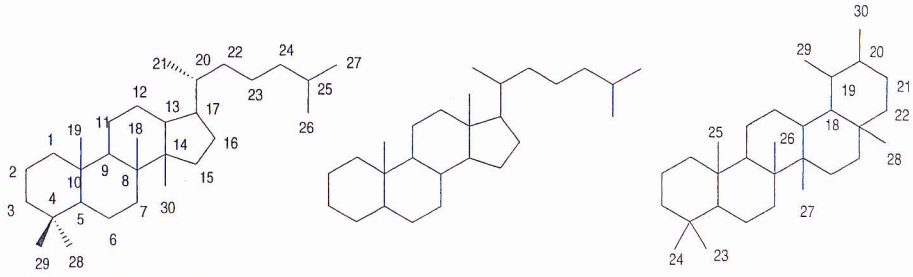
Triterpenes and Steroids

Generalities

Triterpenes—4,000 compounds built upon over 40 different skeletons—are C_{30} compounds arising from the cyclization of epoxy-3*S*-2,3-epoxy-2,3-dihydrosqualene, or, in rare cases, from squalene itself. Almost always hydroxylated at C-3 (because they arise from the opening of the epoxide), triterpenes present very high structural homogeneity: the major differences are in configuration, and are linked to the conformation adopted by the squalene epoxide (or by squalene) prior to cyclization; the cation resulting from this cyclization can subsequently undergo a series of 1,2 proton and methyl group shifts, which can be used to rationalize the occurrence of the different tetra- and pentacyclic skeleta characteristic of this group.

Structural homogeneity is also marked among the steroids: compounds as different, by their properties, as phytosterols, saponins, ecdysteroids, cardiac glycosides, or steroidal alkamines, all have the same basic skeleton.



*Tetracyclic triterpene** *Steroid** *Pentacyclic triterpene*

* extra carbons at the 24-position are numbered 24¹ et 24²

At first glance, it is fair to say that there are no fundamental differences between triterpenes and steroids, as these may be considered to be tetracyclic triterpenes having lost, at a minimum, three methyl groups (in fact it is the presence of the methyl groups at C-4 and C-14 which was initially used to distinguish steroids from triterpenes [Ourisson]). Only by considering the biosynthesis is it possible to separate the two groups: a compound like cycloartenol (C_{30}) must be considered a 4,4-dimethylsterol (it is a sterol precursor), whereas euphol or the dammaranes (also with 30 carbon atoms) are tetracyclic triterpenes. Separating the two groups is not always easy: for example, where do the cucurbitacins belong? With sterols—which are derived from protostane—or, according to many authors, in the group of tetracyclic triterpenes? A 1991 publication covers the lanostanes in the chapter on triterpenoids and lanosterol in the chapter on phytosterols.

APPLICATIONS OF TRITERPENES AND STEROIDS

In view of their therapeutic and industrial applications, triterpenes and steroids constitute a group of secondary metabolites of the utmost importance.

- Consider the applications of cardiac glycosides, which no synthetic product has completely replaced.

- Consider the applications of spirostane-type saponins, of sitosterol, and of stigmasterol, which are valuable starting materials for many approaches in biotechnology. They remain irreplaceable to fulfill the needs of the pharmaceutical industry for steroidal medications (contraceptive, anabolic, anti-inflammatory agents).

- Consider the therapeutic applications of many saponin-containing drugs, used for the extraction of active compounds (aescin, glycyrrhizin), to obtain simple galenicals, or to prepare phytotherapeutic products.

- Consider the economic importance of licorice, a low-calorie sweetener, widely used in food technology.

- Consider the importance of saponins, in that their presence can substantially decrease the nutritional value of fodder (alfalfa), or impart to plants that are familiar in our every day environment a non-trivial toxicity.

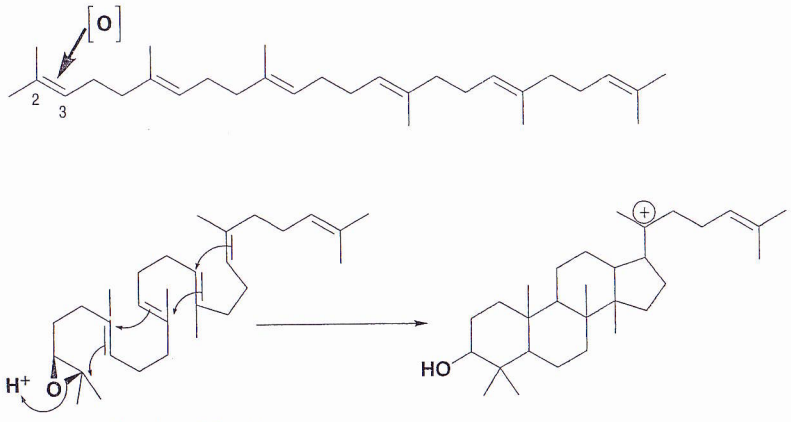
- Consider their therapeutic potential in many varied fields: as cytostatics, insecticides, anti-inflammatory agents, products toxic to molluscs, analgesics, and more.

BIOSYNTHESIS OF TRITERPENES AND STEROIDS

Although steroids in animals, fungi, algae, and higher plants all arise from a common pathway which leads, from acetate, *via* mevalonate, to squalene epoxide, upon closer examination it appears that from that point, the biosynthetic pathways diverge considerably. The first sterol synthesized by animals and fungi is lanosterol; which is converted to cholesterol in most animals, and to ergosterol in fungi. In the case of the Eucaryotes capable of photosynthesis (Algae, Bryophytes, Pteridophytes, Spermatophytes), all of the sterols (phytosterols, cardenolides, spirostanes, solanidanes) arise from the stepwise demethylation of cycloartenol and from the opening of its 9β,19-cyclopropane ring. In addition, these vegetable organisms are capable of cyclizing squalene epoxide in a conformation that leads specifically to the tetracyclic triterpenes of the laticiferous ducts of the Euphorbiaceae, as well as to the saponins with a pentacyclic triterpenoid aglycone, or to the modified triterpenes of the Rutales (quassinoids, meliacins, limonoids).

Initial cyclization

It is the opening of the epoxide which initiates the cyclization. In order for this cyclization to take place, the cyclization enzyme must stabilize the conformation of the polyisoprene in a manner that fulfills the stereoelectronic requirements of the cyclization. It is the initial conformation of the squalene epoxide which determines the orientation of the biosynthesis toward either steroids and cucurbitacins, or else triterpenes in the strict sense of the term.



Squalene-2,3-oxide

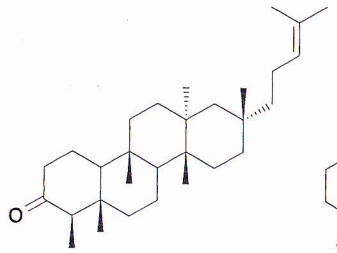
1. If the squalene epoxide is maintained in a chair-boat-chair conformation, the cyclization leads to a protostane cation, which is the immediate precursor of cycloartanes and cucurbitanes, by a series of 1,2-proton and methyl group shifts (these shifts are made possible by the *trans*-antiparallel arrangement of the protons and methyl groups at C-17, C-13, C-14, and C-8).

2. If the squalene epoxide is maintained in a chair-chair-chair conformation, the cyclization leads to a dammarane cation (see, for example, the aglycones of the saponins of ginseng), which can also undergo rearrangement:

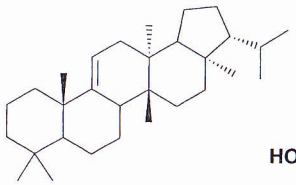
- either by concerted migrations leading to tirucallol and euphol, the precursors of limonoids and quassinoids,
- or, in most cases, by the formation of an extra ring, leading to pentacyclic triterpenes (oleananes, ursanes, lupanes, friedelanes, taraxastanes),
- or, in rare cases, to form tetracyclic compounds with a six-membered D ring (baccharanes, shionanes).

3. A somewhat special case is that of triterpenes devoid of a hydroxyl group at C-3. They are generally derived from the direct cyclization of squalene: hopanes (characteristic of natural sediments), fernanes (the conformation of the precursor is of the chair-chair-chair-boat type).

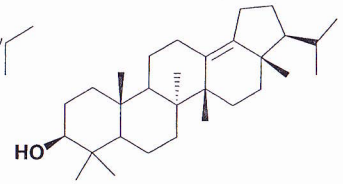
4. In some cases, the cyclization is only partial (polypodatetraenes, malabaricanes), or on the contrary it is complete, and completely concerted (boehmerol, arborinol: chair-boat-chair-chair-boat), or even initiated from both ends of the precursor (onoceranes). Some unusual structures have also been described (C_{31} aldehydes from *Iris*), especially in the animal kingdom (siphonales from the Spongiae).



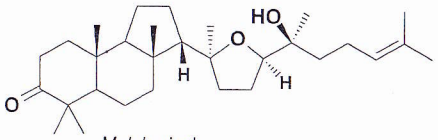
Shionone



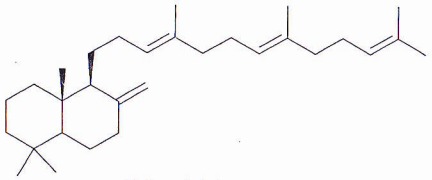
$\Delta^{9(11)}$-Fernene



Boehmerol



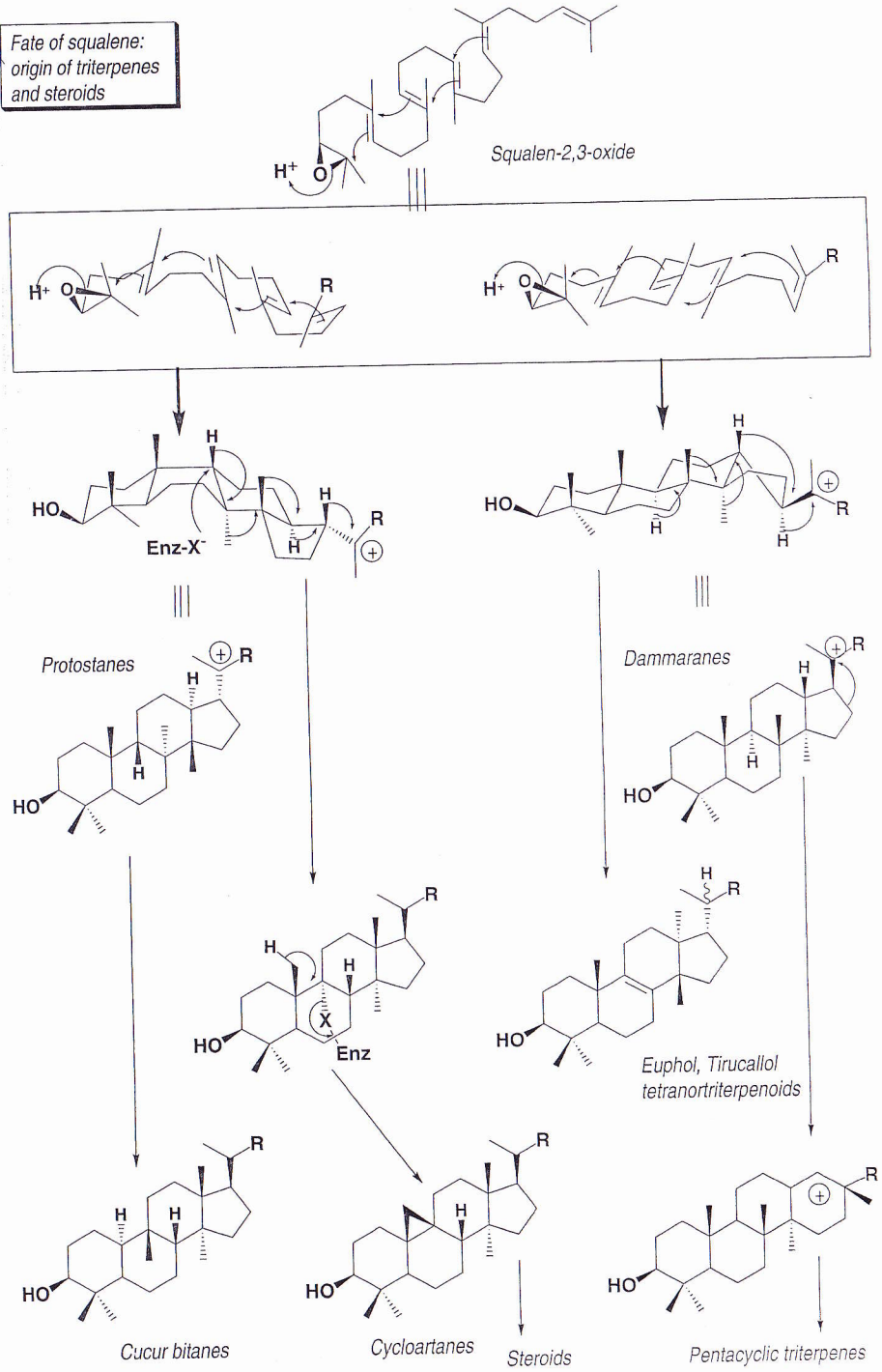
Malabaricol



α-Polypodatetraene

Triterpenes resulting from an alternate mode of cyclisation

Fate of squalene: origin of triterpenes and steroids

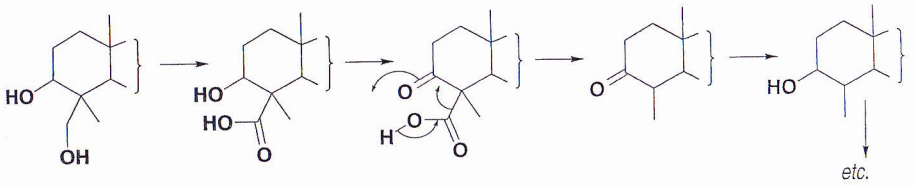


CITRULLUS COLOCYNTHIS Arn.

Formation of the steroids

As described above, animals and fungi elaborate lanosterol—the rearrangement ends with the release of the proton at C-9—whereas plants make cycloartenol: the rearrangement ends with the formation of cyclopropane by a reaction which is probably catalyzed by an enzyme.

The conversion of a C_{30} skeleton to a C_{27} or smaller skeleton, in other words to a steroid, involves—at a minimum—a stepwise demethylation at C-4 and C-14; note also the opening of the cyclopropane ring and a migration of the double bond resulting from this opening. Both methyl groups at C-4 are lost through a series of oxidations (CH_3 —> CH_2OH —> CHO —> CO_2H) which ends with a decarboxylation. A preliminary oxidation of the hydroxyl group at C-3 leads to an α-ketoacid, which facilitates the final decarboxylation. The methyl group at C-14 is eliminated by oxidation to formic acid.

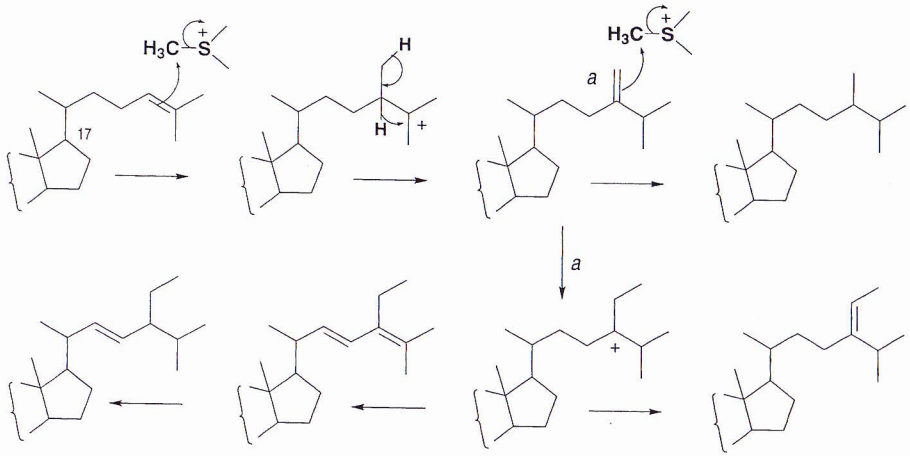


Principle of the elimination of the methyl group at the 4-position

In animals, the side chain remains intact (cholestane), becomes truncated (C_{24} cholanes, C_{21} pregnanes), or even eliminated (C_{19} androstanes, C_{18} estranes), but in plants, the side chain may be functionalized and cyclized (spiroketals, alkamines, ecdysteroids), shortened (pregnanes) and functionalized (cardenolides, conanines), or—this is frequent—it may possess one or two extra carbons in the form of a methyl (or methylene) group, or an ethyl (or ethylidene) group at C-24. This characteristic of C_{28} or C_{29} phytosterols from higher plants is also found in the algae (fucosterol), fungi (ergosterol), and marine organisms.

The introduction of extra carbon(s) into the side chain of the steroids results from transmethylations involving *S*-adenosylmethionine. The first transmethylation generally precedes the demethylations at C-4 and C-14, and the second one commonly takes place at a later time. Sterols with a 24-methylene or a 24-ethylidene group can isomerize to the corresponding 24-methyl- and 24-ethyl Δ^{24} sterols, which can yield, by a stereospecific reduction, the 24α-alkylsterols characteristic of plants, for example, sitosterol (ethyl group) or campesterol (methyl group).

The other possible fates of the steroid nucleus appear in the figure on p. 670, and will be summarized below.



Examples of modifications of the C-17 side chain of phytosterols

Formation of triterpenes

The guiding principles which result in the elaboration of the chief triterpenoid skeletons appear in the figure on p.535. Although a fair number of the proposed sequences remain hypothetical, they are, however, supported by the fact that some of the proposed reactions can be achieved *in vitro* in acidic medium, under conditions that mimic biosynthesis. Furthermore, a certain number of experiments with labeled compounds—among others, with ^{13}C double-labeled acetate—have been conducted: they demonstrate the validity of several of the proposed mechanisms.

The secondary modifications of triterpenes are rather limited: additional hydroxylations, dehydrogenations, functionalizations of the angular methyl groups, and lactonizations are the most common (see below: saponins). One exception lies in several Rutales families (Rutaceae, Meliaceae, Simaroubaceae, Cneoraceae) in which the initial tetracyclic skeleton can undergo profound modifications: oxidation, ring opening and closure, side chain elimination, and more (see p. 764 and 766).

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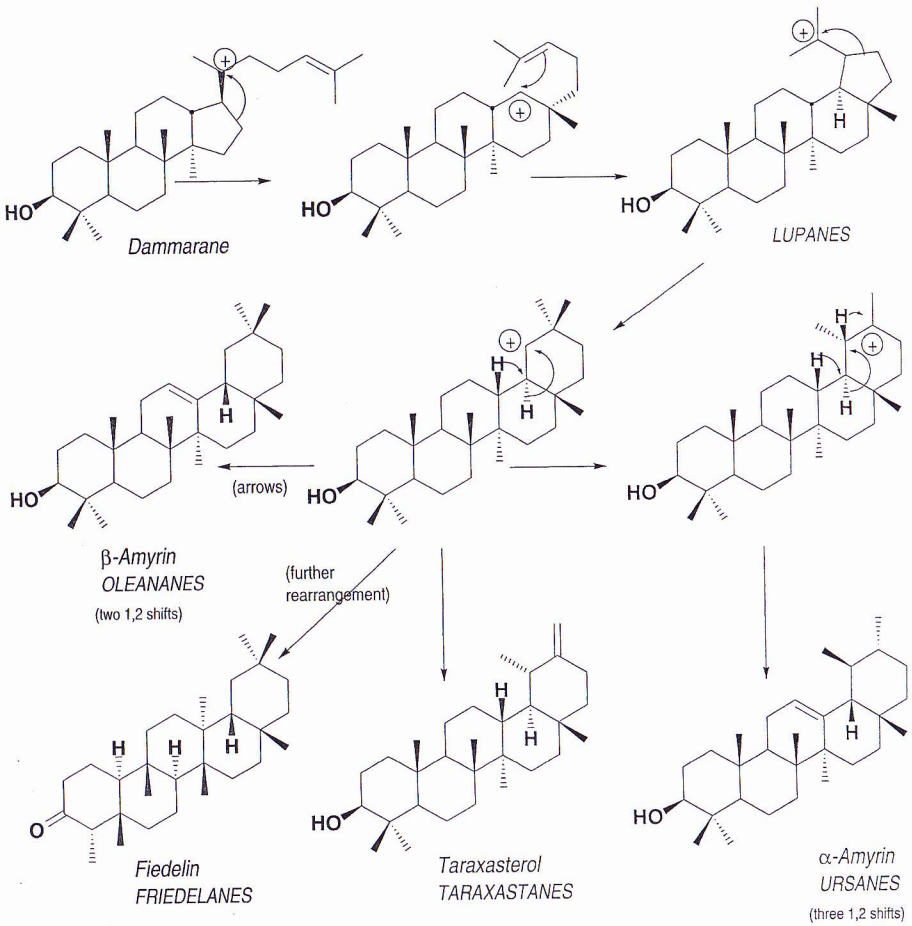
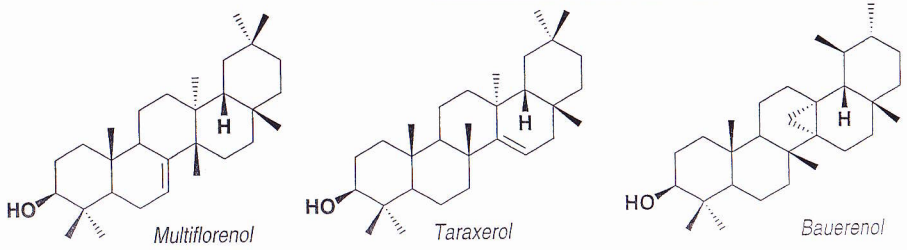
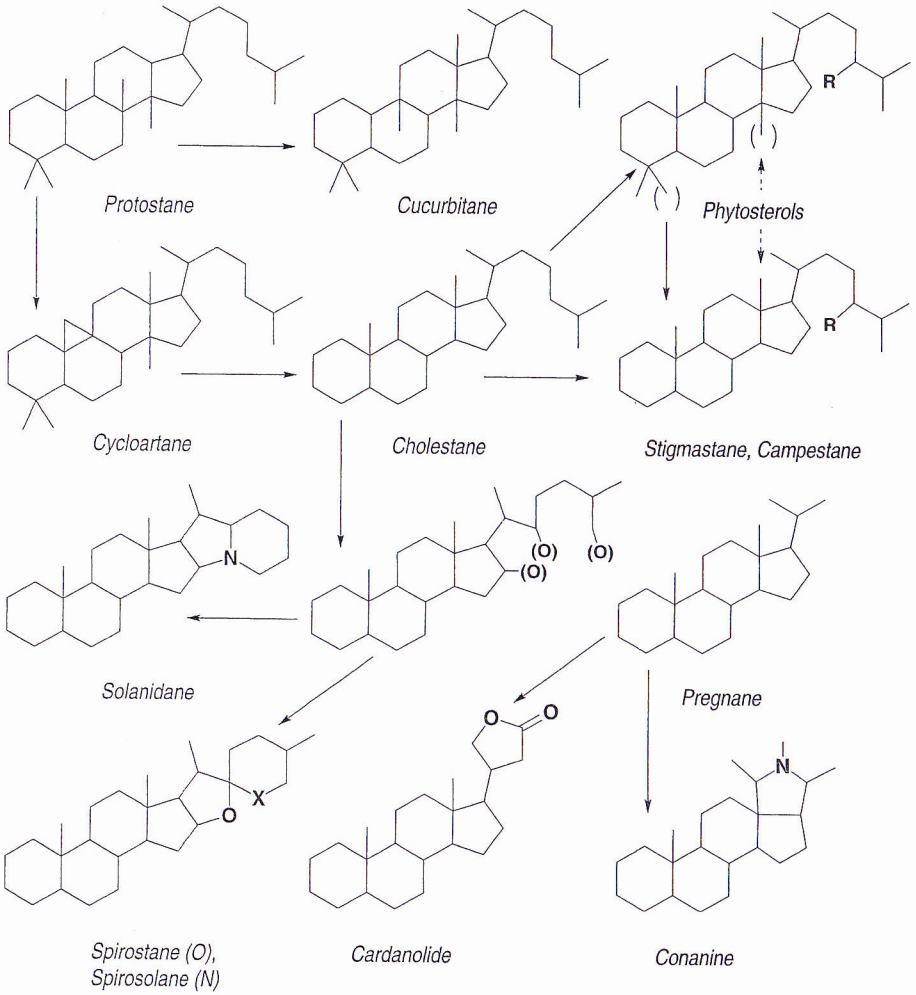


Interconversions in the pentacyclic triterpene series (examples)

Other examples of pentacyclic triterpenes





Chief basic steroidal skeleton found in higher plants

Triterpenes and Steroids

Saponins