA) molecule, which is an exact copy of the gene (as described in the section Splicing). In general, prokaryotic mRNAs are degraded very rapidly, whereas the cap structure and the polyA tail of eukaryotic mRNAs greatly enhance their stability.

Ribosomal RNA (rRNA)

Ribosomal RNA (rRNA) molecules are the structural components of the ribosome. The rRNAs form extensive secondary structures and play an active role in recognizing conserved portions of mRNAs and tRNAs. They also assist with the catalysis of protein synthesis. In the prokaryote *E. coli*, seven copies of the rRNA genes synthesize about 15,000 ribosomes per cell. In eukaryotes the numbers are much larger. Anywhere from 50 to 5,000 sets of rRNA genes and as many as 10 million ribosomes may be present in a single cell. In eukaryotes these rRNA genes are looped out of the main chromosomal fibres and coalesce in the presence of proteins to form an organelle called the nucleolus. The nucleolus is where the rRNA genes are transcribed and the early assembly of ribosomes takes place.

Transfer RNA (tRNA)

Transfer RNA (tRNA) carries individual amino acids into the ribosome for assembly into the growing polypeptide chain. The tRNA molecules contain 70 to 80 nucleotides and fold into a characteristic cloverleaf structure. Specialized tRNAs exist for each of the 20 amino acids needed for protein synthesis, and in many cases more than one tRNA for each amino acid is present. The nucleotide sequence is converted into a protein sequence by translating each three-base sequence (called a codon) with a specific protein. The 61 codons used to code amino acids can be read by many fewer than 61 distinct tRNAs (as described in the section Translation). In *E. coli* a total of 40 different tRNAs are used to translate the 61 codons. The amino acids are loaded onto the tRNAs by specialized enzymes called aminoacyl tRNA synthetases, usually with one synthetase for each amino acid. However, in some organisms, less than the full complement of 20 synthetases are required because some amino acids, such as glutamine and asparagine, can be synthesized on their respective tRNAs. All tRNAs adopt similar structures because they all have to interact with the same sites on the ribosome.

Ribozymes

Not all catalysis within the cell is carried out exclusively by proteins. Thomas Cech and Sidney Altman, jointly awarded a Nobel Prize in 1989, discovered that certain RNAs, now known as ribozymes, showed enzymatic activity. Cech showed that a noncoding sequence (intron) in the small subunit rRNA of protozoans, which had to be removed before the rRNA was functional, can excise itself from a much longer precursor RNA molecule and rejoin the two ends in an autocatalytic reaction. Altman showed that the RNA component of an RNA protein complex called ribonuclease P can cleave a precursor tRNA to generate a mature tRNA. In addition to self-splicing RNAs similar to the one discovered by Cech, artificial RNAs have been made that show a variety of catalytic reactions. It is now widely held that there was a stage during evolution when only RNA catalyzed and stored genetic information. This period, sometimes called “the RNA world,” is believed to have preceded the function of DNA as genetic material.

Antisense RNAs

Most antisense RNAs are synthetically modified derivatives of RNA or DNA with potential therapeutic value. In nature, antisense RNAs contain sequences that are the complement of the normal coding sequences found in mRNAs (also called sense RNAs). Like mRNAs, antisense RNAs are single-stranded, but they cannot be translated into protein. They can inactivate their complementary mRNA by forming a double-stranded structure that blocks the translation of the base sequence. Artificially introducing antisense RNAs into cells selectively inactivates genes by interfering with normal RNA metabolism.

Viral genomes

Many viruses use RNA for their genetic material. This is most prevalent among eukaryotic viruses, but a few prokaryotic RNA viruses are also known. Some common examples include poliovirus, human immunodeficiency virus (HIV), and influenza virus, all of which affect humans, and tobacco mosaic virus, which infects plants. In some viruses the entire genetic material is encoded in a single RNA molecule, while in the segmented RNA viruses several RNA molecules may be present. Many RNA viruses such as HIV use a specialized enzyme called reverse transcriptase that permits replication of the virus through a DNA intermediate. In some cases this DNA intermediate becomes integrated into the host chromosome during infection; the virus then exists in a dormant state and effectively evades the host immune system.

Other RNAs

Many other small RNA molecules with specialized functions are present in cells. For example, small nuclear RNAs (snRNAs) are involved in RNA splicing (*see below*), and other small RNAs that form part of the enzymes telomerase or ribonuclease P are part of ribonucleoprotein particles. The RNA component of telomerase contains a short sequence that serves as a template for the addition of small strings of oligonucleotides at the ends of eukaryotic chromosomes. Other RNA molecules serve as guide RNAs for editing, or they are complementary to small sections of rRNA and either direct the positions at which methyl groups need to be added or mark U residues for conversion to the isomer pseudouridine.

RNA processing

Cleavage

Following synthesis by transcription, most RNA molecules are processed before reaching their final form. Many rRNA molecules are cleaved from much larger transcripts and may also be methylated or enzymatically modified. In addition, tRNAs are usually formed as longer precursor molecules that are cleaved by ribonuclease P to generate the mature 5′ end and often have extra residues added to their 3′ end to form the sequence CCA. The hydroxyl group on the ribose ring of the terminal A of the 3′-CCA sequence acts as the amino acid acceptor necessary for the function of RNA in protein building.

Splicing

In prokaryotes the protein coding sequence occupies one continuous linear segment of DNA. However, in eukaryotic genes the coding sequences are frequently “split” in the genome—a discovery reached independently in the 1970s by Richard J. Roberts (the author of this article) and Phillip A. Sharp, whose work won them a Nobel Prize in 1993. The segments of DNA or RNA coding for protein are called exons, and the noncoding regions separating the exons are called introns. Following transcription, these coding sequences must be joined together before the mRNAs can function. The process of removal of the introns and subsequent rejoining of the exons is called RNA splicing. Each intron is removed in a separate series of reactions by a complicated piece of enzymatic machinery called a spliceosome. This machinery consists of a number of small nuclear ribonucleoprotein particles (snRNPs) that contain small nuclear RNAs (snRNAs).

RNA editing

Some RNA molecules, particularly those in protozoan mitochondria, undergo extensive editing following their initial synthesis. During this editing process, residues are added or deleted by a posttranscriptional mechanism under the influence of guide RNAs. In some cases as much as 40 percent of the final RNA molecule may be derived by this editing process, rather than being coded directly in the genome. Some examples of editing have also been found in mRNA molecules, but these appear much more limited in scope.

Practical Plant Biochemistry

1- Determination of Pigments

2- Determination of sugars

3- Determination of protein

4- Determination of amino acids

5- Determination of terpenoids

6- Determination of anthocyanin

7- Determination of lipids

8- Determination of alkaloids

9- Determination of phenols

10- Determination of DNA and RNA