A COMPARISON OF TRITERPENOIDS WITH STEROIDS AS MEMBRANE COMPONENTS

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Steroids have, aside from their role as emulsifying agents (bile acids and alcohols), two principal functions in living systems: they may act as hormones (1–3) or as architectural components of membranes (4–6). Because pentacyclic triterpenoids have also been shown to play a role in membranes (5, 7), it has been suggested that the sequence in which squalene, pentacyclic triterpenoids, and sterols appear in biosynthesis and during the evolutionary process is related to their membranous functions (7–9).

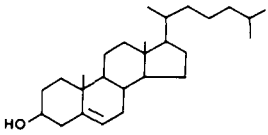
This article summarizes some of the literature that was not covered by earlier reviews on the role of steroids in membranes (4–6). Our attention has been focused on plant systems and we do not claim completeness of coverage. Extending the discussion to the triterpenoids, we will seek answers from the literature to some of the following questions: what are the molecular features of triterpenoids and sterols which are required for growth in organisms that are auxotrophic or heterotrophic for sterols; are there any differences in membrane composition among plants throughout the evolutionary hierarchy or between plants and animals; to what extent do physico-chemical studies of sterol-lecithin interactions reflect the architectural fit of sterols and other polycyclic isopentenoids into natural biological membranes; do tracheophytes prefer 24-alkylsterols over 24-desalkylsterols, e.g., cholesterol and triterpenoids, in their membranes?

The structures and trivial names of the principal sterols and triterpenoids associated with subcellular fractions are shown in figs. 1 and 2, respectively. Their biogenetic relations are outlined in fig. 3. Cyclization of the acyclic triterpene 2,3-oxidosqualene leads via the protosteroid cation to lanosterol or cycloartenol (10, 11). The tetracyclic or pentacyclic triterpenoids are produced either by direct cyclization of squalene or via the triterpenoid cation. The direct cyclization process is anaerobic, and only pentacyclic triterpenoid products are formed (8, 12). It should be noted that, although steroids and certain triterpenoids have a tetracyclic ring system in common, the rings of steroids are stereochemically different from triterpenoids. The difference results from the mechanism by which 2,3-oxidosqualene passes through one of two possible transitory cation intermediates. Although lanosterol and cycloartenol are steroids on biosynthetic grounds (4), we will classify all C–30 polycyclic isopentenoids as triterpenoids.

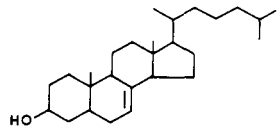
EVIDENCE FOR THE ROLE OF STERIODS AND TRITERPENOIDS IN MEMBRANES

Organismic Occurrence

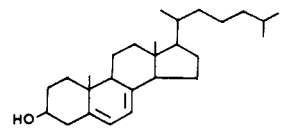
As is evident from tables 1, 2, and 3, sterols are found throughout the evolutionary hierarchy, from the cyanophytic photosynthetic bacteria to tracheophytes, as well as from nonphotosynthetic bacteria to man. Triterpenoids (excluding lanosterol and cycloartenol) appear to be confined to the photosynthetic and lower nonphotosynthetic organisms. The first three tables also show that 24-



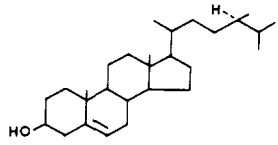
CHOLESTEROL



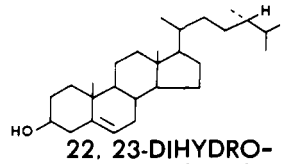
LATHOSTEROL



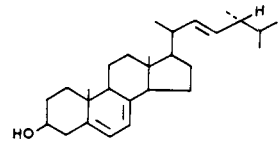
7-DEHYDROCHOLESTEROL



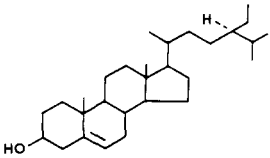
CAMPESTEROL



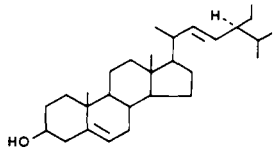
22, 23-DIHYDRO-BRASSICASTEROL



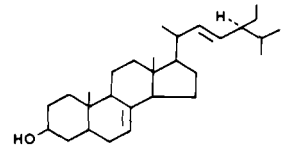
ERGOSTEROL



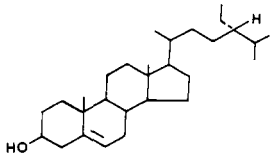
SITOSTEROL



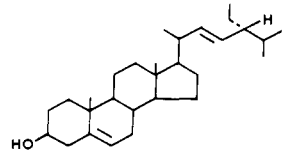
STIGMASTEROL



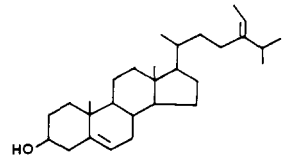
SPINASTEROL



CLIONASTEROL

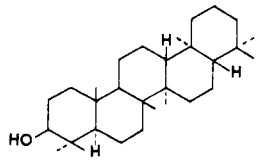


PORIFERASTEROL

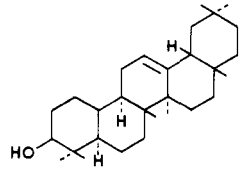


FUCOSTEROL

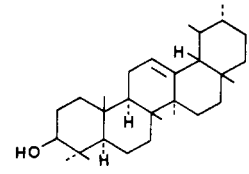
FIG. 1. Principal membrane sterols.



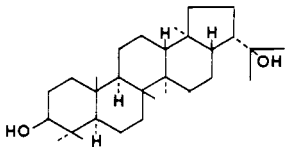
TETRAHYMANOL



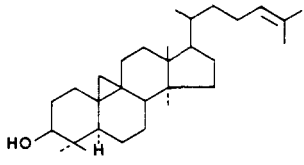
β-AMYRIN



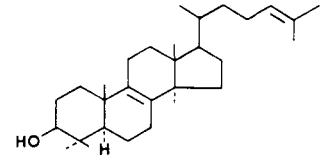
α-AMYRIN



DIPLOPTEROL



CYCLOARTENOL



LANOSTEROL

FIG. 2. Principal membrane triterpenoids.

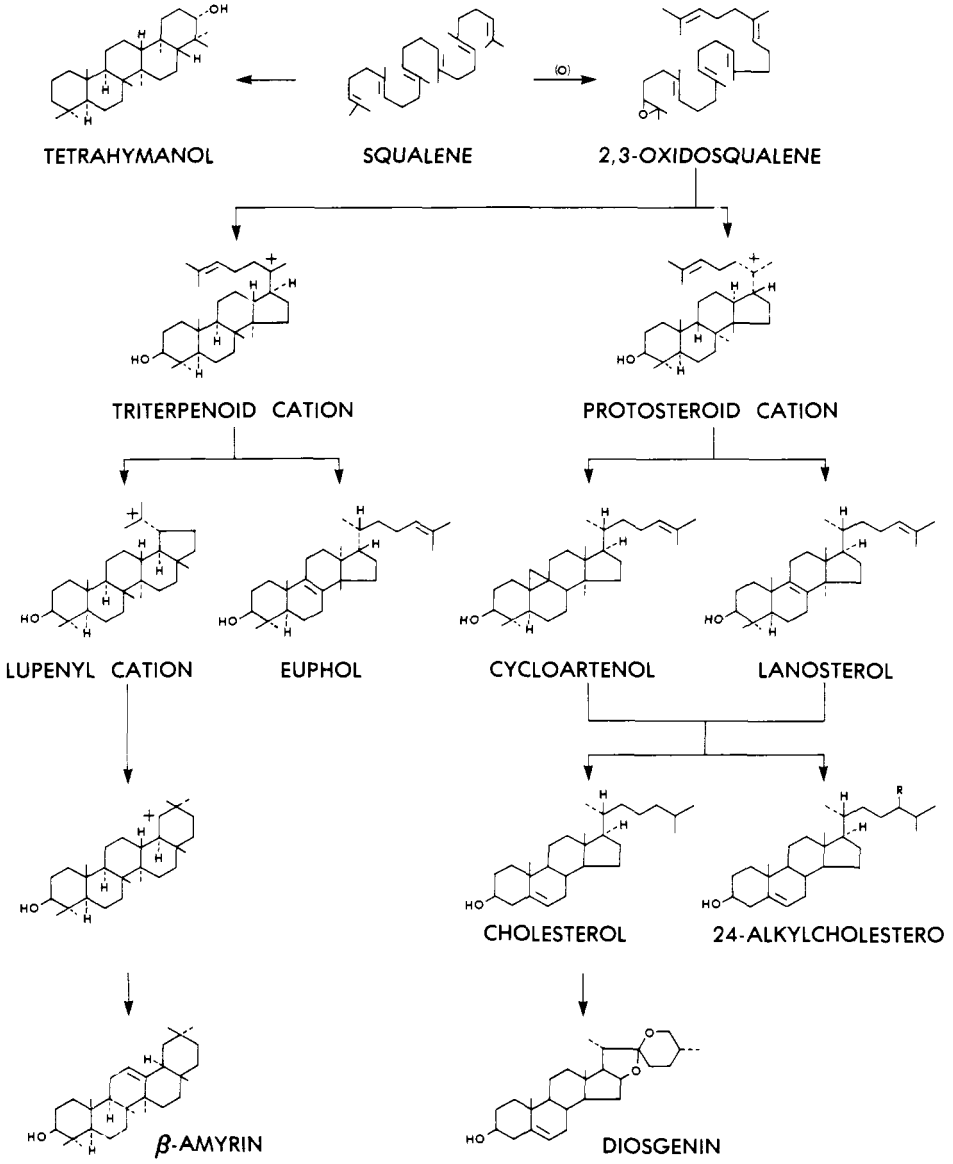


FIG. 3. Biosynthetic pathways of sterols and triterpenoids.

alkylsterols are found throughout the photosynthetic kingdom, while their production by nonphotosynthetic organisms becomes increasingly rare as one ascends the evolutionary ladder. The total sterol concentration in eukaryotic organisms is ca. 0.1–1.0% of their wet weight (5). In bacteria the sterol concentration is usually within one to two orders of magnitude lower than that reported for eukaryotes. The concentration range of triterpenoids in bacteria is approximately the same as that of sterols (7), and the concentration of triterpenoids in tracheophyte seeds (25) and leaves (26) is of the same order of magnitude as that of sterols isolated from the same sources.

TABLE 1. Prokaryotic organisms containing sterols or triterpenoids.

Sterols	Refs.	Pentacyclic triterpenoids	Refs.
Nonphotosynthetic bacteria		*Nonphotosynthetic bacteria*	7,8
Methylobacterium organophilum	13	*Methylococcus sp.*	7,8
Methylococcus capsulatus	14,15	*Methylomonas sp.*	7,8
Cellulomonas dehydrogenans	16	*Methylocystis sp.*	7,8
Azotobacter chroococcum	17	*Methylosinus sp.*	7,8
Streptomyces olivaceus	17	*Hyphomicrobium sp.*	7,8
		Nitrosomonas europoea	7,8
Cyanobacteria		*Pseudomonas cepacia*	7,8
Anacystis nidulans	18	*Azotobacter vinelandii*	7,8
Nostoc commune	19	*Bacillus acidocaldarius*	7,8
Spirulina platenis	19	*Streptomyces chartreusi*	7,8
Phormidium luridium	20		
Anabena cylindrica	21	*Photosynthetic bacteria and Cyanobacteria*	
Calothrix sp.	19	*Anabaena sp.*	7,8
Tremyella diplosiphon	18	*Nostoc sp.*	7,8
Spirulina maxima	22	*Synechocystis sp.*	7,8
		Rhodopseudomonas sp.	7,8
		Rhodospirillun sp.	7,8
		Rhodomicrobium sp.	7,8

TABLE 2. Synthesis of sterols and triterpenoids in the eukaryote kingdom, Photosynthetica.

Organismic group	Sterols		Triterpenoids	
	Synthesis[a]	Alkylation	Tetracyclic[b]	Pentacyclic
I. Algae				
Division Euglenophyta	+	+	−	−
Division Chlorophyta	+	+	−	−
Division Chrysophyta	+	+	−	−
Division Pyrrophyta	+	+	−	−
Division Phaeophyta	+	+	−	−[b]
Division Rhodophyta	+	−	−	−
II. Less Advanced Plants				
Division Bryophyta	+	+	−	−
III. More Advanced Plants				
Division Tracheophyta				
Subdivision Psilopsida	+	+	?	−
Subdivision Lycopsida	+	+	?	+
Subdivision Sphenopsida	+	+	?	+
Subdivision Pteropsida	+	+	+	+
Subdivision Spermopsida	+	+	+	+

[a]Including lanosterol and cycloartenol.
[b]Except for certain species (4,23,24).

TABLE 3. Synthesis of sterols and triterpenoids in the eukaryote kingdom, Nonphotosynthetica.

Organismic group	Sterols		Triterpenoids	
	Synthesis[a]	Alkylation	Tetracyclic	Pentacyclic
I. Fungi				
Division Myxomycophyta	−	+	−	−
Division Eumycophyta	+[c]	+	−	−[c]
II. Primitive Animals				
Subkingdom Protozoa				
Phylum Protozoa	−	−[b]	−	−[c]
Subkingdom Metazoa				
Phylum Coelenterata	−	?	−	−
Phylum Nemathelminthes	−	−	−	−
Phylum Platyhelminthes	−	?	−	−
Phylum Aschelminthes	−	?	−	−
Subkingdom Parazoa				
Phylum Porifera	+[c]	?	−	−
III. Invertebrate Animals				
Phylum Mollusca	+	+	−	−
Phylum Annelida	+[c]	−	−	−
Phylum Arthropoda	+[c]	−[b]	−	−
Phylum Echinodermata	+	−	−	−
IV. Vertebrate Animals				
Phylum Chordata	+	−[c]	−	−

[a]Including lanosterol and cycloartenol.
[b]Some genera may dealkylate (4).
[c]Except for certain species (4, 23).

Subcellular Distribution

24-Desalkyl- and 24-alkylsterols, as the free alcohols, have been shown to occur in subcellular fractions of tracheophytes with 24-alkylsterols predominating (table 4). While 24-desalkylsterols, e.g., cholesterol, are the major sterols occurring in all subcellular fractions of mammals (4, 35), it has recently been found that 24-alkylsterols may also accumulate in mammalian membranes, depending on the amount of "phytosterols" in the diet (table 5). Mammals are known to synthesize free sterols, steryl esters, and steryl sulfates, but usually sterols occur only as the free alcohols in their various membrane systems to any significant extent (38). One exception to this rule appears to be cholesteryl sulfate, which accumulates in red blood cell membranes (39).

Plants, unlike mammals, synthesize free sterols, steryl esters, steryl glucosides, and acyl steryl glucosides (40–44), all of which occur in subcellular fractions (cf. refs. in table 4). The major sterol fraction occurring in plant membranes is not always in the free form. Steryl esters may predominate in particular membranes. For instance, much of the total cholesterol in certain tracheophytic leaves occurs in the form of esters in the nucleus, mitochondria, or chloroplast (27, 29, 45). Moreover, there are reports that certain molecular species of the 24-alkylsterols, conjugated with sugars, may also accumulate in membranes, depending on age or the degree of light received by the plants (46, 47).

Mammals are apparently incapable of synthesizing steroidal sapogenins, pentacyclic triterpenoids, and tetracyclic triterpenoids formed via the triterpenoid cation (cf. fig. 3). These compounds have never been found in mammalian membranes, although they are known to be present, together with the

TABLE 4. Subcellular distribution of free 4-desmethylsterols in tracheophytes as percent of total sterols.

	Nucleus		Mitochondria					Microsomes					Chloroplasts						Plasmalemma		
Plant Organ	*Zea mays* Shoots	*Nicotiana tabacum* Leaves	*Phaseolus vulgaris* Leaves	*Nicotiana tabacum* Leaves	*Zea mays* Shoots	*Zea mays* Coleoptiles	*Solanum tuberosum* Tuber	*Phaseolus vulgaris* Leaves	*Nicotiana tabacum* Leaves	*Zea mays* Shoots	*Zea mays* Coleoptiles	*Solanum tuberosum* Tuber	*Phaseolus vulgaris* Leaves	*Nicotiana tabacum* Leaves	*Zea mays* Shoots	*Zea mays* Coleoptiles	*Antirrhinum majus* Leaves	*Spinacia oleracea* Leaves	*Glycine max* Roots	*Zea mays* Coleoptiles	*Phaseolus vulgaris* Cotyledons
Sterol[a]																					
Cholesterol	22	11	27	9	1	2	51	6	10	1	2	18	24	12	2	2	—	3	tr.	1	1
Lathosterol	—	—	3	—	—	—	—	2	—	—	—	—	1	—	—	—	—	—	—	—	—
Campesterol	9	15	5	14	12	18	—	8	15	12	19	—	5	16	13	19	—	—	11	19	5
Stigmasterol	18	25	17	25	26	56	4	23	23	27	53	6	22	24	26	56	—	—	31	56	41
Sitosterol	51	49	37	52	61	24	45	55	52	60	26	76	48	48	59	23	89	—	58	24	41
Isofucosterol	—	—	11	—	—	—	—	6	—	—	—	—	tr.	—	—	—	—	—	—	—	12
Unknown	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	11	3	—	—	—
7-Stigmasterol	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	48	—	—	—
Spinasterol	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	46	—	—	—
Ref.	27	28	29	28	27	30	31	29	28	27	30	31	29	28	27	30	32	32	33	30	34

[a]Dash (—) indicates absence.

"phytosterols", in significant amounts in the diet of herbivores and omnivores. With modern analytical methods, sapogenins and triterpenoids should have been detected. On the other hand, the so-called phytosterols, i.e., 24-alkylsterols, have been identified in mammalian tissues such as adrenal glands (48), aorta (49), skin (50), erythrocytes (51), brain (52), and in cancerous tissues (53–56). In many cases, 24-methylsterols occur in mammals in higher concentrations than do 24-ethylsterols (57, 58). The absence of steroidal sapogenins and triterpenoids from mammalian membranes indicates that the discriminatory mechanisms of the intestinal mucosa are very efficient.

Alternatively, plants that synthesize steroidal sapogenins and triterpenoids incorporate them into their membranes (tables 6 and 7). As is shown in table 4,

TABLE 5. Distribution of dietary plant sterols in mammalian subcellular fractions. Ratio of cholesterol: campesterol: sitosterol.

	Nuclei	Mitochondria	Microsomes	Plasmalemma	Refs.
Liver of rats fed low-phytosterol diet	93:2:5	91:3:6	92:2:6	N.E.[a]	36
Rats fed high-phytosterol diet	88:7:5	86:10:4	84:9:7	N.E.	36
L-cell fibroblast, incubated with sitosterol	3:0:1	2:0:1	2:0:1	2:0:1	37

[a]Not examined.

sterols are found in the nucleus, but triterpenoids, which were sought after in the same plant, are apparently excluded from the nucleus (cf. table 7). That plants select certain sterols in specific molecular forms, e.g., cholesteryl esters (27, 29, 45), for the nucleus while excluding monohydroxy triterpenoids indicates that plant membranes are capable of discriminating among structurally similar polycyclic compounds. Not only do plants select particular compounds for deposition in the lipid leaflet of subcellular fractions, but plants as well as animals selectively deposit sterols on either the outer or inner surface of the cell membranes (table 8). This asymmetric distribution of sterols and other lipids in membranes has been referred to as "lipid asymmetry" (71).

TABLE 6. Subcellular distribution of free steroidal sapogenins in *Dioscorea* sp. in percent of total sapogenins.

	Nuclei			Mitochondria			Microsomes			Chloroplasts	
	D. tokoro	*D. tokoro*	*D. bernoulliana*	*D. tokoro*	*D. tokoro*	*D. bernoulliana*	*D. tokoro*	*D. tokoro*	*D. bernoulliana*	*D. tokoro*	*D. tokoro*
	Leaves	Rhizomes	Tubers	Leaves	Rhizomes	Tubers	Leaves	Rhizomes	Tubers	Leaves	Rhizomes
Refs.	59	59	60	59	59	60	59	59	60	59	59
Sapogenin											
Diosgenin	–[a]	–	4	–	–	13	–	–	52	–	–
Yonogenin	96	50	6	75	50	–	15	50	16	96	50
Tokorogenin	4	50	–	25	50	14	25	50	7	4	50
Kryptogenin	–	–	39	–	–	19	–	–	–	–	–
Hecogenin	–	–	30	–	–	20	–	–	8	–	–
Tigogenin	–	–	7	–	–	–	–	–	11	–	–
Yamogenin	–	–	14	–	–	18	–	–	–	–	–
Smilagenin	–	–	–	–	–	16	–	–	6	–	–

[a]Dash (–) indicates absence.

It is generally believed that sterols comprise 2–25% of the lipid leaflet in nonphotosynthetic organisms (4). However, depending on the basis by which the sterol content is calculated (e.g., as percent of the dry weight, as percent of the wet weight, as percent of the proteins, or as percent of the phosphorus or phospholipids), the organelles isolated from plants may contain significantly more or less sterol than analogous membranes from nonphotosynthetic organisms (table 9).

Not all photosynthetic plants contain phospholipids as their main polar lipids in membranes, e.g., chloroplasts (86, 87). Therefore, plant biochemists rarely report the sterol content as percent of the phospholipids, and such data have been excluded from table 9. The sterol to phospholipid ratio is high in mammalian plasma membranes (67, 78) as it is in plants (30, 34, 74). In plant membranes,

TABLE 7. Subcellular distribution of free triterpenoids in eukaryotes.

	Nuclei		Mitochondria			Microsomes			Choroplasts				Cilia	
	Tetrahymena pyriformis	*Calendula officinalis*	*Tetrahymena pyriformis*	*Calendula officinalis*	*Zea mays*	*Tetrahymena pyriformis*	*Calendula officinalis*	*Zea mays*	*Calendula officinalis*	*Zea mays*	*Lycopersicon esculentum*	*Sinapis alba*	*Nicotiana tabacum*	*Tetrahymena pyriformis*
Refs.	61	62	61,63	62	27	61	62	27	62	27	64	64	64	63
Triterpenoid[a]														
Tetrahymanol	−	−	+	−	−	+	−	−	−	−	−	−	−	+
β-Amyrin	−	−	−	+	+	−	+	+	+	+	−	−	−	−
α-Amyrin	−	−	−	−	+	−	−	+	−	+	−	−	−	−
Erythrodiol	−	−	−	−	−	−	+	−	+	−	−	−	−	−
Cycloartenol	−	−	−	−	+	−	−	+	−	+	+	+	+	−
24-Methylene-cycloartanol	−	−	−	−	+	−	−	+	−	+	−	−	−	−

[a]Dash line (−) indicates not present; plus sign (+) indicates present. Only leaves of the plants were examined; the whole organism in the case of *T. pyriformis*.

however, the sterol to phospholipid ratio is significantly altered during senescence (34), indicating that developmental processes influence the sterol composition of some plants.

Except for the Golgi apparatus and chloroplastic membranes, plant membranes, as a general rule, contain sterols in amounts of the same order of magnitude as do their counterparts in the nonphotosynthetic organisms when the amounts are calculated on the basis of percent of the proteins. However, when the amounts are calculated on the basis of percent of the wet weight, photosynthetic plants

TABLE 8. Asymmetric distribution of sterols in membranes.

Sterol	Organism	Membrane system	Ratio of amount, outer: inner membrane	Refs.
Ergosterol	Fungus (*Neurospora crassa*)	mitochondria	100:0	65
Cholesterol	mammal (guinea pig liver)	mitochondria	6:1	66
	rat liver	mitochondria	100:1	67
Cholesterol	bacterium (*Mycobacterium capricolum*)	cytoplasmic envelope	2:1	68
Cholesterol	mammal	myelin	2:1	69
Sitosterol	tracheophyte (cauliflower bud)	mitochondria	3:1	70

may contain significantly less membranous sterol. Triterpenoid concentrations are usually reported as percent of the wet weight, and when they occur together with sterols in membranes, their contribution to the mixture is ca. 10%. In protozoa, triterpenoids occur in the same range of concentrations as do sterols in the membranes (88).

Generally, the various membranes within a cell contain different amounts of sterols and triterpenoids. For instance, in photosynthetic plants, chloroplastic and mitochondrial membranes usually contain less of the polycyclic isopentenoids than do the endoplasmic reticulum and plasmalemma. This indicates that there may be a relationship between membrane composition (structure as well as concentration) and membrane function.

Biological Studies

The occurrence of sterols in subcellular organelles throughout the evolutionary hierarchy is good reason to believe that they act as architectural components of membranes. An increase in the amount of free sterols has been observed to accompany biogenesis of membranes in plants as well as in animals (4, 33). This has led various investigators to suggest that the free sterol content of cells is a measure of the membranous sterols (4, 89, 90). Additional observations support the association of sterol accretion with membrane development. Thus, studies concerned with sterol metabolism in animals (4) have shown that cholesterol or related sterols undergo a slow turnover. This points toward a nonmetabolic role for sterols, which implies that the sterols function as an architectural component. Investigations of the growth and morphological characteristics—generally taken to be a measure of membrane response—of mycoplasmas and other prokaryotes (91–96), pythiaceous fungi (90, 97–101), anaerobic yeast (102–104), and protozoa (105–107) have demonstrated that dietary sterols are incorporated into their membranes (108–111). This results in altered membrane permeability (111–114) and growth stimulation or inhibition, depending on the structure of the added sterols. Mutations of the sterol pathway in yeast (115–118) and mammalian tissue culture cells which prevent the formation of the end-product, e.g., ergosterol in yeast and cholesterol in mammals, lead to alterations in growth and membrane permeability (119) and may even lead to cell death (120), depending on the intermediate that accumulates.

In organisms that are auxotrophic or heterotrophic for sterols, the addition of sterol-complexing agents (SCA), e.g., polyene antibiotics (121), steroidal (122) or triterpenoid saponins (123), or glycoalkaloids (122), to the growth medium usually has no significant effect on growth when their concentrations are low. However, the simultaneous addition of sterols and SCA inhibits growth (124). Addition of SCA to cells normally synthesizing sterols also results in growth inhibition (125). Presumably, complexing of sterols with SCA occurs in the membrane (126). This assumption is supported by the fact that sterols complex with SCA in the bilayers of model systems, e.g., sterols combine with digitonin (127) and various polyene antibiotics (121). Also, alterations in the function of membranes are observed *in vitro* when SCA are added to isolated chloroplasts of peas (128), mitochondria of beans (129), and protoplasts of petunia and tobacco (130). A further observation associating specifically free sterols with biogenesis of membranes is that aerobic cultures of yeast synthesize primarily free sterols during their early growth phase, but, as the stationary phase approaches, steryl esters are formed (131, 132) and are deposited along with triglycerides in lipid

droplets (108). Also, during germination of seeds (133) and development of seedlings (134), and in suspension cultures of higher plants (135) an increase in free sterols is observed. While free desmethylsterols undoubtedly accompany biogenesis of membranes in a range of nonphotosynthetic and photosynthetic organisms, other sterol forms also accompany membrane development, and this apparently depends on the organ studied. For instance, depending on the organ examined, steryl esters (136–138) and acyl steryl glucosides may accumulate in tracheophyte tissues (139, 140), and both of these forms are demonstrably incorporated into subcellular fractions (cf. refs. in table 4).

Arguments in favor of the role of sterols as membrane components have been documented for triterpenoids. Thus, the subcellular occurrence of triterpenoids as the free alcohols in bacteria and higher plants is analogous to that of the sterols

TABLE 9. Total sterol concentration in membranes.

	Sterols in lipid leaflet as percent of			
	Dry Weight	Wet Weight	Protein	Refs.
Photosynthetic Organisms				
Blue-green bacteria[b]		$3x10^{-3}$		20
Chloroplasts (tracheophyte)	$2x10^{-2}$–$2x10^{-1}$	$8x10^{-6}$–$5x10^{-4}$	10^{-1}	27,28,30
Endoplasmic reticulum	$8x10^{-1}$	$8x10^{-5}$–$7.5x10^{-3}$	10^{0}–10^{2}	27–32
Mitochondria	$1x10^{-1}$	$8x10^{-5}$–$4x10^{-3}$	$5x10^{-1}$–10^{1}	27–32,45, 72,73
Plasmalemma		$8x10^{-4}$	$4x10^{0}$–$2x10^{1}$	30,74
Nuclei	$3x10^{-2}$	$4x10^{-5}$–10^{-4}		27,28
Golgi app.		$9x10^{-5}$	$3x10^{2}$	72
Nonphotosynthetic organisms[a]				
Bacteria[b]	$5x10^{-2}$–$2x10^{-1}$			14,16,17
Bacteria[c]	$2x10^{0}$		$2x10^{1}$	75,76
Endoplasmic reticulum		$3x10^{-1}$–$2x10^{0}$	$4x10^{0}$	36,77–79
Mitochondria (rat liver)		$3x10^{-2}$	$2x10^{-1}$	36,79
Mitochondria (fungus)	10^{-1}–$2x10^{-1}$		$2x10^{-1}$	80
Nuclei		10^{-1}		36
Erythrocyte ghosts	$2x10^{1}$		$7x10^{0}$	36,78
Myelin	$2x10^{1}$		$6x10^{1}$	81,78
Plasmalemma (mammal)			$5x10^{0}$–10^{1}	78,82,83
Plasmalemma (slime mold)			10^{1}	84
Plasmalemma (fungi)	$2x10^{-1}$–$4x10^{0}$			80
Yeast protoplast	10^{1}			85
Golgi app.			$7x10^{0}$	79

[a]Data for mammals, unless otherwise indicated.
[b]Bacteria synthesizing sterols.
[c]Bacteria requiring sterols for growth.

found in membranes (table 9). When tetrahymanol, β-amyrin, or α-amyrin are added to vegetative *Phytophthora cactorum*, growth is stimulated without any metabolic transformations of the triterpenoids (101). The absence of triterpenoid metabolism is an indication that these substances act as mycelial membrane components. Similar observations were reported for mycoplasmas incubated with β-amyrin (141). Once triterpenoids are metabolized to their corresponding saponin, they no longer function as membrane constituents; rather, at least in insects (142) and fungi (123, 143), they may act as feeding inhibitors, perhaps by complexing with membrane sterols. Saponins and glycoalkaloids are not believed

to be present in the subcellular particles of the plants that synthesize them. It is assumed that they are deposited into intracellular vacuoles (122). The absence of saponins from the membranes of plants that synthesize them indicates that they are prevented from entering their sterol-containing membranes. This suggests a very specific intracellular discriminatory process operating in saponin-synthesizing plants which is lacking in animals and fungi, where saponins have deleterious effects (123, 142, 143).

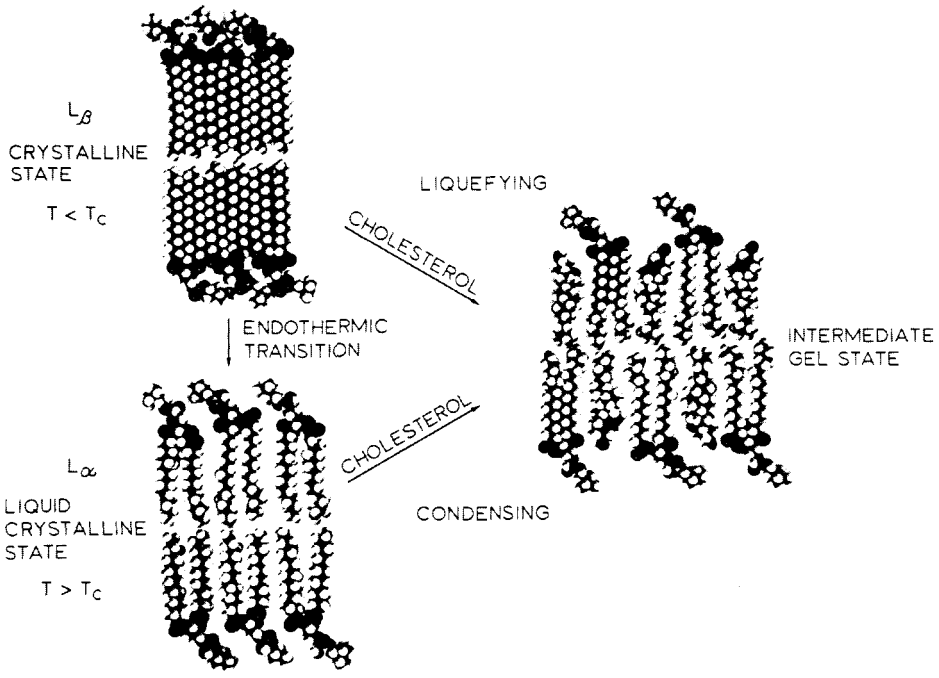
Some years ago, it was suggested that triterpenoids which are structurally equivalent to sterols could act as substitutes for sterols under certain circumstances (5). For instance, during germination, pea seeds do not initially synthesize sterols, but rather β-amyrin (25). Two days after the start of germination, β-amyrin synthesis decreases and sterol production is initiated. Apparently, a functional feedback on the 2,3-oxidosqualene cyclase (144) is derepressed (145) and the enzymes of the steroid pathway begin forming sterols in place of pentacyclic triterpenoids (fig. 3). The switch to the formation of sterols in peas favors their preferential incorporation into subcellular membranes, where they presumably fit better in the bilayer than triterpenoids.

When *Tetrahymena pyriformis* is incubated with a sterol, the synthesis of the native pentacyclic triterpenoid, tetrahymanol, is inhibited, the sterol is incorporated into the membranes of the protozoa (88), and growth of the organism is stimulated (107). On the assumption that growth reflects membrane function, the sterols seem to be preferred to the triterpenoids for membrane construction. It has been suggested on phylogenetic grounds that hopanoids preceded sterols in the evolution of bacteria (7, 8). If this is correct, then evolutionary ascent was accompanied by selection against triterpenoids, preference being given to the synthesis and incorporation of sterols into the membranes. The data on subcellular occurrence and biological effects tend to support this view.

Physico-Chemical Studies

Sterols and triterpenoids have been used in physico-chemical studies to demonstrate, among other things, that polycyclic isopentenoids can be incorporated into the lipid leaflet, align with the fatty acyl chains of phospholipids, and exhibit *in vitro* membrane properties similar to those of natural membranes with similar composition. Partially on the basis of model studies, it is generally accepted that membrane fluidity is due to the unsaturation of fatty acyl chains of phospholipids (146). In organisms which utilize sterols as architectural components, the degree of fluidity of phospholipids is presumed to be controlled by the addition of sterol (6). Whether the fatty acyl chains are in an ordered (gel) or a disordered (liquid) state is determined by temperature and sterol (4, 6, 147, 148). Thus, depending on the transition temperature and amount of sterol present in the model system, sterols can exhibit a condensing effect (more ordered or rigid) or a liquefying effect (less ordered) (cf. fig. 4). There is much speculation on the nature of cholesterol-phospholipid interactions, based primarily on model studies. The various membrane models, in which sterols are shown to be imbedded in the lipid leaflet, fall into two broad, but not mutually exclusive, categories. One has to do with the spatial fit of cholesterol interacting with the paraffinic chains of phospholipids (149–152), and the other one has to do with the capacity of the 3β-hydroxyl group of cholesterol to undergo hydrogen bonding with some other membrane component (152–159).

Cholesterol has been the principal sterol examined in model systems. The



results indicate that cholesterol is oriented perpendicularly to the bilayer surface (6) and forms a molecular complex with fatty acids (6, 148). The alterations in fluidity induced by the addition of cholesterol to phospholipid-containing liposomes was originally believed to be due principally to the repulsion by the sterol nucleus (149). Moreover, on the basis of chemical evidence, the cholesterol molecule was shown to affect the first 10 carbon atoms of the acyl chain, resulting in a reduction in the number of chain conformations, i.e., more order along the upper part of the chain, while simultaneously increasing the conformational freedom of the acyl chain along the lower half (6, 149, 152). Recent studies of the effects of a series of naturally occurring and synthetic sterols on the molecular order and motion in liposomes have shown that the "bulky" sterol nucleus is not the single molecular feature responsible for changing order. For instance, the longitudinal dimensions (160–162) as well as the extent of unsaturation of the side chain (163, 164), but not the degree of alkylation at C–24 (165), exert a significant influence on membrane rigidity.

Triterpenoids have also been studied, but less well than sterols, in model systems. While their physico-chemical properties are essentially similar, they differ from those of 24-desalkyl- and 24-alkylsterols in some respects. For instance, a glycolipid containing hopane, isolated from *Bacillus acidocaldarius*, exhibited a fluidizing effect on monolayers that was somewhat different from that of cholesterol (166). Monohydroxy triterpenoids, like sterols, orient themselves in monolayers perpendicular to the air-water interface and exhibit condensed isotherms (167),

but simple viscosity determinations on monolayers containing cholesterol, lanosterol, and α-amyrin have indicated that cholesterol films are in a more liquid state than monolayers containing lanosterol and α-amyrin, which produce rigid films (167). More direct data from micro-viscosity experiments with model and natural membranes containing a desmethyl sterol and sterols and triterpenoids with C–4 and C–14 methyl substituents gave the highest micro-viscosity value with desmethyl sterol, viz., cholesterol, lower values with the C–4 methyl-substituted sterols, and the lowest value with lanosterol, a 4,4,14-trimethyl steroid (168, 169). When cholesterol and lanosterol were incubated separately in lecithin-containing liposomes in which glucose had been trapped inside, the former was more effective in releasing glucose (169). From these model studies it has been inferred that the predominant reason why lanosterol cannot adequately replace cholesterol in model systems is not that it has double substitution at C–4 but that the 14α-methyl group is present (170, 171). The 14α-methyl group is known to destroy the planarity of the α- or back face of the sterol molecule (4, 171) and, as a result, presumably weakens sterol-phospholipid interactions, i.e., lessens the cooperative interactions with the fatty acyl chains, thereby altering bilayer fluidity (168). *In vitro* observations with tetrahymanol have also shown that pentacyclic triterpenoids alter fluidity in model systems (7).

Structure-Growth Relationships

It is generally believed that all microorganisms, those auxotrophic as well as those heterotrophic for sterols, incorporate exogenous sterols from the growth medium into their membrane structure and that their growth response reflects the effectiveness of those sterols in contributing to membrane function (4, 6). The premise is based on the striking correlation between the structural requirements for *in vivo* and *in vitro* sterol-SCA interactions (121–125, 172), for sterol-lipid interactions in model systems (6, 148, 152, 173, 174), and for the support of growth in sterol-dependent organisms (3, 4, 6). Thus, membranes generally have very similar structural requirements for sterols according to these studies. The structural requirements for function are: a free hydroxyl group at C–3, a

TABLE 10. Effect of sterols on the growth of sterol-requiring prokaryotes.

Sterols	Organisms				
	Mycoplasma hominis, Strain 07	*Mycoplasma mycoides*, Strain Y–5	*Mycoplasma* spp.	*Mycoplasma mycoides* subs. *capri*	*Treponema hyodysenteriae*
Cholesterol	++	++	++	++	++
Sitosterol	++	+	++	++	++
Stigmasterol	+		+		0
5α-Cholestane	0		0		
5α-Cholestanol	++	+	+	++	++
5β-Cholestanol	0		0	0	
7-Dehydrocholesterol	0	0	+		
Ergosterol	+	+	+	++	
Refs.	91,92	93	94	95	96

(++) Indicates maximal growth support, (+) minimal growth support (<75% of maximal growth), (0) no growth or growth inhibition; blank spaces indicate absence of data.

planar tetracyclic nucleus, and an intact side chain of 8 to 10 carbon atoms (3, 4, 6). However, more recently, structural features postulated for polycyclic isopentenoid membrane function have undergone some revisions in light of new information from various sources.

Numerous species of mycoplasmas and *Treponema hydysenteriae* have an obligatory nutritional sterol requirement for growth. The species shown in table 10 occur as parasites in mammals; their habitat is, therefore, rich in cholesterol (91–96). One would expect that cholesterol fits the membrane architecture of these organisms best and that no other polycyclic isopentenoids would be more effective. In fact, when sterol was added to the growth medium, no sterol induced better growth of these prokaryotes than cholesterol, but they were capable of discriminating between stigmasterol and sitosterol, the latter being more effective. Analogous observations in monolayer (175, 176) and bilayer (164, 165) systems

TABLE 11. Effect of sterols on the growth of sterol-requiring eukaryotes.

Organisms	*Paramecium aurelli*	*Tetrahymena pyriformis*	*Saccharomyces cerevisiae*	*Phytophthora cactorum*	*Phytophthora cactorum*	*Phytophthora infestans*	*Phytophthora megasperma*	*Phytophthora parasitica*	*Phytophthora cactorum*	*Phytophthora cactorum*	*Pythium periplocum*	*Drosophila pachea*	*Dermestes vulpinus*	*Drosophila melanogaster*	*Xylaborus ferrugineas*
Organismic Type	Protozoum[a]	Protozoum[a]	Yeast[b]	Fungus[a]	Fungus[a]	Fungus[a]	Fungus[c]	Fungus[c]	Fungus[c]	Fungus[c]	Fungus[c]	Insect[c]	Insect[c]	Insect[c]	Insect[c]
Refs.	105,106	107	102,103 104	90	97	98	99	99	100	101	99	177	178,179	180,181	182
Sterols															
Cholesterol	0	+	+	++	++	+	++	++	++	0	++	0	++	++	0
Campesterol			+	++						+			++		
22,23-Dihydrobrassicasterol			+							0			++		
Sitosterol	+		++	++	++	+	+	+	++	++	+	0	++	++	
Clionasterol	+		++												
Stigmasterol	++		++		+	++	+	+	++	++	+	0	+		
Poriferasterol	++		++												
Fucosterol			+							0			0		
7-Dehychocholesterol	0		+									++	++		++
Lathosterol			+									++			
22,23-Dihydroergosterol										0			++		
Brassicasterol	0		++							0			++		
Ergosterol	0	++	++			+	++	++	+	0	0	0	0	++	++
Ergostanol	0									0					
Cholestanol	0		+	0	++	++			++	0			++	++	
Sitostanol										+					
5α-Cholestane	0			0						0					
Coprostanol				0											
7-Dehydrositosterol												+	++		
7-Stigmastenol												++			
5,22-*trans*-Cholestadienol			+										++		

[a]Organism cultured in aerobic liquid medium.
[b]Organism cultured in anaerobic liquid medium.
[c]Organism cultured on solid medium.
For symbols, see Table 10.

have shown that sitosterol acts similar to cholesterol and is more effective than its 22-*trans*-dehydro derivative, stigmasterol. In the prokaryotes, only a few naturally occurring sterols have been examined for structure-growth response, but the eukaryotes that require a dietary source of sterols for growth have been studied more extensively. The growth response of twelve sterol-requiring eukaryotic organisms to various sterols is summarized in table 11.

Many of the eukaryotic organisms which require sterols are parasites of higher plants, e.g., the pythiaceous fungi and herbivorous insects. Unlike the prokaryotic parasites requiring sterols, the eukaryotic parasites that infest tracheophytes would be expected to prefer 24-alkylsterols to 24-desalkylsterols for optimum growth. For organisms which discriminate among 24-alkylsterols, not only is the degree of alkylation important for activity, but the stereochemistry of the

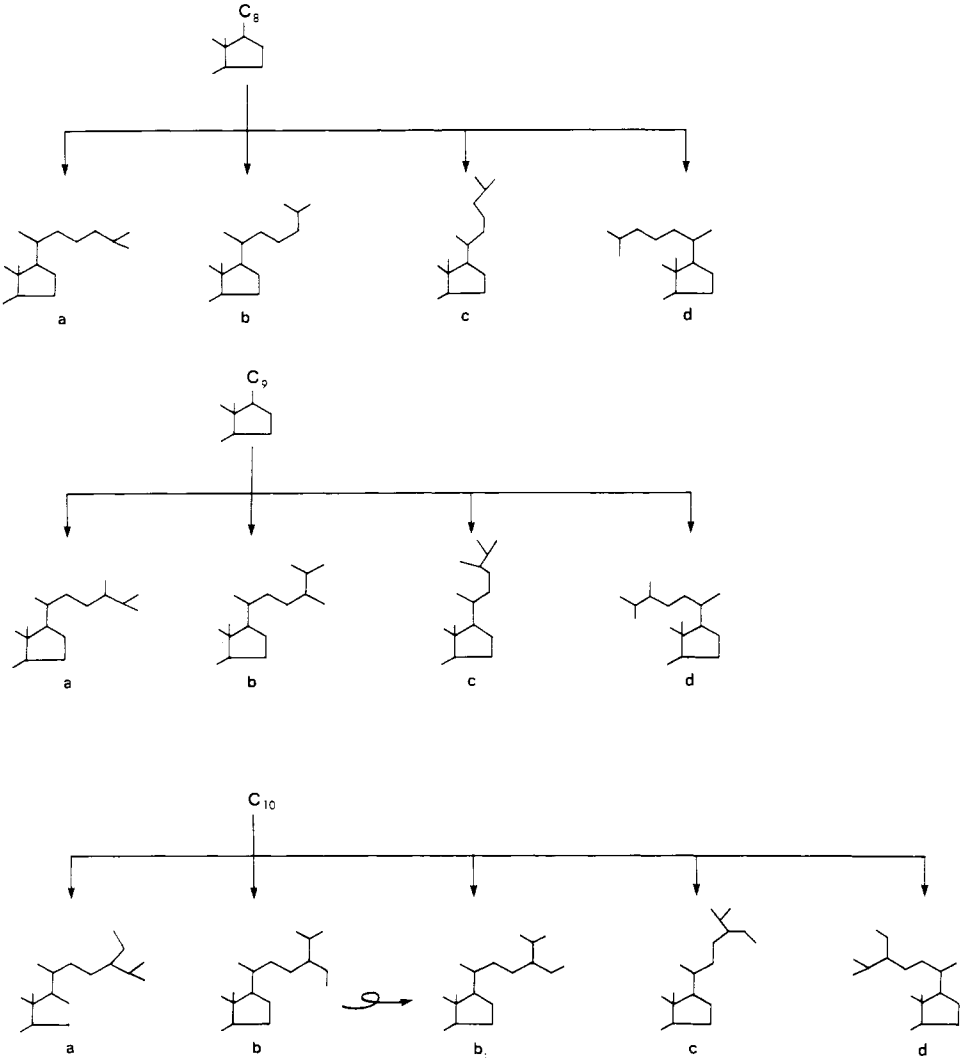


FIG. 5. Various rotameric states of 24-alkyl- and 24-desalkylsterols.

chiral carbons of the side chain also influences growth. Thus, many organisms prefer the sterol found in the host, which for highly evolved tracheophytes are 24α-ethylsterols (4), to sterols normally synthesized in fungi and animals, e.g., 24β-methylsterols and 24-desalkylsterols. The substantial differences among organisms in their structural requirements for growth contrast sharply with the assertion that biological membranes exhibit little specificity for sterol structures (183). The underlying assumption is that as long as polycyclic isopentenoid molecules possess the same nuclear features as cholesterol and an intact side chain of 8 to 10 carbon atoms, they can intercalate in the phospholipid bilayer in analogy to cholesterol. Recent *in vivo* and *in vitro* studies have cast doubt on these generalities and have demonstrated, for instance, that the chain length is crucial for membrane activity. Thus, a side chain having 10 carbon atoms will only act in a positive fashion when the addition of 2 extra methyl groups occurs at C–24. As is shown in fig. 5, naturally occurring membrane sterols, which normally are alkylated at C–24, do not possess a greater side chain length than cholesterol when the aliphatic chain assumes the staggered conformation or in any other rotomeric state. Thus, rotation by 180° about C–24 does not lengthen the longitudinal dimensions of the side chain in the case of 24-alkylsterols, and inversion at C–24 does not affect length to any significant degree. Addition of a methyl or ethyl group at C–24 does not lengthen the chain, but addition of a methyl or ethyl substituent at C–26(27) does increase the overall dimensions of the side chain. In order to test the influence of side chain on membrane structure, a homologous series of sterols was tested in a bilayer system with and without the addition of SCA (160–162) and in the metabolism and growth of anaerobic yeast (102–104, 184), protozoa (184, 185), and pythiaceous fungi (101, cf. also table 12). All of these studies have shown, in essence, that when activities are plotted against the number of carbon atoms in the side chain, a bell-shaped curve is obtained. Maximal activity coincides with a chain length of 5 or 6 carbon atoms, counting

TABLE 12. Effect of synthetic sterols on the growth of sterol-requiring organisms.

Sterols	Organisms			
	Saccharomyces cerevisiae	*Phytophthora cactorum*	*Tetrahymena pyriformis*	*Mycoplasma capricolum*
Ref.	4,102,103	101	187	188
Cholesterol	+	0	+	++
20-Isocholesterol	0		0	
20-Methylcholesterol	0		0	
20α-Hydroxycholesterol	0	0	0	
29β-Hydroxyporiferasterol		0		
trans-17(20)-Dehydrocholesterol	+		0	
Halosterol			0	
27-Desmethylcholesterol			0	
29-N-Ethyl-27-desmethylcholesterol			0	
26-Homocholesterol		+		
cis-17(20)-Dehydrocholesterol	0		0	
Cholesteryl methyl ether	+			+
Ergosterol	++	0	++	

For symbols, see table 10.

from C-20, which corresponds to all 24-desalkyl- and 24-alkylsterols found in natural membranes. The rare exceptions to this rule for naturally occurring sterols appear to be the unusual 10- or 11-carbon side chains of sterols found in sponges (186).

Following the discovery that *Tetrahymena pyriformis* synthesizes a pentacyclic triterpenoid in place of sterols as its membrane component (4, 5), it was of interest to investigate whether sterol-requiring organisms could utilize triterpenoids as sterol replacements. As is shown in table 13, in some organisms triterpenoids

TABLE 13. Effect of triterpenoids on the growth of sterol-requiring organisms.

	Organisms						
	Saccharomyces cerevisiae	*Saccharomyces cerevisiae*	*Phytophthora cactorum*	*Mycoplasma capricolum*	*Mycoplasma capricolum*	*Tetrahymena pyriformis*	*Dermestes vulpinus*
Refs.	4,102,103	189	101	141	168	107	178
Triterpenoids							
Cycloartenol	0		0	++			
Lanosterol	0	+	0	++	+		0
Lophenol	0						
β-Amyrin	0		+	+			
Tetrahymanol	0		+			+	
Lupeol	0	0	0				
Friedelin			0				
22(29)-Hopene			0				
Euphol				0			
α-Amyrin			+				
Sterols[a]	++	++	++	++	++	++	++

[a]Any of the 3β-hydroxysterols known to stimulate maximal growth.
For symbols, see Table 10.

may replace sterols for growth and in others they may not. This does not depend on whether the organism is a prokaryote or eukaryote, since *Mycoplasma capricolum* (a bacterial pathogen) and *Phytophthora cactorum* (a fungal pathogen) can utilize β-amyrin as an effective sterol replacement, albeit less well than sitosterol. The inability of *Saccharomyces cerevisiae* and *Dermestes vulpinus* to use triterpenoids as membrane components shows that some organisms have more specific structural requirements for membranes than others.

Structural Requirements for Membrane Function

The data on subcellular distribution of steroids and triterpenoids and on the growth response of sterol-requiring organisms exemplifies the structural specificity one would expect on the basis of their evolutionary advancement and ecological niche. Thus, the available evidence strongly suggests a relationship between the availability and selectivity towards polycyclic isopentenoids (whether formed endogenously or obtained exogeneously) and their utilization by cells in terms of membrane function.

Model studies indicate that when steroids and triterpenoids are added to lecithin-containing monolayers and bilayers, no other isopentenoid—synthetic or natural—exceeds the ability of cholesterol to modulate membrane fluidity (168–170). Moreover, cholesterol is more effective than many 24-alkylsterols (e.g., those that have unsaturated side chains) and triterpenoids in influencing membrane rigidity. Thus, cholesterol is the most suitable polycyclic isopentenoid for the control of fluidity in mammal and other vertebrate membranes. Biological data for mammals, where cholesterol is the dominant membrane sterol, are in agreement with those from model studies (cf. above). Mammals discriminate against 24-alkylsterols prior to entry into the circulating system at the level of intestinal membranes and those that finally enter the blood at the erythrocyte membranes (4, 148). They not only discriminate against dietary 24-alkylsterols but also against triterpenoids. Some triterpenoids are normally synthesized by mammals, e.g., lanosterol. However, when lanosterol accumulates in a cell culture, the cells lyse rapidly (120), and this indicates that it is deleterious to membranes. In short, cholesterol is generally preferred by mammalian membranes and by membranes of those pathogens that infest mammals; this is in agreement with the model studies.

The data and assertions derived from an examination of model systems and animal membranes do not necessarily reflect the structural requirements for plant membranes. Thus, subcellular organelles of photosynthetic and nonphotosynthetic plants have evolved the capacity to utilize polycyclic isopentenoids other than cholesterol.

The principal molecular features common to all polycyclic isopentenoids found in biological membranes is that the overall dimension approximates 20 Å. For membrane function, the process whereby the dimension is achieved is a matter of detail and is not necessarily the same for all intracellular membranes in nature. Generally, the structural requirements for triterpenoids are similar to those for sterols, except in the case of pentacyclic triterpenoids. Thus, isopentenoids do not have to be tetracyclic nor completely planar to serve as membrane components in certain plant and bacterial systems. β-Amyrin, having a D/E *cis*-ring juncture, which produces a puckered, rather than a flat shape at that ring juncture, serves as a sterol replacement in certain fungi and bacteria. All pentacyclic triterpenoids found in membranes so far have axial groups on both sides of the molecule. Thus, the presence of axial groups at C–4 and C–14 does not prevent the pentacyclic compound from acting as a sterol replacement, as the 14α-methyl group of certain tetracycles is believed to be deleterious to activity in natural and model systems (4, 171). In the pentacyclic triterpenoids, the 14α-methyl group does not destroy the α-face planarity as it does in sterols. For instance, examination of a molecular model of tetrahymanol shows that it approximates a flat structure in its ring system and possesses four axial methyl groups at C–4, C–14, C–18, and C–22 (triterpenoid numbering system (23)), all of which lie 90° to the plane of the molecule. These four methyls, in effect, produce a plane of their own, which retains a flatness or smoothness of the molecule.

While the stereochemistry and position of methyl groups of the ring systems is of some importance in sterols and triterpenoids, the configuration at C–17, C–20, and C–24 is a determining factor for biological activity of tetracyclic isopentenoids. For instance, a polycyclic isopentenoid having a 17β-oriented side chain is necessary for supporting growth since lanosterol supports growth, while its 17α-enantiomer euphol does not (141). The *R*-configuration at C–20 is obliga-

tory for activity in yeast (102) and protozoa (187) since cholesterol but not 20-isocholesterol is used. The configuration and extent of alkylation are also determining factors for growth support in many organisms. Thus, 24β-methylcholesterol supports better growth than its epimer in anaerobic yeast cultures (102), and 24α-methylcholesterol stimulates vegetative growth to a greater extent than its epimer in *Phytophthora cactorum* (101). Another molecular feature which may be a determinant factor for function is the introduction of additional double bonds at C-22(23), C-24(25), C-24(28), and C-25(27), which normally occur in plants. Some examples of sterols with unsaturated side chains that accumulate in plants are fucosterol in brown algae, ergosterol in yeast, stigmasterol in tracheophytes, poriferasterol in green algae, and desmosterol in red algae (4). The presence of these double bonds may have two effects: (a) freezing the side chain into a particular conformation and thereby reducing the flexing of the aliphatic chains, which would tend to diminish cooperative interactions with *n*-acyl chains in the membranes; and (b) introducing a π-lobe system which, like the double bonds in fatty acids, would tend to increase Van der Waals attraction, also tending to influence lipid-lipid interactions and membrane fluidity. Thus, the primary difference between sterols and pentacyclic triterpenoids, both of which may occur in membranes, is that sterols possess an acyclic side chain, while the pentacyclic compounds do not. We suggest that the mobility and flexibility of the side chain is a contributing structural feature of polycyclic isopentenoids for membrane function.

We have discussed the role of sterols, steroidal sapogenins and triterpenoids as affecting membrane function by acting as a structural component, perhaps through an interaction with phospholipids in certain membrane systems. Undoubtedly, steroids have not only structural, but also dynamic functions in biological membranes which are again related to structure. Thus, steroids such as progesterone, ouabain, and digitoxin may exert their physiological effects in mammals (190) or plants (191) through, possibly, interaction with membrane proteins (192). The mechanism of action of steroid hormones was earlier believed to be due to membrane effects resulting from their penetration into the lipid leaflet and interaction with phospholipids (193). Subsequent monolayer studies showed that the presence of more than one functional group militates against membrane activity (194). Recently, some steroid hormones have been shown to have protein receptors both in subcellular fractions and in the cytosol (192, 195), and a membrane-action of progesterone has been postulated (192). Chemotactic effects of steroids, such as antheridiol and oogoniol, may also be due to action on membranes. More work will be required to establish the physical basis of such dynamic effects.

That proteins present in membranes recognize subtle differences in steroid structure is significant, and we suggest that polycyclic isopentenoid-protein binding may be a common phenomenon in plants. While certain plant membranes contain sterols in concentrations well below those required for altering fluidity in model systems (e.g., 22–33 mol%), they may contain very high sterol to protein ratios. How then do sterols and other isopentenoids function in these membranes? It has been shown that certain lipids surround the lipid annulus of membrane proteins (196), and we think it is probable that some polycyclic isopentenoid-protein complex may be involved, together with phospholipids, in membranes. This complex could then regulate the characteristics of the polar lipid bilayer in at least two imaginable ways: (a) by acting as a bridge, i.e., simultaneously binding with protein and polar lipid (phospholipid, sulfolipid, glycolipid);

and (b) by altering the higher structure of the protein, which in turn alters the architecture and function of the membrane. One can readily imagine binding to protein as occurring on the front (β-face) of the sterol and triterpenoid, which contains the angular methyl groups. The latter would allow a 3-dimensional "hold-fast". All naturally occurring sterols and triterpenoids possess these two angular methyl groups. The suggestion of β-face binding is supported by the stereochemical requirements at C–20 of sterols, which have been previously described. When a CH_3- or OH-group protrudes on the β-face of a sterol, the molecule does not support the growth of anaerobic yeast (102) and inhibits the growth of protozoa (187) and pythiaceous fungi (90). Binding to polar lipid could then occur (bridge-model) on the back (α-face), which is smooth in the sense that only H-atoms protrude more or less in a single plane. In the case of pentacyclic triterpenoids, where more than one axial methyl group is present on the back face, the α-face retains 3-dimensionally smooth characteristics. The idea of polycyclic triterpenoids being bound to proteins suggests that the recognition sites on the protein for sterols and triterpenoids are influenced by evolutionary pressures.

Thus, it is not unreasonable to believe that these pressures, which are mated to membrane structure and function, have selected in favor of a biosynthetic pathway through the cyclization of squalene epoxide to the protosteroid cation rather than to the prototriterpenoid cation. As previously shown, tetracyclic triterpenoids formed *via* the triterpenoid cation are rather ineffective membrane components, while those formed *via* the protosteroid cation either act as membrane components—albeit not as effectively as sterols—or they are metabolized to sterols. Binding of the polycyclic isopentenoid to the protein does not contravene the existence of pure sterol-phospholipid complexes, but the latter is dependent less on steric phenomena and more on the absolute concentration of sterol and on the mol% of the added isopentenoid, while the former entails greater dependence on the stereochemistry of the molecule. Finally, in accordance with the bridge-model, cells must have evolved the proteins but did not have to evolve the synthetic machinery for isopentenoid production since in many organisms the isopentenoid may be obtained from dietary sources. The absence of a protein specific, e.g., for tetrahymanol in yeast, would explain why tetrahymanol is deleterious to membrane function in yeast but not in *Tetrahymena pyriformis* or *Phytophthora cactorum*.

In summary, while there is a great deal of information available on the intracellular localization of sterols in mammals and on the function of cholesterol in the lipid leaflet of artificial and natural membranes, much less work has been done on the subcellular occurrence and function of 24-alkyl sterols and of tetracyclic and pentacyclic triterpenoids. Although the histological structure and biological processes of membranes from photosynthetic and nonphotosynthetic organisms are basically similar, the composition of polycyclic isopentenoids in these organelles may vary significantly from one organismic group to another. We are only beginning to understand the contribution made by the structure of sterol and triterpenoid molecules toward membrane functions in mammalian systems and we know even less about this in plants. Apparently, the membranes of photosynthetic plants utilize 24-alkyl sterols in preference to 24-desalkyl sterols and triterpenoids, whereas membranes of animal origin show a preference for 24-desalkyl sterols and exclude triterpenoids. The reason for this is still obscure. The mechanisms whereby pentacyclic triterpenoids, 24-alkyl sterols, and sterol

conjugates regulate the fluidity of the bilayer and other functional activities of plant membranes await further study.

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