6 August 1982, Volume 217, Number 4559 SCIENCE

Chemical Fossils: The Geological Fate of Steroids

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Summary. Steroids are used to illustrate some of the significant advances that have been made in recent years in understanding the biological origin and geological fate of the organic compounds in sediments. The precursor sterols are transformed, initially by microbial activity and later by physicochemical constraints, into thermodynamically more stable saturated and aromatic hydrocarbons in mature sediments and petroleums. The steps in this transformation result in a complex web linking biogenesis, diagenesis, and catagenesis. Indeed, the complexity and variety of biological lipids such as the steroids are evidently matched in the corresponding geolipids. The extent of preservation of the biochemical imprint in the structures and stereochemistry of these geolipids, even over hundreds of millions of years, is startling, as is the systematic and sequential nature of the geochemical changes they evidently undergo. This new understanding of molecular organic geochemistry has applications in petroleum geochemistry, where biological marker compounds are valuable in the assessment of sediment maturity and in correlation work.

The number of compounds with an obvious biological origin detected in the geosphere has increased dramatically in recent years, as shown in Fig. 1 for sterols and for steroid and pentacyclic triterpenoid alkanes. Such compounds have been termed chemical fossils (*1*), biological marker compounds, or molecular fossils. A chemical fossil is any organic compound in the geosphere whose carbon skeleton suggests an unambiguous link with a known natural product. Common examples are listed in Fig. 2 along with their presumed precursors.

The increase in the numbers of chemical fossils found (see Fig. 1) is linked partly to the development over the past decade of computerized gas chromatography–mass spectrometry (GC-MS). Sedimentary organic extracts and crude oils are complex mixtures; they are frequently separated by "wet chemical" methods into fractions containing more than an estimated 10^3 simple components amenable to partial analysis by gas chromatography. Thus the combination of a glass or fused silica capillary column with high resolving power and a fast scanning mass spectrometer (< 2 seconds per decade) is a particularly valuable asset. The data acquired in a single GC-MS analysis are of sufficient amount and complexity to require a computer for both acquisition and appraisal. Much use is made of computer-constructed mass fragmentograms (plots of the intensity of an ion characteristic of a particular compound type against GC retention time). Fractionation before GC-MS is mainly by chromatography (thin-layer, column, and high-performance liquid) and selective adduction (for instance, urea) and occlusion (molecular sieves).

At present, the most convenient part of the organic matter for the molecular approach is that (the lipids) extracted from a sedimentary rock with organic solvents. This fraction rarely forms more than 5 percent of the total organic matter; the other 95 percent comprises (depending on the maturity of the sediment) fulvic and humic acids, humin (all soluble in aqueous base), and kerogen, a complex polymeric material, insoluble in organic and aqueous solvents (*2*). It appears that the components soluble in base disappear with a mild increase in diagenetic temperature (to about 50°C). Chemical fossils can also be released from kerogen by pyrolysis in the laboratory (*3*, *4*), but such products have not been examined in the same detail as their extractable equivalents. However, as the understanding of extractable chemical fossils improves, the molecular approach should be applied to kerogen. In addition, chemical degradation (*5*) of kerogen should be studied further. The potential has been indicated by treatment of kerogens or sedimentary polar fractions with reagents which selectively cleave ether linkages, yielding products which contain acyclic isoprenoids with head-to-head linkages, thought to arise from the glyceryl ethers originally contributed by methanogenic bacteria (Fig. 2) (*6*).

The pyrolysis of kerogens is thought to resemble, in part, the processes of oil generation in sedimentary sequences. The soluble components formed, together with the soluble portion inherited from deposition, are believed to be expelled from the source rock. Migration and accumulation of the resultant hydrocarbons lead to oil pools. Increasingly, the knowledge and analytical methodology developed for chemical fossils in sedimentary rock extracts are being applied to investigation of the same compounds in crude oils and hence to elucidation of the processes involved in their formation.

Detailed pathways, connecting natural products to stable geochemical intermediates, can now be drawn for several types of compound, encompassing earth processes ranging from chemical alteration in water columns to crude oil formation. These geochemical degradative pathways have analogies with biosynthetic pathways; indeed, the two can be linked as significant features of the global carbon cycle. In this article the geochemical fate of steroids is taken as an

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0036-8075/82/0806-0491$01.00/0

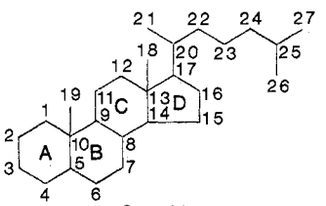
example. The reactions involved allow assessment of (i) early diagenetic effects (mainly microbial reworking), (ii) the region of thermal maturation between Recent sediments and the onset of greenschist facies metamorphism, and (iii) aspects of the environment of deposition, including the nature of the original organic input.

The history of steroidal compounds in geological materials stems from attempts to explain the optical activity of petroleum (*7*). Similarities in the properties of sterols and of petroleum fractions were used to demonstrate a genetic relationship and thence the biogenic nature of the latter (*8*). Indeed, the supposition that steroids were constituents of petroleum (*9*) and the isolation of two "sterols" from soil (*10*), although they were probably triterpene mixtures (*11*), preceded the elucidation of the structure of cholesterol (*12*). However, the recognition that sterols could be transformed into petroleum-like substances by heat or chemical treatment (*13*), despite the inappropriateness in geological terms of the conditions used, was an important step. The first confirmation of steroidal compounds in sedimentary material, β-sitosterol and β-sitostanol in peat (*14*), was made in 1958, although earlier mass spectrometric studies had shown C_{27} to C_{29} sterols in soil extracts (*15*).

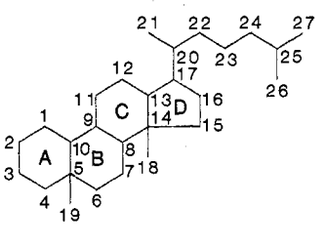
Derivatives of numerous other biolipids occur widely in sediments and petroleums. Figure 2 includes selected precursor-product relationships not discussed here.

The basic scheme of sterol diagenesis or catagenesis with major classes of compound arranged according to functional group, in relation to their occurrence and major reaction types, is shown in Fig. 3. The term diagenesis, as used by the organic geochemist, refers to alteration effects during sediment consolidation, where microbial reworking is a major aspect. Long-term, mainly thermal reactions at lower depths include those in the zone of oil generation and are covered by the term catagenesis. The pathways of sterol diagenesis and catagenesis shown in Fig. 3 are discussed in turn, including stereochemical aspects. Stereochemical changes as geochemical indicators were first used for Quaternary dating with α-amino acids (*16*). Configurational isomerization reactions also occur in steroidal hydrocarbons, but these compounds can, in the absence of deep burial, take longer than the Phanerozoic (about 600 million years) to reach equilibrium.

The insert below shows the carbon skeletons of steroids and rearranged steroids (diasteroids), illustrating the numbering system and ring nomenclature. In the text, specific positions are given as, for example, C-20 with Δ used to denote positions of unsaturation, as in Δ^5-stenols.



Steroids



Rearranged steroids (diasteroids)

Imprint of the Biosphere on Contemporary Bottom Sediments

The lipid compositions of contemporary bottom sediments are dependent on the source organisms and on the physical, chemical, and microbial influences that affect or modify them during their passage to the sediment.

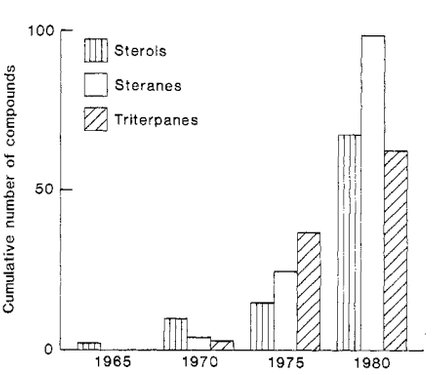


Fig. 1. Growth in the number of sterols, steranes, and triterpanes identified in geological materials over 5-year periods since 1965. The cumulative histograms include compounds recognized by comparison with either authentic reference compounds or suitable homologs, but do not take account of components suggested solely on mass spectral grounds. Sterol, sterane, and triterpane histograms are based on data from the following sources. Sterols: before 1965 (*14*), 1966 to 1970 (*114*), 1971 to 1975 (*41, 115*), 1976 to 1980 (*18, 28–32, 41*); present number = 80 (*116*). Steranes: 1966 to 1970 (*117*), 1971 to 1975 (*35, 70, 118*), 1976 to 1980 (*62, 69, 76, 77, 113, 119*); present number = 105 (*120*). Triterpanes: 1966 to 1970 (*121*), 1971 to 1975 (*122, 123*), 1976 to 1980 (*62, 112, 113, 124, 125*); present number = 73 (*126*).

The lipids of an organism play important roles in membrane structures, in energy budgeting, as protective waxes, and as pigments. Their composition is adapted and balanced to meet the requirements of the organism so that, for example, there are significant differences between the lipids of terrestrial and aquatic plants (*17*). The lipids in contemporary bottom sediments are derived from living systems and reflect inputs from various organisms (*18, 19*). Hence, differences in the nature or relative abundance of contributing organisms can be expected to produce changes in the lipid composition of the underlying sediment. The location (lacustrine, estuarine, shelf, or open marine) can be significant. Similarly, climate (tropical, temperate, or arctic) and seasonal fluctuations, such as periodic algal blooms, play a major part in determining biological populations. Finally, nutrient supply is important; for example, in upwelling areas the supply of silicon and other elements provides favorable conditions for diatom growth and may encourage periodic algal blooms. Single species can dominate modern environments, such as the Black Sea (*20*), and are well documented in the fossil record (*21*).

Many interrelated factors determine the survival of lipids, including the topography of the depositional basin, the oxicity and circulation of the water column, and the rate of sediment accumulation. In particular, these factors influence the microbial populations of the water column and sediment, which in turn play a significant role in the degradation and recycling of organic matter. In marine systems more than 95 percent of the total productivity of the euphotic zone is typically recycled within the upper portion of the water column (*22*), so that the fraction of this pelagic production reaching the sediment may not be representative of the whole. In most environments the particulate flux of organic matter represents a significant transportation pathway through the water column, especially as capsular fecal pellets of zooplankton (*23*). The mechanisms whereby terrigenous materials are introduced to bottom sediments include river inputs and land runoff (*24*). Eolian inputs are also significant in some areas (*25*) and turbidity currents can bring materials from continental shelves into the deep sea (*26*). The extent to which lipids are associated with particulate and dissolved organic matter in aquatic systems has yet to be suitably documented. Overall, the various combinations of the factors that influence sedimentary lipid distributions can lead to marked changes in

lipid composition within a somewhat restricted area (*27*).

The result of the processes of biosynthesis, predation, degradation, and transportation is the introduction into sediments of a complex mixture of compounds from terrestrial, bacterial, and algal and other aquatic sources. In many instances the structural specificity or distribution of lipids reflects their origin and provides an imprint of the biota contributing to the environment.

Here, we focus on sterols in the marine system. The major sources appear to be phytoplankton (*18, 28–32*), for example, diatoms (*29*) and other algae such as dinoflagellates (*30*), terrestrial plants (*27*), and higher organisms such as coelenterates (*29*) and sponges (*18*). The contributions of such different organisms can be recognized from the structures of the sterol side chains (Fig. 4). For example, it appears that 4-desmethylsterols $\leq C_{26}$ and $\geq C_{30}$ are characteristic of algae or higher marine organisms such as sponges, coelenterates, and scallops (*18, 33, 34*), since higher plants are not known to synthesize 4-desmethylsterols outside the C_{27} to C_{29} range (or 4α-methylsterols). In addition, the configuration at C-24 is of taxonomic importance (*35*); the 24β(H) configuration is dominant in marine plankton, whereas higher plants biosynthesize mainly 24α(H) diastereoisomers (Fig. 4).

The range of sterol markers is limited since many are biosynthesized almost ubiquitously (Fig. 4). Also, the more specific compounds may only be minor

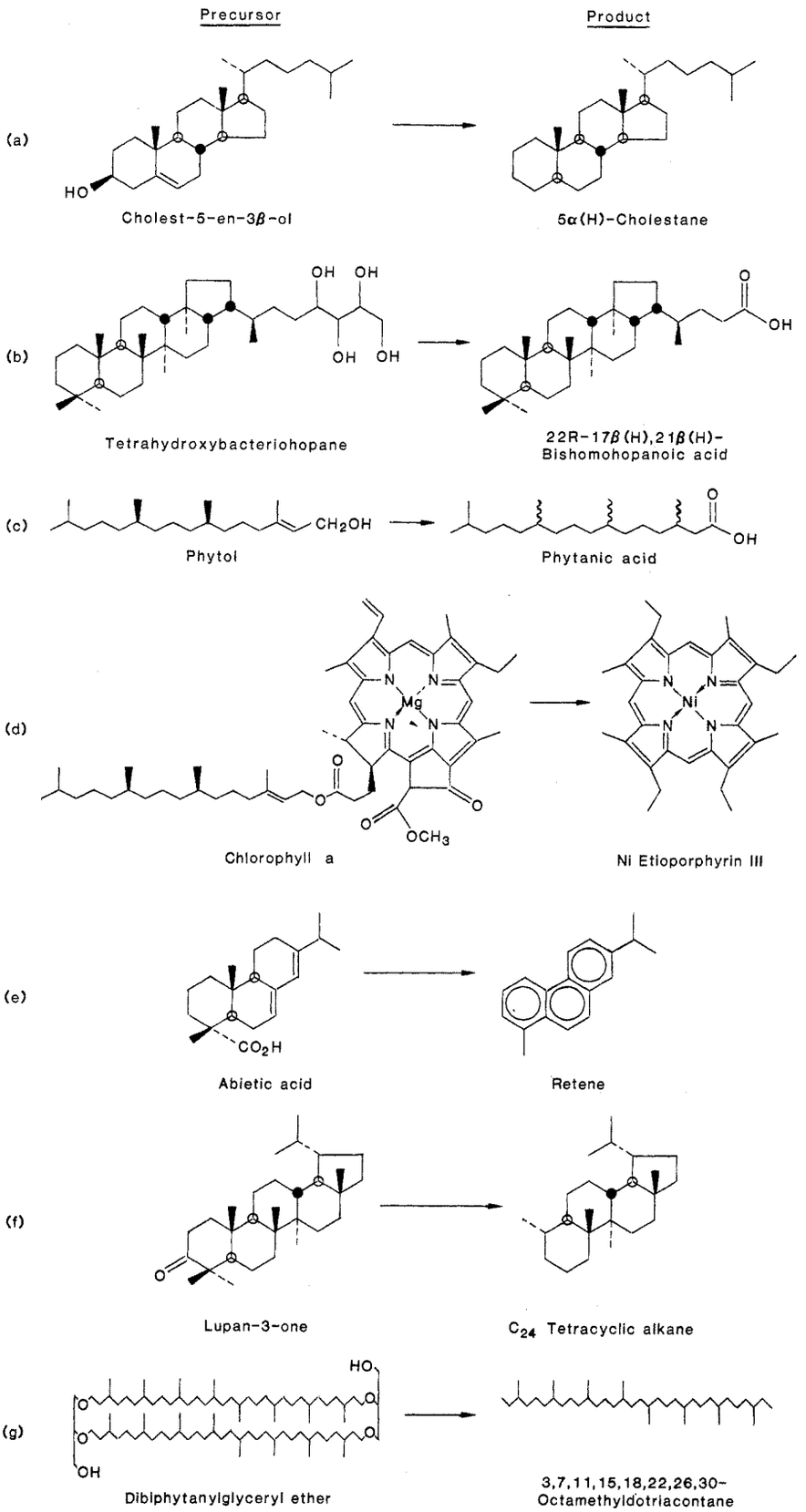


Fig. 2. Examples of postulated precursor-product relationships. The precursors are well-known natural products (biolipids). None of the products are biosynthesized directly, but all occur in sediments. [The conventions of thickened and dashed lines to represent β and α substituents, respectively, and solid and open circles to represent β(H) and α(H) substitution, respectively, are used. Sinuous lines represent substituents of uncertain or mixed (that is, both α and β) stereochemistry.] (a) Steroid transformations are the focus of this article; the diagenetic conversion of sterols to steranes is discussed in detail. (b) Polyhydroxybacteriohopanes are considered precursors of the ubiquitous extended hopanoids found in Recent and ancient sediments and in petroleums (*111, 127*). Mild oxidation of the tetrahydroxyhopane shown (*127*) would generate the C_{32} 17β(H),21β(H) acid, which is usually the dominant hopanoid acid in sediments (*18, 29, 123, 128*). (c) The phytanic acid in immature sediments (*129*) is thought to derive from phytol, a supposition backed by stereochemical studies (*129*). In more mature samples mixtures of all the stereoisomers are found (*76*). (d) The alkylation pattern of the C_{32} etioporphyrin III component of gilsonite bitumen (*130*) is consistent with an origin from chlorophyll a, allowing for loss of the phytol side chain, reduction of the vinyl and carbonyl groups, loss of the methyl ester function with cleavage of the isocyclic ring, and aromatization of the macrocycle. (e) The diagenetic formation of retene from abietic acid is supported by the recognition of intermediates (notably dehydroabietic acid, dehydroabietane, dehydroabietin, simonellite, and 1,2,3,4-tetrahydroretene) in sediments (*112*). (f) The C_{24} tetracyclic alkane is thought to form by photomimetic or photochemical degradation of the A ring of the 3-oxytriterpenoid lupan-3-one (*125*). Alkene and carboxylic acid intermediates of this degradation have been identified in sediments (*125*). (g) The acyclic isoprenoid alkanes with head-to-head linkages found in petroleums (*131*) are thought to derive from the glyceryl ethers of archaebacteria (*109*), the only organisms known to possess such a linkage.

components of the sterols, making their recognition difficult. It is also not clear whether all the side chains are equally resistant to degradation before incorporation in the sediment. For instance, zooplankton can generate cholest-5-en-3β-ol (C_{27}) by dealkylation of C_{28} and C_{29} sterols (*36*).

Variations in the nucleus are less numerous and virtually nonspecific (except C-4 substitution). For example, $\Delta^{5,7}$-stenols occur in fungi, marine yeasts, algae, and invertebrates (*37, 38*), whereas Δ^5-stenols and 5α(H)-stanols appear to be ubiquitous and Δ^7-stenols are found in many marine organisms (*39*). In addition, there are microbial and diagenetic pathways for the interconversion of the nuclei before deposition or within the upper levels of sediments (see below), which further restrict their useful specificity. However, the structural relationship between 4α,23,24-trimethyl-5α(H)-cholest-22-en-3β-ol and its 5α(H)-stan-3-one counterpart makes obvious their common origin from dinoflagellates (*40*).

A major discrepancy between the known sterol distributions of organisms and sediments is the greater abundance of 5α(H)-stanols in the latter. Although this has been attributed to reduction of Δ^5-stenols in the sediment (*41*), it now appears that stanols are more abundant in certain organisms than previously recognized, especially at specific growth stages (*38*). In addition, Δ^5-stenols appear to be more susceptible to oxidative degradation in sediments than 5α(H)-stanols (*42*). That sediments act as a sink for lipids from many sources is demonstrated by a comparison of the simple sterol distribution of a marine alga with the complex one in a diatomaceous ooze from the Japan Trench (Fig. 5). In the latter, several compounds can be related to individual inputs on the basis of their structural specificity (see Figs. 4 and 5). In nonmarine sediments the distribution of sterols is usually much simpler (*41, 43*), with fewer than 15 sterols compared to more than 50 in many marine sediments (*18, 29, 44*). This difference may reflect the smaller variety of organisms contributing to lacustrine sediments and the apparently simpler sterol composition of nonmarine organisms (*45*).

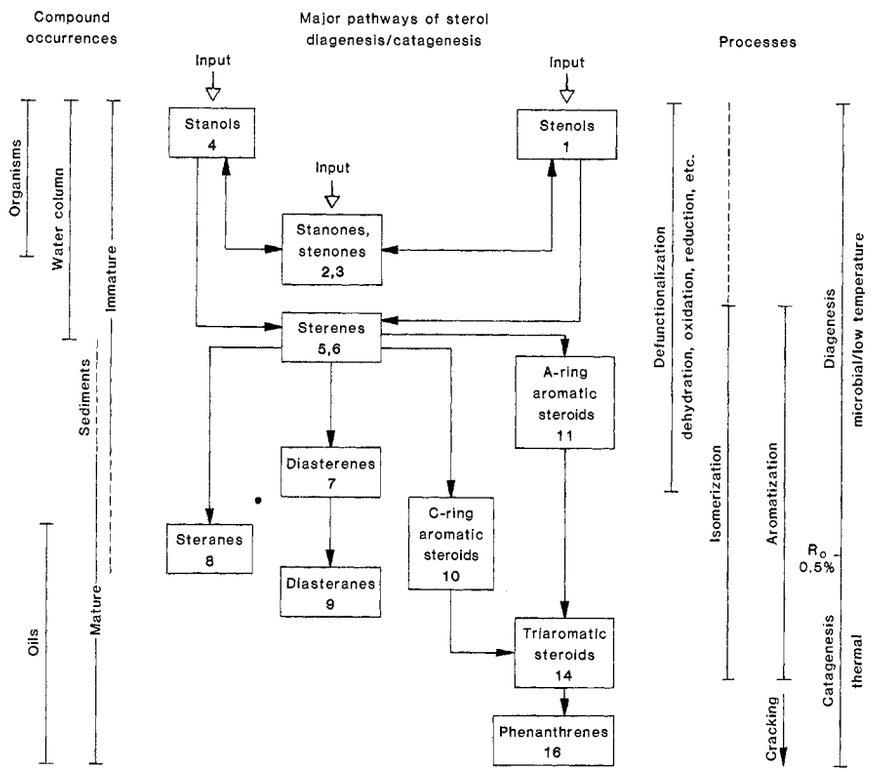


Fig. 3. A schematic summary of the presumed geological fate of the major natural product steroids. The biological and geological ranges of occurrence of steroids and the main processes that effect the transformations of steroids are also shown. A more detailed picture of the alteration of steroids, including structural and stereochemical changes, is presented in Fig. 6 and discussed in the text. The terms stenol and stanol refer to nuclear unsaturated and saturated sterols, respectively. Similarly, stenone and stanone refer to nuclear unsaturated and saturated sterones, respectively. The only natural product inputs to sediments considered here are sterols (**1** and **4**) and sterones (**2** and **3**), although steroidal acids (*48*), steryl esters (*49*), and steryl ethers (*18, 50*) may be additional sources of sedimentary steroids.

Diagenesis and Catagenesis

Tissot and Welte (*46*) propose a division of the progressive alteration of sedimentary organic matter between deposition and greenschist facies metamorphism (~200° to 300°C) into three stages: diagenesis, catagenesis, and metagenesis. Metagenesis involves the degradation of most organic molecules with the evolution of methane and formation of graphite; chemical fossils have little bearing on this stage. Tissot and Welte place the diagenesis-catagenesis boundary at a vitrinite reflectance (R_0) of ~0.5 percent. Here we consider two zones: (i) early diagenesis, and (ii) late diagenesis and catagenesis (Fig. 3). In early diagenesis the main agents of change are biochemical (microbial) and low-energy physicochemical reactions, which convert biolipids into more stable geolipids (chemical fossils). These relatively short-term reactions may be facilitated by the pore water chemistry, reactions with inorganic species, and the slight temperature rise consequent upon burial. Temperature and pressure rises are generally small in this zone, whereas for the longer term changes associated with late diagenesis and catagenesis, the main agent of alteration is thought to be heat, temperature rises being ~40° to 200°C. A convenient arbitrary division between early and late diagenesis corresponds approximately to the stage where natural product compounds have essentially lost their functional groups and (i) are present as alkanes and as partially and fully aromatized hydrocarbons, (ii) have been bound into kerogen, or (iii) presumably exist, in some cases, as stabilized heteroatomic aromatic compounds (*47*).

Early diagenesis. During early diagenesis, steroidal biolipids are converted into geolipids, whereby their functionalities are altered but stereochemical features remain largely unaffected. The major steroidal classes derived from organisms are sterols and sterones, with the former markedly more dominant. Steroidal carboxylic acids and esters are also biosynthetic products that occur in sediments (*48, 49*), but their diagenetic fate is not considered here. Steryl ethers are also found in sediments but their origin remains uncertain (*18, 50*). They appear to represent direct inputs rather than diagenetic products, in view of the discrepancies between the carbon number distributions of their sterol moieties and those of the co-occurring sterols (*18*).

In the water column and upper sediment, transformations mediated by microbes may occur (Fig. 6). First, Δ^5-

stenols (**1**) may be reduced to 5α(H)- and 5β(H)-stanols (**4a** and **4b**) (*41*) through sterone intermediates, although the stanols and sterones can also originate directly from organisms. Indeed, it now appears that the major source of 5α(H)-stanols is direct biological input, as illustrated by sediments where there is little correlation between the distributions of Δ^5-stenols and 5α(H)-stanols (*18*). Radiolabeling studies have also suggested that 5α(H)-stanols (**4a**) can be oxidized to their 5α(H)-stanone (**3a**) counterparts (*51*), illustrating the possibility of their diagenetic interconversion. Second, an isomerization that can occur in shallow sediments is conversion of 5β(H)-stan-3β-ols (**4b**) to their more stable 5β(H)-stan-3α-ol (**4c**) counterparts (*29, 52*). The appropriate 5β(H)-stan-3-ones are probable intermediates, and it remains open whether 5β(H)-stan-3α-ols are derived directly from these components, themselves formed from ster-4-en-3-ones (**2**), or from precursor 5β(H)-stan-3β-ols. Third, widespread products of early diagenesis (*52, 53*) appear to be Δ^2-sterenes (**5a**) and $\Delta^{3,5}$-steradienes (**6** in Fig. 6), which can also form rapidly within the water column (*54*). The possibility that sterenes are derived directly from organisms is supported by a single report of $\Delta^{3,5}$-cholestadiene in the isopod *Ligia oceanica* (*55*), and such a source seems unlikely. Rather, sterenes are generated from dehydration, with Δ^5-sterols giving rise to $\Delta^{3,5}$-steradienes and 5α(H)-stanols to Δ^2-sterenes (*52, 53*). These are the main sterene series in immature sediments and occur, like sterols, with a wide range of side chains (*18, 29, 52*). Indeed, the similarity between the side chain distributions of Δ^2-sterenes and 5α(H)-stanols and $\Delta^{3,5}$-steradienes and Δ^5-stenols, respectively, in a few sediments (see below) provides circumstantial evidence for their product-precursor relationships. The 5β(H)-ster-2-enes (**5b**) tentatively identified in sediments (*18*) can be envisaged as analogous dehydration products of the minor 5β(H)-stanols (Fig. 6). Other sterols (Δ^7-stenols, $\Delta^{5,7}$-stenols) may also undergo dehydration, but the expected products ($\Delta^{2,7}$-sterenes) have not been identified, perhaps because of the relatively low abundance of their precursors relative to Δ^5-stenols and 5α(H)-stanols (*18*).

Another process that may be partly microbially mediated is the formation of A-ring monoaromatic steroids (**11** in Fig. 6). These compounds occur in shallow or immature sediments (*53, 56–58*) and are thought to derive from $\Delta^{3,5}$-steradienes by microbial hydroxylation and subsequent dehydration (*53, 58*). Their formation from other sterol precursors (such as $\Delta^{5,7}$-stenols) has also been suggested (*18, 53*). The A-ring aromatization (whether it follows triene formation or occurs at the same time as dehydration is uncertain) is accompanied by the appearance of the C-19 methyl at C-1 or C-4, a reaction that may be catalyzed by clays (*58*). Analogous B-ring aromatics have yet to be fully characterized, but may form by similar mechanisms.

The sterenes generated in the water column and upper levels of sediment are not stable during subsequent diagenesis. The Δ^2-sterenes isomerize to the more stable Δ^4- and Δ^5-sterenes (**5c** and **5d**) (*18, 29, 59*) and, under suitably acidic conditions, can undergo in part clay-catalyzed rearrangement to diasterenes (**7** in Fig. 6) (*59*). This rearrangement has been simulated by heating 5α(H)-cholestan-3β-ol with montmorillonite (*60*), but it can also be achieved by acidic treatment of the same substrate or the Δ^5-sterene (*61*). This is a good illustration of a geological transformation following that observed in the laboratory. The initial products have $\Delta^{13(17)}$ unsaturation and unchanged 20*R* stereochemistry, but

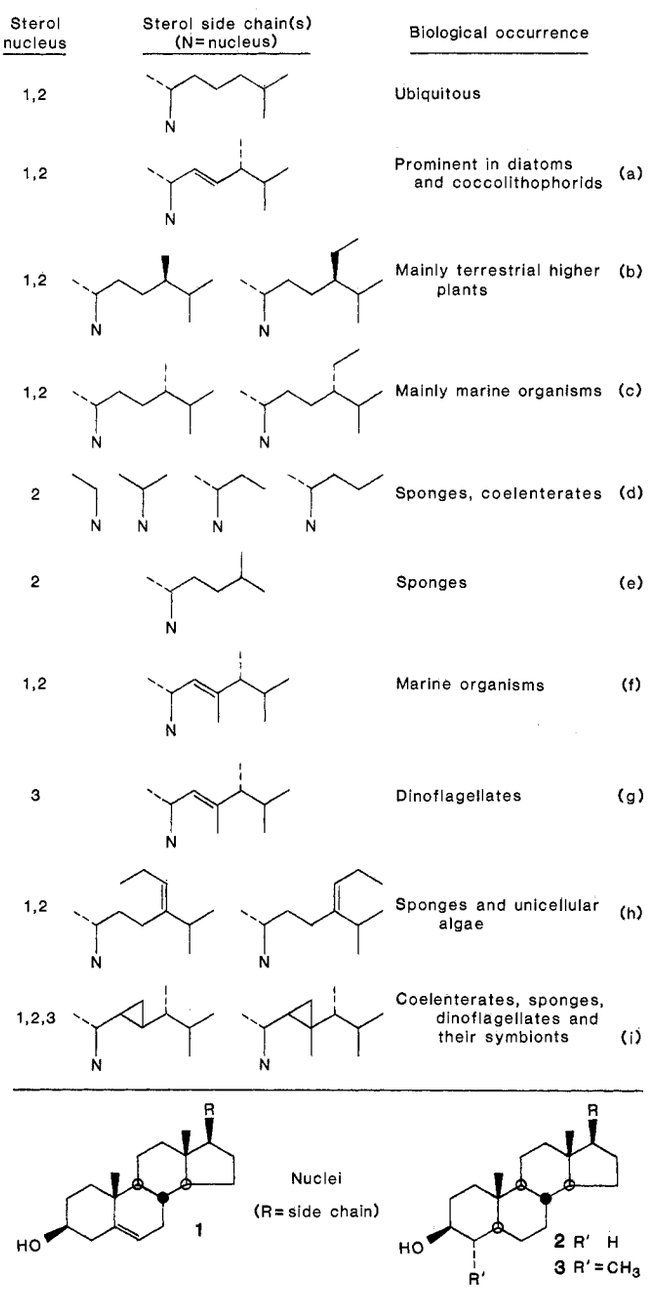


Fig. 4. Sterol types (nucleus and side chain) indicative of inputs from specific classes of organisms to sediments. All have been recognized in sediments (*18, 29–32, 116*). In some instances the assignment of sterols that may have originated from more than a single class of organism may be complemented by independent information, such as the abundance of sponge spicules in the sediments (*18, 44*) or of jellyfish (coelenterates) in the water column (*29*). (a) (*132*). (b) and (c) In terrestrial higher plants and marine organisms the stereochemistry of the sterol C-24 alkylation is dominantly β and α, respectively (*35*), although certain organisms do not conform to this pattern. (d) Of the variety of sterols with short ($< C_7$) side chains found in organisms (*133*), only the four shown have been identified in sediments to date (*18, 44*). (e) (*134*). (f) 23,24-Dimethyl alkylation appears to be restricted to marine organisms (*135*). (g) (*136*). (h) Sponges and unicellular algae seem more probable sources of these sterols in oceanic sediments than scallops or oysters (*33*). (i) 22,23-Methylene substitution now appears to be more widespread in organisms than was originally thought (*34, 45*).

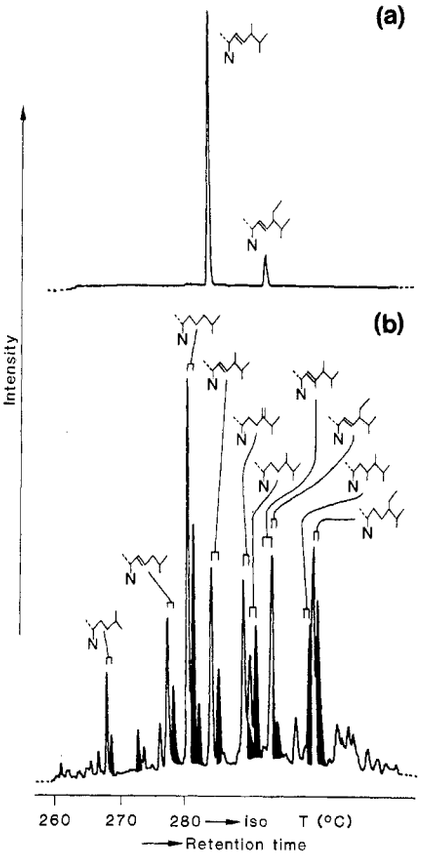


Fig. 5. Gas chromatograms of (a) the 4-desmethylsterols of the coccolithophore *Crystallolithus hyalinus* (*132*) and (b) the 4-desmethylsterol fraction of a Pleistocene diatomaceous ooze from the Japan Trench (*18*). (Gas chromatograph conditions: 20-m OV-1 flexible silica column, temperature programmed from 80° to 280°C at 5.8°C per minute after splitless injection at ambient temperature.) ■, 5α(H)-Stanols (**4a**); □, Δ^5-stenols (**1**); and *N*, nucleus.

facile isomerization at C-20, probably through a C-20 carbonium ion, then occurs to give equal proportions of 20*R* and 20*S* diastereoisomers (*61*).

Ring C monoaromatic steroid hydrocarbons occur in sediments and petroleums (*62, 63*) and include components with 10β,17β-dimethyl substitution (**10** in Fig. 6) (*64, 65*). These compounds appear as a mixture of isomers at C-20 [and presumably at C-24 (*66*)]. Since the methyl group is retained at C-10, it seems unlikely that they arise in sediments from diasterenes. Although no simulation studies with steradienes have been reported, it has been shown in the laboratory that under the acid conditions that rearrange Δ^5-sterenes to diasterenes, cholesta-3,5-diene can be converted to a mixture of hydrocarbons containing two C-ring monoaromatic steroids, thought to have the 5α(H) configuration and to be isomeric at C-20, as minor products (*65*). That steradienes are not found in other than highly immature sediments might be explained by their conversion into A- and C-ring monoaromatic steroids in early diagenesis. In addition, some circumstantial evidence suggests that the steratrienes found in relatively immature sediments (*18, 53*) may be intermediates in the formation of monoaromatic steroids, although this remains to be confirmed.

The final transformation considered here is the reduction of sterenes and diasterenes (Fig. 6). The appearance of steranes (**8a**) before diasteranes (**9**) during diagenesis (*67*) presumably reflects the ease of reduction of monounsaturated sterenes containing a di- or trisubstituted double bond compared with the tetrasubstituted double bond ($\Delta^{13(17)}$) in diasterenes. The steranes first generated are 5α(H) and 5β(H) components (**8a**), which retain the biosynthetic configurations at their other chiral centers (*68*). The diasteranes formed are principally 13β(H),17α(H) components (**9a**) with lesser amounts of 13α(H),17β(H) isomers (**9b**) (*69*); all the other centers have the configuration expected from rearrangement of the original biological skeleton. These alkanes often (*69*), but not always (*67*), inherit almost equal mixtures of the 20*R* and 20*S* configurations from the corresponding alkenes.

At the stage of maturity where diasteranes appear sterols can normally no longer be detected, and in slightly more mature sediments only trace amounts of sterones and sterenes survive. Hence, at the onset of late-stage diagenesis, the lipid extracts contain alkanes and monoaromatics as their major steroidal components.

The diagenetic fate of 4-methylsterols is less well understood. It appears that the 4α-methyl configuration dominates by far, both in organisms and in immature sediments (*18, 29, 30, 70, 71*). The apparent low ratio of 4-methylsterenes to sterenes in immature sediments (*18, 32*) is difficult to explain, in that certain 4α-methyl-5α(H)-stanols are often dominant components of marine sedimentary sterol distributions (*18, 29, 30, 32, 71*). This suggests that there are rate differences in the dehydration of sterols and 4-methylsterols to give the corresponding alkenes (*18, 32, 71, 72*). Yet at the stage of diasterene formation 4-methyldiasterenes, presumed to be solely 4α-methyl, are also generated (*70*), possibly indicating that any 4-methylsterenes, once formed, undergo rapid transformation to their rearranged counterparts (*18, 72*) or reduction to 4-methylsteranes (*70*). In the latter compounds the ratio of 4β- to 4α-methyl isomers is much higher than would be expected from the original sterol configuration (*70*). It has been suggested that this could arise from selectivity in the backbone rearrangement with 4β-methylsterenes not undergoing rearrangement due to steric hindrance (*70*). The 4-methyldiasterenes, like the diasterenes, are also reduced to the corresponding alkanes, and retain their preferred configuration at C-4 [presumably 4α-methyl (*69*)].

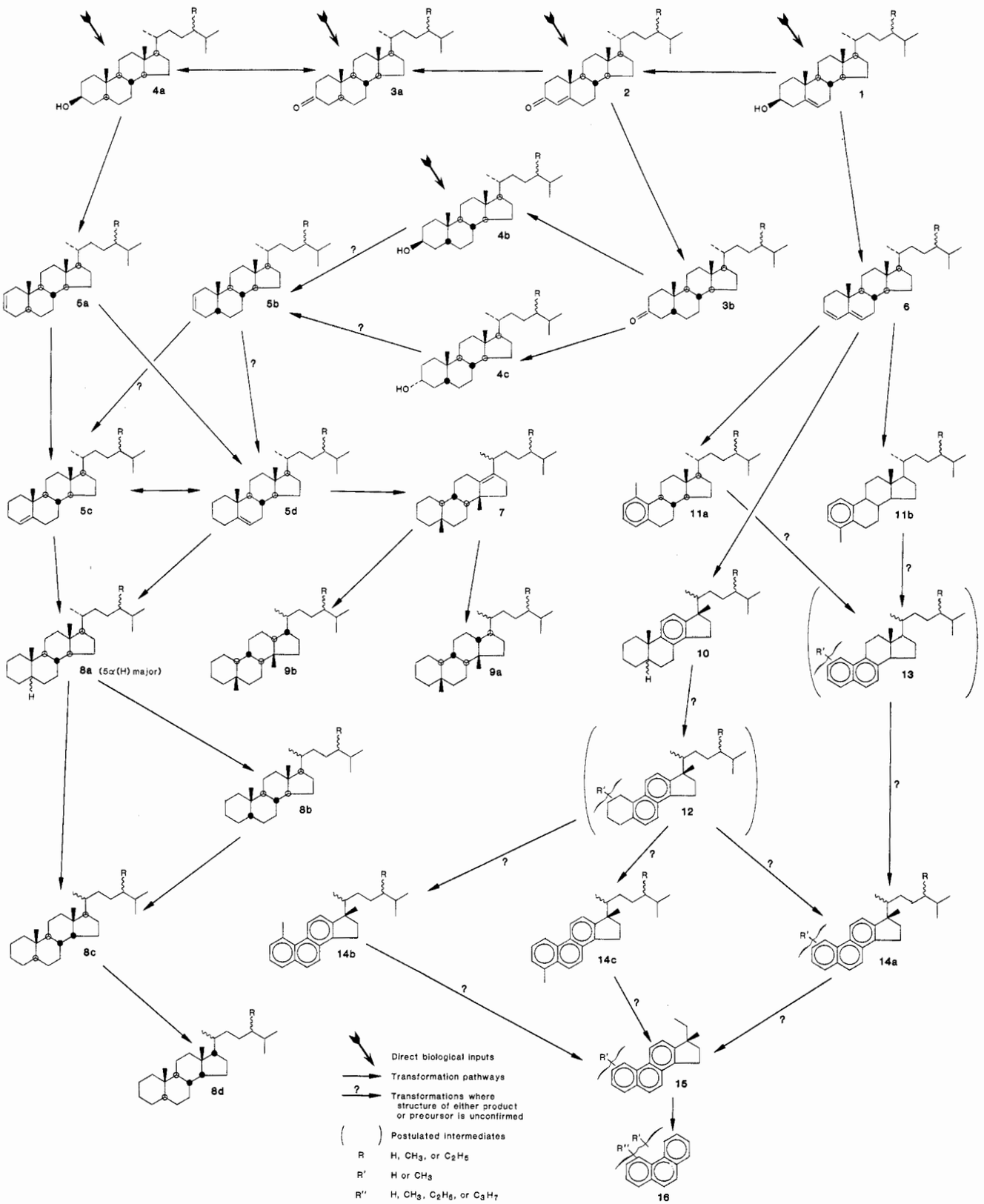
In this section changes in the nucleus have been emphasized without reference to the fate of unsaturated sterol side chains, despite the prominence of Δ^{22}- and $\Delta^{24(28)}$-sterols in bottom sediments (*18, 28–32, 41*). That such sterols undergo dehydration is shown by the occurrence of Δ^{22}- and $\Delta^{24(28)}$-steradienes in immature sediments (*73*), but their nuclear unsaturation appears to be at C-4 or C-5 rather than C-2 on mass spectral evidence (*73*). The subsequent fate of steradienes with side chain unsaturation is not clear; there are few reports of saturated steroidal nuclei with unsaturated side chains (*72*), and none of rearranged or C-ring aromatic steroidal skeletons. It seems probable that Δ^{22} and $\Delta^{24(28)}$ unsaturations are susceptible to diagenetic reduction or incorporation into kerogen.

Late-stage diagenesis and catagenesis—isomerization. For sterols to function as rigid inserts in cell membranes a certain degree of "molecular flatness" is required, which is achieved by an all-chair conformation with all the ring junctions fused in a *trans* configuration [that is, 8β(H),9α(H),10β(CH_3),13β-(CH_3),14α(H),17α(H), 20*R* (Fig. 7)]. Either the α(H) or the β(H) configuration is present at C-24 (Fig. 4). The processes of defunctionalization leave the biological configuration essentially intact in the nonrearranged steranes. With increased temperatures in the catagenetic zone, however, hydrogen exchange occurs by an unknown mechanism (*35, 62, 68, 74*). Sedimentary steranes tend to adopt the

Fig. 6. Schematic summary of proposed pathways of steroid diagenesis and catagenesis. The processes include defunctionalization, reduction, rearrangement, isomerization, and aromatization. Individual transformations are discussed and referenced in the text. Other steroids, such as Δ^3-sterenes (*29*) and steratrienes (*66*), are known constituents of sediments, but have been omitted because their place in the diagenetic scheme is not well understood. For simplicity, only direct biological inputs of sterols and sterones are considered (see Fig. 3). In a few instances the transformations indicated are well documented, but the majority can only be regarded as plausible on present evidence.

thermodynamically more stable structure and lose the original molecular flatness. At certain carbons (C-8 and C-9) the most stable configuration is already present, while that at C-10 and C-13 cannot be changed by hydrogen exchange (Fig. 7).

Immature sediments (maximum temperature <40°C) therefore contain steranes mainly with the "sterol configuration" (*35, 68*) with a mixture of isomers at C-5 resulting both indirectly from reduction during early diagenesis [mainly to the more stable 5α(H) configuration] and directly from the precursor stanols. Typically, the major nonrearranged steranes (up to 75 percent) of mature sediments and petroleums have, however, a 5α(H),14β(H),17β(H) configuration (**8d** in Fig. 6) (*62, 68, 75*) with the sterol-derived configuration preserved at the



other nuclear centers. The other nonrearranged steranes are mainly the "flat" sterol-derived 5α(H),14α(H), 17α(H) configuration (**8a**), although lesser amounts of other isomers are present (*62, 76, 77*). A mixture analogous to that in mature sediments and petroleums has been obtained by the isomerization of 5α(H)-cholestane on platinized carbon with H_2 at 300°C (*62, 78*). The sedimentary steranes with both 5α(H),14α(H), 17α(H) and 5α(H),14β(H),17β(H) configurations exist as isomeric mixtures at C-20 and probably also C-24 (*35, 66*). Thus isomerization at C-14, C-17, C-20, and C-24 has been proposed to occur with increasing catagenesis (*35, 62, 63*). In the Paris Basin Toarcian shales, isomerization at C-24 appears complete (50:50) in samples with a proposed maximum burial depth of less than 1000 meters (*79*). At the more sterically hindered C-20 position, a maximum burial depth of ~ 2500 m is required for complete isomerization (*68*). For the two centers (C-20 and C-24) in the side chain, where both isomers appear to have similar stability, it seems that the more hindered the center, the slower the isomerization rate (*68*).

Aromatization. Aromatic steroid hydrocarbons have been examined in a number of sedimentary sequences (*63, 65, 76, 80*), which contain a range of skeletal types, including two families [ABC-ring triaromatics, **14**, and the C-ring monoaromatics, **10** (*63, 65*)]. Changes in the relative abundances of these families with increasing maturation indicate that aromatization (C → ABC) of C-ring monoaromatics occurs in rings B and A and appears to proceed mainly with loss of the nuclear methyl group on the A-B ring junction. This process generates, in the case of the C_{27}–C_{29} 4-desmethyl skeletons, C_{26}–C_{28} compounds (**14a** in Fig. 6) with a 17β(CH_3) substituent (*63, 65*) as the major triaromatic products. Aromatization of the C-ring monoaromatics, with retention of the nuclear methyl group originally at the A-B junction, may also occur, the methyl group being transferred to ring A at positions corresponding to C-1 (**14c**) and C-4 (**14b**) in the original skeleton (*63*). Alternatively, or in addition, the ring A monoaromatic hydrocarbons (**11**) found in immature sediments with the angular methyl group originally at C-10 migrated to C-1 (**11a**) or C-4 (**11b**) may aromatize to form the same triaromatic components (**14c** and **14b**) (*58*). With increasing thermal maturation the C-ring monoaromatic hydrocarbons (**10**) appear to increase in abundance relative to the A-ring monoaromatic hydrocarbons (**11**) (*56, 58, 63*). The reasons for this phenomenon are not clear, but a more rapid conversion to the triaromatic steroid hydrocarbons (*58, 63, 65*) through progressive aromatization of rings A → AB → ABC as compared with C → CB → CBA could provide an explanation (Fig. 6). Unidentified AB- and BC-ring diaromatic components (**13** and **12**) have also been recognized (*56, 63, 81*) in smaller amounts than the C-ring monoaromatics (**10**) and triaromatics (**14a**). Aromatization of the B ring in both C-ring (**10**) and A-ring (**11**) monoaromatic steroid hydrocarbons appears, therefore, to be rapidly followed by aromatization of ring A and ring C, respectively.

Within the zone of late diagenesis and catagenesis the C-ring monoaromatic and the triaromatic hydrocarbons exist as diastereoisomers at C-20. They are also presumably a mixture of isomers at C-24 (*66*) as a result both of the configurations inherited from their precursor sterols and of configurational isomerization (compare the steranes) at this maturity stage. The C-ring monoaromatic distributions are further complicated by the existence of components also thought to be stereoisomeric at C-5 as a result of

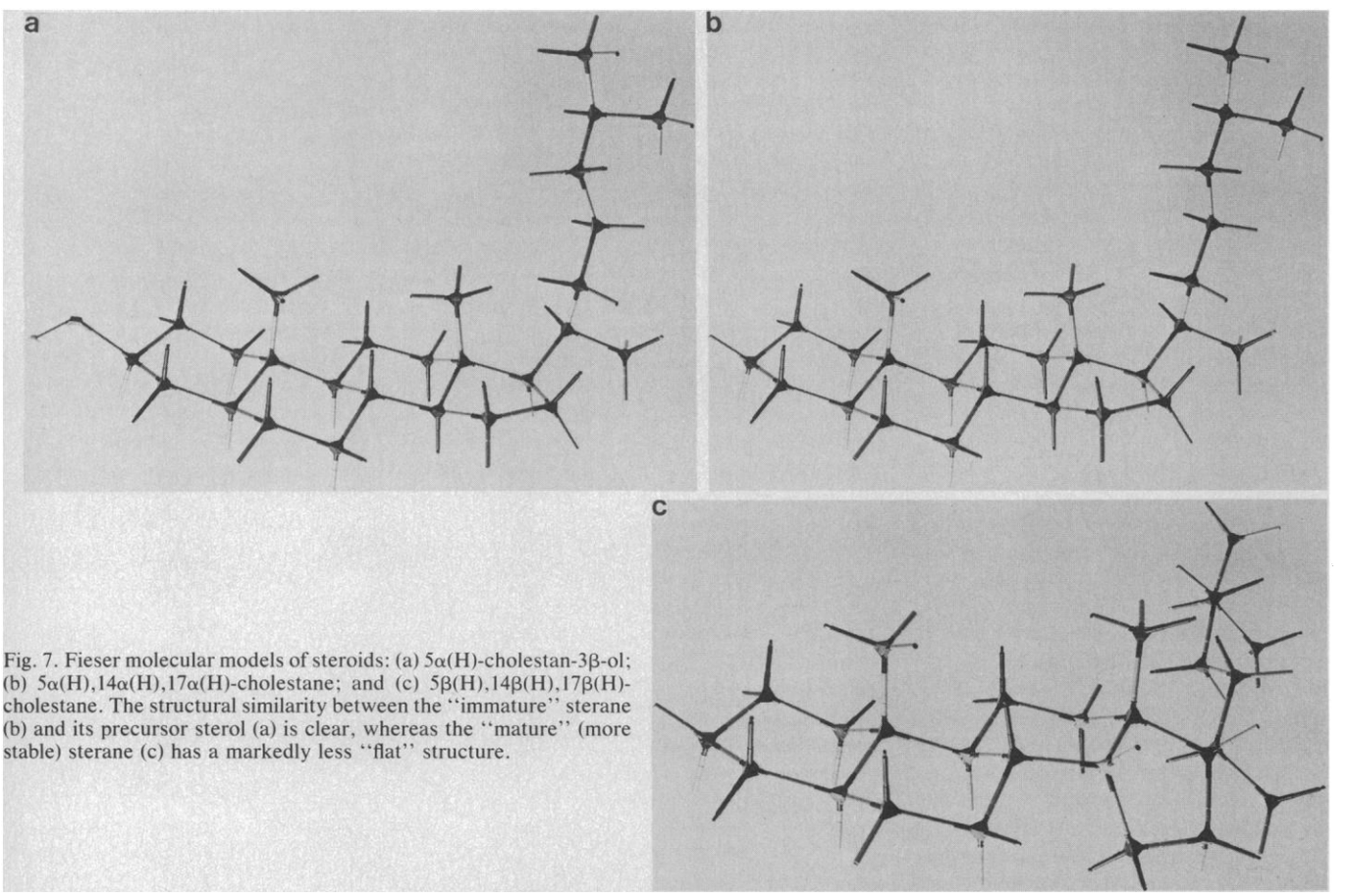


Fig. 7. Fieser molecular models of steroids: (a) 5α(H)-cholestan-3β-ol; (b) 5α(H),14α(H),17α(H)-cholestane; and (c) 5β(H),14β(H),17β(H)-cholestane. The structural similarity between the "immature" sterane (b) and its precursor sterol (a) is clear, whereas the "mature" (more stable) sterane (c) has a markedly less "flat" structure.

Table 1. Occurrences and relative abundance of steroids in organisms and in sediments of different maturities. ++, Major component; +, present in significant amount; Tr., trace component; −, absent.

Source	Sterols (1, 4)	Sterones (2, 3)	Sterenes (5, 6)	Mono-aromatic steroids (10, 11)	Diasterenes (7)	Steranes (8)	Diasteranes (9)	Poly-aromatic steroids (12–16)	Comments
Living organisms	++	+	−	−	−	−	−	−	Only biosynthetic compounds present
Water column and surface sediments	++	+	+	Tr.	Tr.	−	−	−	Steroid defunctionalization begins
Immature sediments	+	+	+	+	+	+	Tr.	−	Defunctionalization trends continue
Mature sediments and petroleums	−	−	−	+	−	++	+	++	Only alkane and aromatic components survive

isomerization in the late diagenesis–catagenesis zone.

With increased extent of catagenesis, an increase is observed in the relative abundance of monoaromatic and triaromatic steroid hydrocarbons with C_2 and C_3 side chains (**15**) relative to the corresponding components with C_8 to C_{10} side chains (**14**) (*62, 63*). These low molecular weight components could arise in part from alteration of sterols with short side chains (see above), or from thermal bond cleavage in the C_8 to C_{10} side chain of the higher molecular weight components. Further catagenetic bond cleavage in the side chain and the D ring may be responsible for some of the alkyl phenanthrenes (**16**) thought to occur in all crude petroleums (*82*) (Fig. 6).

Since the later stages of most of the reactions described in this section occur within the zone of oil generation (*76*), the contribution to the organic extract by the thermal breakdown of kerogen must be considered. Intact steroid skeletons are certainly present in kerogen (*83*), since laboratory pyrolysis has shown the presence of nonrearranged steranes in the pyrolysate, although no diasteranes or aromatic steroid hydrocarbons have been detected (*4, 62*). The latter are generated, however, when the products of heating are retained at the reaction site (*84*). The differences emphasize the care that must be taken in using the results of laboratory simulation experiments in the evaluation of sediments in the oil generation zone.

Summary of Diagenetic-Catagenetic Processes

During diagenesis and catagenesis the biolipids are altered in a systematic rather than a random way. Some information about their biological origin is lost during diagenesis, such as evidence of positions of unsaturation, but details like the carbon number distribution and structural features such as 4-methyl substitution and side chain alkylation patterns (for instance, differences between 23,24-dimethyl and 24-ethyl substitution) are retained until the later stages of catagenesis.

In essence, the changes involve transformation of biolipids with their enzymatically controlled skeletons into more stable geolipids, by a combination of processes such as defunctionalization, reduction, aromatization, structural rearrangement, and stereochemical isomerization. The relative abundances of the types of steroidal lipids in sediments of different maturity (Table 1) illustrate the effects of diagenesis and catagenesis, as biogenic sterols and sterones are converted into the steroidal hydrocarbons found in petroleums and mature sediments. The greatest diversity of steroids is found in immature sediments, where both residual biolipids and newly formed geolipids coexist.

All of the diagenetic and catagenetic changes outlined above (Fig. 6) are based on empirical evidence from observations of the occurrence of biolipids and geolipids in sediments of differing maturity, supported in part by radiolabeled incubation studies in the field (*41, 51*) and by laboratory heating experiments (*59, 85*). To better express these transformations there is a need for a more mathematically based approach to lipid diagenesis and catagenesis, similar to that invoked to elucidate early-stage inorganic diagenetic changes (*86*). The microbially mediated transformations of lipids may prove difficult to treat in this manner, with later stage diagenesis and catagenesis more amenable to such expression. Further understanding of the processes involved will undoubtedly come from the evaluation of their kinetics. In addition, the continuing study of new sedimentary environments and basins of deposition will help to clarify uncertain areas, such as the transformation pathways of 4-methylsteroids and of steroids with unsaturated side chains. It is important to recognize that the proclivity of specific reactions can differ between environments.

Applications

The contemporary position of chemical fossils can be compared to that existing during the early days of the study of conventional fossils, when description and documentation were essential preliminaries to the unifying theories of biological diversity and evolution and to the development of key tools such as biostratigraphy. In organic geochemistry a significant portion of the molecular fossil record can now be documented and exploited.

The detailed study of the structures of chemical fossils is a prerequisite to any assessment of their biological origins and of the diagenetic and catagenetic reactions they have undergone. This approach has much in common with the established field of natural product organic chemistry. It also has geochemical applications. At present, only a small proportion of the organic carbon content is amenable to precise molecular description. In contrast, the physical characterization of the total organic carbon content allows an averaged or statistical approach [for instance, (*2*)]. Both approaches can benefit from each other.

Correlation of chemical fossil data with aspects of environments of sediment deposition. It has become apparent that geolipids preserve a signature of depositional environments, reflecting the organisms extant within the system and the processes influencing the preservation of these lipids. The presence, absence, and relative abundances of several hundred frequently encountered compounds provide an approach to the characterization of environment and sedimentary processes and sediment deposition. One basis for paleoenviron-

mental assessment with lipid data relies on the structural relationships between bio- and geolipids. At early stages of diagenesis these relationships may be apparent, but they often become less evident as diagenesis progresses. From the sterol and sterene distributions of a diatomaceous ooze from the Japan Trench (Fig. 8), it can be seen that the side chain distributions of sterols are inherited by the resulting sterenes, but this conversion is only one step in a process of diagenetic and catagenetic transformation. In theory, if the components of an ancient sediment could be linked exactly to their precursors, a "lipid picture" comparable to that obtained for Recent sediments would be realized. This objective remains distant, despite the increasing ability to differentiate between present-day lipid facies. The oceanic organic geochemical record is often well preserved, however, especially in areas of low geothermal gradient. For example, in the Japan Trench many of the original marker compounds survive in Miocene sediments (~ 6 million years old) (*18*). Similarly, the lipids of an Albian black shale from the Hess Rise in the central North Pacific Ocean include many unaltered lipids (for instance, sterols) (*72*) that can be related to their presumed source organisms.

Attempts to relate the lipids of ancient sediments to present-day precursors of necessity assume that the principal biosynthetic pathways of the organisms that contributed to the sediment are comparable with those of modern biota. For example, the recognition of dinosterol in a Cretaceous black shale (*72*) confirms that it has been synthesized for at least 90 million years and perhaps indicates a dinoflagellate input (Fig. 4). Many other lipid components are believed to be uniquely synthesized by specific classes of organisms and can be used to identify contributions from particular organisms, such as methanogens (*87*). In addition, lipids can sometimes provide evidence of biological inputs that paleontological studies cannot give, such as an indication of contributions from coccolithophores to sediments that contain no skeletal debris because they were deposited below the carbonate compensation depth (*18, 88*). Chemical fossils can also provide evidence of microbial activity (*87, 89*), as bacteria can leave a signature in the form of specific molecules (*87, 89*), although they leave no morphological remains.

Overall, the use of markers that can be routinely recognized enables the assessment and distinction of lipid inputs from algae, bacteria, and terrestrial higher plants. In addition, particular components can be indicators for specific classes of organism (Fig. 4). The range of indicators extends, however, beyond sterols (Fig. 2); for example, certain cyclic diterpenoids and specific C_{40} acyclic isoprenoids are thought to be markers for terrestrial resinous plants and archaebacteria, respectively.

The use of triangular diagrams that describe the relative abundances of C_{27}, C_{28}, and C_{29} sterols (*90*) is of limited value in differentiating environments, except in extreme cases—for example, where there is a high abundance of C_{29} components with 24-ethyl–substituted side chains in sedimentary organic matter dominated by higher plant inputs. The range and complexity of the sterol distributions of marine organisms and sediments [for instance, see Fig. 5 and (*18, 29–32*)] indicate that this approach is too simplistic. A similar diagram of certain steranes (*80*) can, however, be useful in empirical geochemical correlations.

Evidence of other environmental features is more difficult to recognize, but the abundance of unsaturated fatty acids in a sediment core from Lake Biwa appears to be correlated with cooler climatic periods (*91*). Other lipids, such as long-chain alkenones from coccolithophores (*92*), may provide similar information.

The assessment of oxicity or anoxicity conditions in both water column and sediment from chemical fossils poses major problems (*93*). The presence of marker compounds for methanogenic (*87*) or sulfate-reducing (*89*) bacteria places oxicity constraints on sediments, but may reflect activity below, rather than at, the sediment-water interface. Similarly, the presence of sterols thought to derive from sponges (Fig. 4) (*18*) may imply bottom-water oxicity. At present, no chemical fossils that are definitive indicators of water column oxicity or anoxicity conditions are known, but in general markedly better preservation of labile lipids occurs in anoxic environments (*93*).

Other aspects that seem appropriate for organic geochemical studies are key event horizons in the geological record, such as boundaries where major evolutionary developments have occurred (for example, the Cretaceous-Tertiary boundary). It would be expected that such catastrophic changes in faunal and floral assemblages (*94*) may be reflected in the lipid compositions of sediments spanning the crucial interval. This possibility has yet to be explored.

Oil exploration. The distributions of chemical fossils in sedimentary rocks and petroleums reflect the environment of deposition and the extent and nature of thermally induced late diagenesis and catagenesis. They are therefore of obvi-

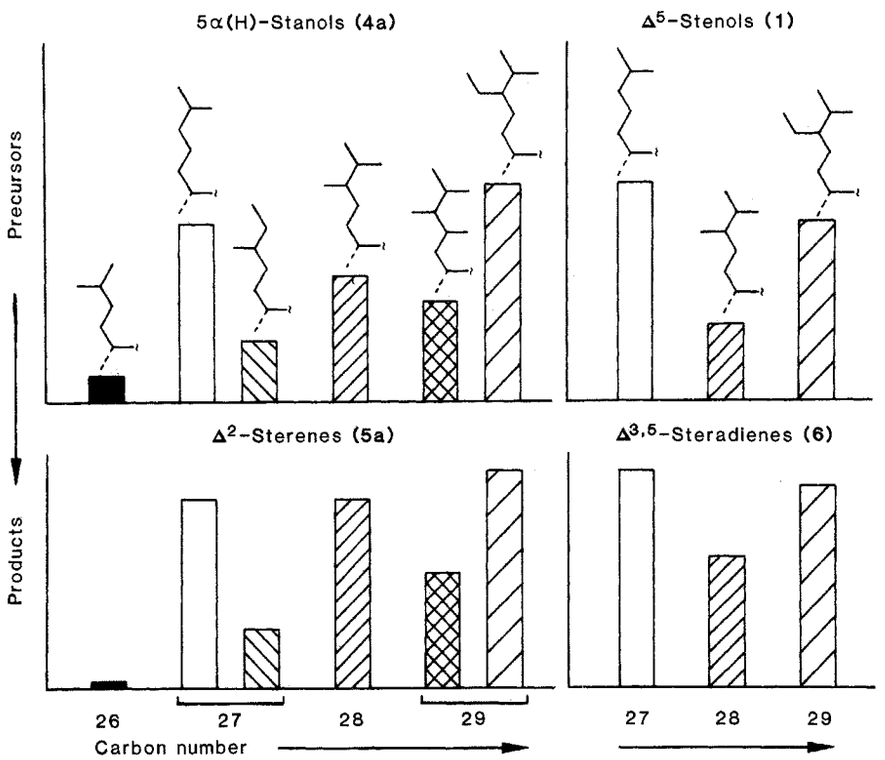


Fig. 8. Abundances of individual steroids in an extract of a Pleistocene diatomaceous ooze from the Japan Trench (DSDP 57-440B-3-5) (*18*). Comparison of the side chain distributions of 5α(H)-stanols (**4a**) with those of Δ^2-sterenes (**5a**) and of Δ^5-stenols (**1**) with those of $\Delta^{3,5}$-steradienes (**6**) suggest precursor-product relationships.

ous value in geochemical correlation studies. For oils, the effects of in-reservoir biodegradation and migration must also be understood.

The initial use of chemical fossils involved the correlation of oils and source rocks by simple pattern matching (*95*). For example, the *m/z* 218 fragmentogram (Fig. 9) of the alkanes from oil Yi-18 correlates with that for shale Yi-21, a slightly more mature equivalent of its suspected source rock. One would not expect the compound distributions in an oil to correlate with a suspected source rock if the two samples have a different depositional environment or a different level of maturity. The reasons for a poor correlation are clearly of interest to the petroleum geologist.

Recent advances in the understanding of the fate of steroids during late diagenesis and catagenesis have suggested measurements of the ratios of selected products and precursors as a means of assessing the extent of thermal maturation (*63, 65, 68, 96*). This approach has been applied to several types of compound. The ranges over which the reactions occur in two lower Jurassic sequences in relation to the zone of oil generation and vitrinite reflections are shown in Fig. 10 (*68, 74, 96*). Certain reactions occur over small ranges of maturation, whereas others extend over larger ranges and together cover the zone of oil generation and above. Some of them can also be applied to the maturity ranking of crude oils. Steroid hydrocarbons appear at present to be the most useful compounds for assessing the extent of maturation.

In Fig. 9 it is apparent that maturity effects are responsible for the lack of correlation between oil Cheng-15 and shale Yi-21. The oil is more mature as it contains higher relative proportions of the products of geological isomerization, namely, 20*R*- and 20*S*-5α(H),14β(H), 17β(H)-steranes and 20*S*-5α(H),14α(H), 17α(H)-steranes. This maturity difference is in agreement with other measurements (*80*).

In applying chemical fossils to petroleum geochemistry an attempt should also be made to distinguish the results of maturation in the source rock from those in the reservoir and hence to evaluate the relative extent of these processes. This will involve a search for reactions that (i) effectively cease when oil moves out of the source rock and becomes a fluid in the reservoir (even when source rock and reservoir are at similar temperatures) and (ii) continue in both mediums, possibly at different rates.

Recognition of a lack of correlation between an oil and a proposed source

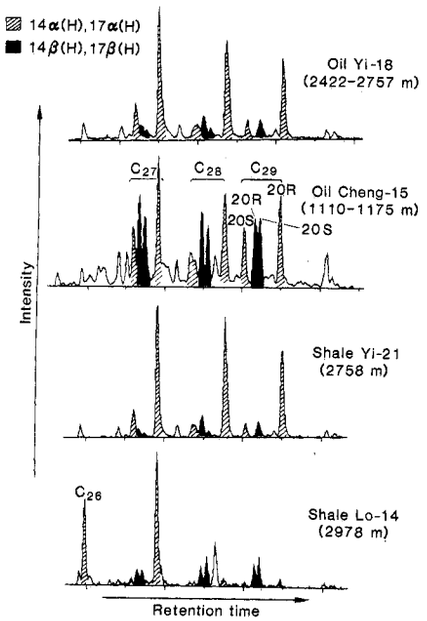


Fig. 9. Comparison of two oils and two shales from the Zhanahua Basin, China (*80*), as illustrated by fragmentograms of the ion with mass-to-charge ratio (*m/z*) 218 [a prominent ion of sterane mass spectra, particularly 14β(H)-steranes (*62, 68, 76, 96, 137*)]. Carbon numbers and stereochemistry at C-20 for C_{29} components are shown for oil Cheng-15.

rock, two oils, or two sediments resulting from differences in depositional environment is more difficult. Much of the original molecular information disappears with thermal maturation. One useful measurement is the carbon number distribution of steranes (*90*). Sterols are mainly C_{27} to C_{29} components and petroleum and sedimentary steroid hydrocarbons are also dominated by this carbon number range (*97*). Triangular diagrams for steranes (C_{27} to C_{29}) with the same stereochemistry [for instance, 5α(H), 14α(H),17α(H),20*R* or *S*] can indicate relationships between samples (*96*). Thus, in Fig. 9 the carbon number distribution of these components in oil Cheng-15 is similar to that in shale Yi-21, perhaps reflecting similarities in their depositional environments, but differs from that in shale Lo-14, reflecting differences in input at the time of deposition. It is not yet possible to infer detailed environmental information by examination of the chemical fossils in a mature source rock, although certain measurements (for instance, ratios of skeletal types such as hopanoids to steroids and 4-methyl steroids to 4-desmethyl steroids) may be of value. For example, source rocks and crude oils dominated by higher plant inputs can have large amounts of C_{29} relative to C_{26}–C_{28} steroid alkanes, together with high hopanoid/steroid ratios (> 5). Assessment of such aspects of the depositional environment must proceed with caution, especially since less than 5 percent of the total organic carbon is used in the case of source rocks, and the marker components are often minor ones in both source rocks and petroleums. It is, however, these structurally specific compounds that show promise of developing a maturity-independent molecular record of original biological input.

There is a problem of the possible effect of migration on the distributions of chemical fossils in petroleums, particularly since the mechanisms and routes of migration are poorly understood (*98*). Chromatography during migration has been proposed as the cause of small changes in sterane stereoisomer ratios, as well as in the relative abundances of other alkanes (*75, 99*). For certain oils with comparatively well understood migration histories (*100*), variations with extent of migration are inferred from

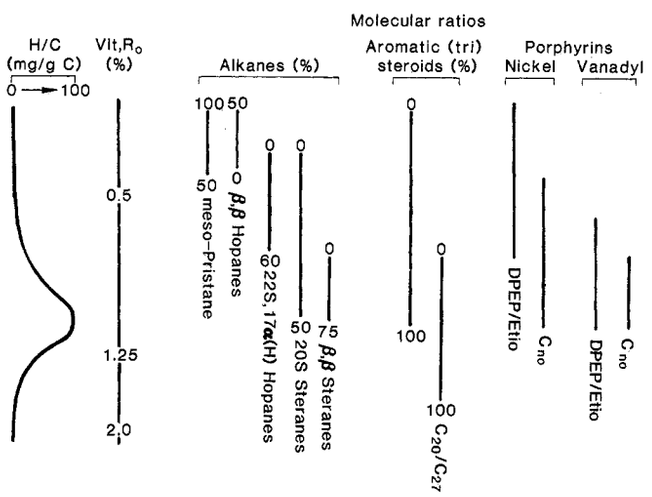


Fig. 10. Ranges of individual molecular measurements for thermal maturation, including those for steroids, plotted against the downhole hydrocarbon generation curve and vitrinite reflectance (R_0) values (*96*). The figure is compiled from results for the Toarcian shales of the Paris Basin and for the Pliensbachian shales of northwest Germany (*96*). *DPEP* and *Etio* refer to desoxyphylloerythroetioporphyrin-type and etiotype porphyrns, respectively; C_{no} denotes carbon numbers.

Table 2. Estimated utility of various compound types in geochemical applications. The estimates are based on the potential of the compound type as reported in the literature: +, of limited use; ++, useful; +++, highly useful; and ?, uncertain.

Compound type	Carbon number range	Depositional environment*	Burial history†	Oil biodegradation‡, (§)
Straight chain	12 to 40	+	++	+++ (+)
Branched chain	14 to 33	+	+	++ (+)
Acyclic isoprenoid	14 to 40	++	++	+++ (++)
Diterpenoid	19 to 30	++	+	+++ (+++)
Steroid	19 to 31	+++	+++	+ (+++)
Triterpenoid	24 to 40	++	+++	+ (+++)
Porphyrin	26 to 40	++	++	? (?)

*Assessment of inputs and conditions of sediment deposition (*18, 19, 27, 42, 71, 87, 90, 92, 112*). †Assessment of diagenesis and catagenesis (*62, 63, 76, 96, 113*). ‡Assessment of extent of degradation (*80, 101, 102, 113*). §Assessment of value in correlation of biodegraded samples (*80, 101, 102, 113*).

ratios of components with extensive differences in molecular shape, size, and polarity (such as mono- and triaromatic steroid hydrocarbons, or alkanes and aromatic hydrocarbons). In this case, however, the alkane stereoisomer ratios were not affected. All of the measurements proposed to change with extent of migration in Alaskan North Slope crudes (*99*) also vary with maturity. No measurement based on the relative abundances of chemical fossils has been proved to assess the extent of migration while being independent of both maturity and depositional environment. Another factor that influences crude oil composition is biodegradation in the reservoir (*101*). However, the steroid hydrocarbons and hopanoid triterpanes appear to be attacked only in extreme cases (*62*).

Environmental. Distributions of chemical fossils are being increasingly used to identify sources of oil pollution as they impart a "fingerprint" to a crude oil. Three of the main processes that affect the composition of oil after spillage are photooxidation, evaporation of volatile components, and bacterial degradation (*102*). Steranes (and hopanoid triterpanes) are, however, relatively involatile and are not as readily biodegraded as many other types of alkane, so they are relatively resistant to these weathering processes. Indeed, the distributions of these compounds obtained by GC-MS provide one of the most efficient methods of correlating weathered oil spillages with their presumed sources (*103*), just as they are widely used to correlate in-reservoir biodegraded petroleums with their nonbiodegraded counterparts (*80*). In adopting this approach to assign pollution sources in present-day sediments, care must be taken, for example, to discriminate against contributions from the reworking of mature, organic-rich shales, which can give a false indication of the level of modern pollution. Similarly, the maturity of sedimentary rocks that have high contents of reworked organic matter or contain migrated hydrocarbons can appear anomalously high (*68, 104*). Such effects can sometimes be recognized from the co-occurrence of compounds that would not normally coexist at the same level of maturity.

Mechanisms and Kinetics

The reactions that generate the changes plotted in Fig. 10 cover different maturity ranges, reflecting different kinetic parameters. Since different reaction types, such as configuration isomerization and aromatization, might be expected to have significantly different kinetic constants (different temperature dependences), the relationships shown in Fig. 10 cannot be considered general. A value that measures the extent of configurational isomerization in an alkane might be expected to correspond to that value for the same type of reaction at another chiral center, because of similarities in their kinetic constants and temperature dependences. For reactions of different types, however, it would be expected that the reaction whose rate increases more rapidly with temperature will be promoted when the sedimentary heating rate [or average rate of increase in temperature since deposition (*105*)] is higher. It has been proposed that isomerization at C-20 in 5α(H),14α(H), 17α(H)-steranes has a lower activation energy than aromatization of C-ring monoaromatic steroid hydrocarbons to triaromatic compounds (*65*). Aromatization is thought to be accelerated relative to isomerization in sedimentary sequences with high average heating rates (> 10°C per million years) compared to those with low average heating rates [< 1°C per million years (*65*)]. Plots of extent of isomerization against extent of aromatization for depth sequences of sediment samples show differences between sedimentary basins with different average heating rates (*65*). A less empirical approach to thermal history will require physicochemical data for the reactions, including kinetic constants (preexponential factor and activation energy). These data should be derived (i) from laboratory heating experiments and (ii) by calculating rate constants for various sediment sequences and integrating them into mathematical geophysical models that attempt to predict the temporal variation in sedimentary heating rates (*106*) since deposition; for a given sequence the reaction extents can then be varied, by altering the estimates for the constants, until they fit the observed values of aromatization and isomerization. These approaches should assist in both elucidating likely reaction mechanisms and providing methodology for determining average sedimentary heating rates. They may eventually be applied in dividing thermal history into a series of events and calculating maximum paleotemperatures. Such research may provide a means of understanding earth processes in the temperature range 50° to 250°C.

A detailed evaluation of the variation in catalytic activity caused by changes in lithology and of the effect of pressure (*107*) will be possible only when appropriate estimates for the kinetic constants involved have been obtained. Competing reactions confuse present studies, while lithological effects include changes in heat conduction, access of fluids, and catalytic activity.

Conclusions

Steroids illustrate the principles and applications of molecular organic geochemistry. There are many uncertainties, but these compounds are at present the most clearly understood and useful. The geological fates of other groups of compounds are under study; Table 2 summarizes the utility of some of these types and their geological products. Examples (Fig. 2) include porphyrins, derived from the chlorin moiety of the chlorophylls, and acyclic isoprenoid compounds, derived from the corresponding phytyl side chain (*93, 108*) and from the lipids of archaebacteria (*109*). There are many triterpenoids and diterpenoids of diverse origins (*110*), but the ubiquitous hopane triterpenoids deserve special mention. Their finding in nearly all geological samples stimulated a fresh appraisal of their role in past and present biospheres (*111*), illustrating the wider insight that chemical fossils can offer.

The scope of molecular organic geochemistry is multidisciplinary. It is based on compound identification and relies for its continued development on synthetic and analytical skills. Knowledge of the pathways and reactions that link chemical fossil molecules to their precursor natural products is now at a sufficient level that integration with geological and geophysical ideas about earth processes can be fruitful. Studies of the slow reactions involved require determination of the kinetic parameters and underlying mechanisms. Investigation of the water column and the shallow zone of early diagenesis requires an appreciation of microbiological constraints, possibly with the help of laboratory cultures. The study of early diagenetic reactions can be aided by isotopic labeling experiments, in a manner similar to their use by biochemists attempting to elucidate biosynthetic pathways.

The reconstruction of postdepositional conditions must necessarily stem from an understanding of present-day environments, integrated with oceanographic and biological studies. Account must also be taken of paleoecological and sedimentological models in assessing environments on the basis of organic geochemical criteria.

References and Notes

1. G. Eglinton and M. Calvin, *Sci. Am.* **216** (No. 1), 32 (1967).
2. B. Durand, Ed., *Kerogen. Insoluble Organic Matter from Sedimentary Rocks* (Editions Technip, Paris, 1980).
3. D. van de Meent, S. C. Brown, R. P. Philp, B. R. T. Simoneit, *Geochim. Cosmochim. Acta* **44**, 999 (1980); W. L. Maters, D. van de Meent, P. J. W. Schuyl, J. W. de Leeuw, P. A. Schenck, H. L. C. Meuzelaar, in *Proceedings, 3rd International Symposium on Analytical Pyrolysis*, C. E. R. Jones and C. A. Cramers, Eds. (Elsevier, Amsterdam, 1977), p. 203; S. R. Larter and A. G. Douglas, in *Advances in Organic Geochemistry 1979*, A. G. Douglas and J. R. Maxwell, Eds. (Pergamon, Oxford, 1980), p. 579.
4. W. K. Seifert, *Geochim. Cosmochim. Acta* **42**, 473 (1978).
5. S. Brown, T. A. Baillie, A. L. Burlingame, in *Advances in Organic Geochemistry 1979*, A. G. Douglas and J. R. Maxwell, Eds. (Pergamon, Oxford, 1980), p. 475.
6. W. Michaelis and P. Albrecht, *Naturwissenschaften* **66**, 420 (1979); B. Chappe, W. Michaelis, P. Albrecht, G. Ourisson, *ibid.*, p. 522; B. Chappe, W. Michaelis, P. Albrecht, in *Advances in Organic Geochemistry 1979*, A. G. Douglas and J. R. Maxwell, Eds. (Pergamon, Oxford, 1980), p. 265; W. Michaelis, B. Chappe, P. Albrecht, in *Advances in Organic Geochemistry 1981*, M. Bjorøy *et al.*, Eds. (Heyden, London, in press).
7. C. Engler, *Z. Angew. Chem.* **21**, 1585 (1909).
8. P. Walden, *Chem. Ztg.* **30**, 291 (1906).
9. M. Steinkopf, A. K. Koss, S. Liebmann, *ibid.* **36**, 653 (1912); M. A. Racusin, *Chem. Abstr.* **1**, 1185 (1907); A. K. Koss, *ibid.* **6**, 291 (1912).
10. O. Schreiner and E. C. Shorey, *J. Am. Chem. Soc.* **31**, 116 (1909); *J. Biol. Chem.* **9**, 9 (1911).
11. M. Steibl and V. Herout, in *Organic Geochemistry. Methods and Results*, G. Eglinton and M. T. J. Murphy, Eds. (Springer-Verlag, Berlin, 1969), p. 401.
12. I. M. Heilbrom, J. C. E. Simpson, F. S. Spring, *J. Chem. Soc.* (1933), p. 626.
13. W. Steinkopf, H. Winterwitz, W. Roederer, A. Wolynski, *J. Prakt. Chem.* **100**, 65 (1919); S. Kawai and S. Kobayashi, *Chem. Ind. (Tokyo)* **26**, 1036 (1923); N. D. Zelinskii, *Chem. Ber.* **60B**, 1793 (1927).
14. J. McLean, G. H. Rettie, F. S. Spring, *Chem. Ind. (London)* (1958), p. 1515.
15. W. G. Meinschein and G. S. Kenny, *Anal. Chem.* **29**, 1153 (1957).
16. Use of amino acids in geochronology was first suggested by P. E. Hare and P. H. Abelson, [*Carnegie Inst. Washington Yearbk.* **66**, 526 (1969)]. For reviews, see G. Dungworth, *Chem. Geol.* **17**, 135 (1976); R. A. Schroeder and J. L. Bada, *Earth Sci. Rev.* **12**, 347 (1976); K. M. Williams and G. G. Smith, *Origins Life* **8**, 91 (1977).
17. The existence of significant variations in the lipid compositions of different organisms has been recognized for many years [for instance, T. P. Hilditch, *The Chemical Constitution of Natural Fats* (Chapman & Hall, London, 1940)] and forms a basis for chemotaxonomic studies.
18. S. C. Brassell, thesis, University of Bristol (1980); S. C. Brassell *et al.*, *Init. Rep. Deep Sea Drill. Proj.* **56/57** (part 2), 1367 (1980); in *Advances in Organic Geochemistry 1979*, A. G. Douglas and J. R. Maxwell, Eds. (Pergamon, Oxford, 1980), p. 375.
19. B. R. T. Simoneit, *Chem. Oceanogr.* **7**, 233 (1978).
20. E. T. Degens and D. A. Ross, *The Black Sea—Geology, Chemistry and Biology* (American Association of Petroleum Geologists, Tulsa, Okla., 1974).
21. An example is the almost pure "Braarudosphaera chalk" found at DSDP site 362 on the Walvis Ridge [*Int. Rep. Deep Sea Drill. Proj.* **40**, 183 (1978)].
22. W. G. Deuser, *Deep Sea Res.* **18**, 995 (1971).
23. H.-J. Schrader, *Science* **174**, 55 (1971); S. Honjo and M. R. Roman, *J. Mar. Res.* **36**, 45 (1978).
24. P. K. Weyl, *Oceanography—An Introduction to the Marine Environment* (Wiley, New York, 1970); G. P. Allen and P. Castaing, *Mar. Geol.* **14**, M47 (1973).
25. B. R. T. Simoneit and G. Eglinton, in *Advances in Organic Geochemistry 1975*, R. Campos and J. Goñi, Eds. (Enadimsa, Madrid, 1977), p. 415; B. R. T. Simoneit, R. Chester, G. Eglinton, *Nature (London)* **267**, 682 (1977).
26. F. P. Shepard and N. F. Marshall, *Bull. Am. Assoc. Pet. Geol.* **57**, 244 (1973).
27. W. Y. Huang and W. G. Meinschein, *Geochim. Cosmochim. Acta* **40**, 323 (1976).
28. C. Lee, R. B. Gagosian, J. W. Farrington, *ibid.* **41**, 985 (1977).
29. A. M. K. Wardroper, J. R. Maxwell, R. J. Morris, *Steroids* **32**, 203 (1978); A. M. K. Wardroper, thesis, University of Bristol (1979).
30. J. J. Boon, W. I. C. Rijpstra, F. De Lange, J. W. de Leeuw, M. Yoshioka, Y. Shimizu, *Nature (London)* **277**, 125 (1979).
31. C. Lee, J. W. Farrington, R. B. Gagosian, *Geochim. Cosmochim. Acta* **43**, 35 (1979).
32. R. B. Gagosian, S. O. Smith, C. Lee, J. W. Farrington, N. M. Frew, in *Advances in Organic Geochemistry 1979*, A. G. Douglas and J. R. Maxwell, Eds. (Pergamon, Oxford, 1980), p. 407.
33. 24-Propylidene sterols have been recognized in sponges [Y. M. Sheikh and C. Djerassi, *Tetrahedron* **30**, 4095 (1974)], unicellular algae [M. Rohmer, W. C. M. C. Kokke, W. Fenical, C. Djerassi, *Steroids* **35**, 219 (1980)], scallops [D. R. Idler, L. M. Safe, E. F. Macdonald, *ibid.* **18**, 545 (1971); D. R. Idler, M. W. Khalil, J. D. Gilbert, C. J. W. Brooks, *ibid.* **27**, 155 (1976); M. Kobayashi and M. Mitsuhashi, *ibid.* **26**, 605 (1975)], and oysters [S. Teshima, G. W. Patterson, S. R. Dutky, *Lipids* **15**, 1004 (1980)].
34. L. S. Ciereszko, M. A. Johnson, R. W. Schmidt, C. B. Koons, *Comp. Biochem. Physiol.* **24**, 899 (1968); F. J. Schmitz and T. Pattabhiramon, *J. Am. Chem. Soc.* **92**, 6073 (1970); A. Kanazawa, S. Teshima, T. Ando, *Comp. Biochem. Physiol. B* **57**, 317 (1977); P. A. Stendler, F. J. Schmitz, L. S. Ciereszko, *ibid.* **56**, 385 (1977); C. Delseth, Y. Kashman, C. Djerassi, *Helv. Chim. Acta* **62**, 2037 (1979); N. W. Winters, W. C. M. C. Kokke, M. Rohmer, W. H. Fenical, C. Djerassi, *Tetrahedron Lett.* (1979), p. 3065.
35. L. J. Mulheirn and G. Ryback, *Nature (London)* **256**, 301 (1975); L. J. Goad, J. R. Lenton, F. F. Knapp, T. W. Goodwin, *Lipids* **9**, 582 (1974).
36. S. Teshima, *Mem. Fac. Fish. Kagashima Univ.* **21**, 69 (1972).
37. G. W. Patterson, *Lipids* **6**, 120 (1971); P. W. Kirk and P. Catalfomo, *Phytochemistry* **9**, 595 (1970); S. Teshima and A. Kanazawa, *Bull. Jpn. Soc. Sci. Fish.* **37**, 68 (1971); W. R. Nes and M. L. McKean, *Biochemistry of Steroids and Other Isopentenoids* (University Park Press, Baltimore, 1977).
38. J. A. Ballantine, A. Lavis, R. J. Morris, *Phytochemistry* **18**, 1459 (1979).
39. D. J. Faulkner and R. J. Andersen, in *The Sea*, E. D. Goldberg, Ed. (Interscience, New York, 1974), vol. 5, p. 679.
40. N. W. Withers, R. C. Tuttle, G. G. Holz, D. H. Beach, L. J. Goad, T. W. Goodwin, *Phytochemistry* **17**, 1987 (1978).
41. S. J. Gaskell and G. Eglinton, *Geochim. Cosmochim. Acta* **40**, 1221 (1976); in *Advances in Organic Geochemistry 1973*, B. Tissot and F. Bienner, Eds. (Editions Technip, Paris, 1974), p. 963; *Nature (London)* **254**, 209 (1975); M. Nishimura and T. Koyama, *Geochim. Cosmochim. Acta* **41**, 379 (1977); M. Nishimura, *ibid.*, p. 1817.
42. M. Nishimura, *Geochim. Cosmochim. Acta* **42**, 349 (1978).
43. P. A. Cranwell, *ibid.*, p. 1523.
44. S. C. Brassell and G. Eglinton, *Nature (London)* **290**, 579 (1981).
45. C. Djerassi, N. Theobald, W. C. M. C. Kokke, C. S. Pak, R. M. K. Carlson, *Pure Appl. Chem.* **51**, 1815 (1979).
46. B. P. Tissot and D. H. Welte, *Petroleum Formation and Occurrence* (Springer-Verlag, Berlin, 1978).
47. J. M. Schmitter, Z. Vatjta, P. J. Arpino, in *Advances in Organic Geochemistry 1979*, A. G. Douglas and J. R. Maxwell, Eds. (Pergamon, Oxford, 1980), p. 67.
48. Steroid acids occur in plants [N. Mandava, J. D. Anderson, S. R. Dutky, M. J. Thompson, *Steroids* **23**, 357 (1974)] and are intermediates in bacterial degradation of cholest-5-en-3β-ol [W. Charney and H. L. Herzog, *Microbial Transformation of Steroids* (Academic Press, New York, 1967)].
49. D. M. Orcutt and B. Richardson, *Steroids* **16**, 429 (1970); E. I. Mercer, R. A. London, I. S. A. Kent, A. J. Taylor, *Phytochemistry* **13**, 845 (1974); P. A. Cranwell and J. K. Volkman, *Chem. Geol.* **32**, 29 (1981).
50. J. J. Boon and J. W. de Leeuw, *Mar. Chem.* **7**, 117 (1979).
51. K. L. H. Edmunds, S. C. Brassell, G. Eglinton, in *Advances in Organic Geochemistry 1979*, A. G. Douglas and J. R. Maxwell, Eds. (Pergamon, Oxford, 1980), p. 427.
52. M. M. Quirk, A. M. K. Wardroper, P. W. Brooks, R. E. Wheatley, J. R. Maxwell, in *Biogéochemie de la Matière Organique à l'Interface Eau-Sédiment Marin*, R. Daumas, Ed. (Editions du CNRS, Paris, 1980), p. 225.
53. R. B. Gagosian and J. W. Farrington, *Geochim. Cosmochim. Acta* **42**, 1091 (1978); M. Dastillung and P. Albrecht, *Nature (London)* **269**, 678 (1977).
54. S. G. Wakeham *et al.*, *Nature (London)* **286**, 798 (1980).
55. R. J. Hamilton, M. Y. Raie, I. Weatherston, C. J. Brooks, J. H. Borthwick, *J. Chem. Soc. Perkin Trans. 1* (1975), p. 354.
56. C. Spyckerelle, thesis, University of Strasbourg (1975).
57. J. Rullkötter, H. von der Dick, D. H. Welte, *Init. Rep. Deep Sea Drill. Proj.*, in press.
58. G. Hussler, B. Chappe, P. Wehrung, P. Albrecht, *Nature (London)* **294**, 556 (1981).
59. I. Rubinstein, O. Sieskind, P. Albrecht, *J. Chem. Soc. Perkin Trans. 1* (1975), p. 1833.
60. O. Sieskind, G. Joly, P. Albrecht, *Geochim. Cosmochim. Acta* **43**, 1675 (1979).
61. J. W. Blunt, M. P. Hartshorn, D. N. Kirk, *Tetrahedron* **22**, 3195 (1966); *ibid.* **25**, 149 (1969); D. N. Kirk and P. M. Shaw, *Chem. Commun.* (1970), p. 806.
62. W. K. Seifert and J. M. Moldowan, *Geochim. Cosmochim. Acta* **42**, 77 (1978).
63. A. S. Mackenzie, C. F. Hoffmann, J. R. Maxwell, *ibid.* **45**, 1345 (1981).
64. That C-ring monoaromatic components occur in geological materials with a 10β,17β-dimethyl skeleton was first demonstrated by comparison with synthetic compounds [B. Ludwig, G. Hussler, P. Wehrung, P. Albrecht, in *Advances in Organic Geochemistry 1981*, M. Bjorøy *et al.*, Eds. (Heyden, London, in press)]. Compounds with the same skeleton have been formed as products of the treatment of 5α(H)-cholestane with Pd/C at 300°C and their occurrence in sedimentary rock and petroleums confirmed (W. K. Seifert, R. M. Carlson, J. M. Moldowan, in *ibid.*).
65. A. S. Mackenzie, N. A. Lamb, J. R. Maxwell, *Nature (London)* **295**, 223 (1982).
66. J. R. Maxwell, A. S. Mackenzie, J. R. Volkman, *ibid.* **286**, 694 (1980).
67. S. C. Brassell *et al.*, *Init. Rep. Deep Sea Drill. Proj.* **50**, 647 (1980).
68. A. S. Mackenzie, R. L. Patience, J. R. Max-

well, M. Vandenbroucke, B. Durand, *Geochim. Cosmochim. Acta* **44**, 1709 (1980).
69. A. Ensminger, G. Joly, P. Albrecht, *Tetrahedron Lett.* (1978), p. 1575.
70. I. Rubinstein and P. Albrecht, *Chem. Commun.* (1975), p. 957.
71. I. D. Thomson, S. C. Brassell, G. Eglinton, J. R. Maxwell, *Init. Rep. Deep Sea Drill. Proj.*, in press; S. C. Brassell, G. Eglinton, J. R. Maxwell, *ibid.* **66**, 557 (1982).
72. P. A. Comet, J. McEvoy, S. C. Brassell, G. Eglinton, J. R. Maxwell, I. D. Thomson, *ibid.* **62**, 923 (1981).
73. $\Delta^{4,22}$- and $\Delta^{5,22}$-steradienes have been identified, on the basis of mass spectral interpretation, in a number of DSDP sediments [C. Cornford, J. Rullkötter, D. Welte, *ibid.* **47** (part 1), 511 (1979); J. Rullkötter, C. Cornford, P. Flekken, D. H. Welte, *ibid.* **56/57** (part 2), 1291 (1980); (*57*)] and in immature horizons of the Miocene Monterey Formation [W. Giger and C. Schaffner, *Naturwissenschaften* **68**, 37 (1981)]. The identification of $\Delta^{4,22}$-steradienes and $\Delta^{3,5,22}$-steratrienes has been confirmed by synthesis and coinjection with DSDP (leg 63) sediment extracts (N. A. Lamb, J. McEvoy, J. R. Maxwell, unpublished results). Steradienes with $\Delta^{24(28)}$ unsaturation have been tentatively identified in a number of marine sediments (*18, 57, 71*), but such assignments have yet to be confirmed by comparison with standard compounds.
74. A. Ensminger, A. van Dorsselaer, C. Spyckerelle, P. Albrecht, G. Ourisson, in *Advances in Organic Geochemistry 1973*, B. Tissot and F. Bienner, Eds. (Editions Technip, Paris, 1974) p. 245.
75. W. K. Seifert and J. M. Moldowan, *Geochim. Cosmochim. Acta* **45**, 783 (1981).
76. A. S. Mackenzie, thesis, University of Bristol (1980).
77. W. K. Seifert, in *Internationales Alfred-Triebs Symposium*, A. Prashnowsky, Ed. (Universitat Würzburg, Würzburg, Federal Republic of Germany, 1980), p. 13.
78. A. A. Petrov, S. D. Pustil'nikova, N. N. Abruiutina, G. R. Kagramonova, *Neftekhimiya* **16**, 411 (1976).
79. G. Deroo, *Intern. Rep. Inst. Fr. Pet. (Paris) No. 14427* (1967).
80. S. Ji-Yang *et al.*, *Chem. Geol.* **35**, 1 (1982).
81. J. Schaeffle, thesis, University of Strasbourg (1979).
82. B. J. Mair and J. L. Martinez-Pico, *Proc. Am. Pet. Inst.* **42**, 173 (1962).
83. E. J. Gallegos, *Anal. Chem.* **47**, 1524 (1975).
84. A. S. Mackenzie, C. A. Lewis, J. R. Maxwell, *Geochim. Cosmochim. Acta* **45**, 2369 (1981).
85. M. M. Rhead, G. Eglinton, G. H. Draffan, *Chem. Geol.* **8**, 277 (1971).
86. R. A. Berner, *Early Diagenesis—A Theoretical Approach* (Princeton Univ. Press, Princeton, N.J., 1979).
87. S. C. Brassell, A. M. K. Wardroper, I. D. Thomson, J. R. Maxwell, G. Eglinton, *Nature (London)* **290**, 693 (1981).
88. J. K. Volkman, G. Eglinton, E. D. S. Corner, J. R. Sargent, in *Advances in Organic Geochemistry 1979*, A. G. Douglas and J. R. Maxwell, Eds. (Pergamon, Oxford, 1980), p. 219.
89. J. J. Boon, W. Liefkens, W. I. C. Rijpstra, M. Baas, J. W. de Leeuw, in *Environmental Biogeochemistry and Geomicrobiology*, W. E. Krumbein, Ed. (Ann Arbor Science, Ann Arbor, Mich., 1978), vol. 1, p. 355.
90. W. Y. Huang and W. G. Meinschein, *Geochim. Cosmochim. Acta* **43**, 739 (1979).
91. K. Kawamura and R. Ishiwatari, *ibid.* **45**, 149 (1981).
92. The alkadienenone/alkatrienone ratios for shallow sediments from the Middle America Trench (*71*) are markedly higher than those of similar sediments from many temperate environments (*88*) where surface water temperatures are significantly lower.
93. B. M. Didyk, B. R. T. Simoneit, S. C. Brassell, G. Eglinton, *Nature (London)* **272**, 216 (1978).
94. T. Birkelund, R. G. Bromley, W. K. Christensen, Eds., *Cretaceous-Tertiary Boundary Events* (Univ. of Copenhagen, Copenhagen, 1979).
95. D. Leythaeuser, A. Hollerbach, H. W. Hagemann, in *Advances in Organic Geochemistry 1975*, R. Campos and J. Goñi, Eds. (Enadimsa, Madrid, 1977), p. 3.
96. A. S. Mackenzie and J. R. Maxwell, in *Organic Maturation Studies and Fossil Fuel Exploration*, J. Brooks, Ed. (Academic Press, London, 1981), p. 239; A. S. Mackenzie, J. M. E. Quirke, J. R. Maxwell, in *Advances in Organic Geochemistry 1979*, A. G. Douglas and J. R. Maxwell, Eds. (Pergamon, Oxford, 1980), p. 239.
97. W. K. Seifert, in *Advances in Organic Geochemistry 1975*, R. Campos and J. Goñi, Eds. (Enadimsa, Madrid, 1977), p. 21.
98. R. W. Jones, in *Problems of Petroleum Migration*, W. H. Roberts and R. J. Cordell, Eds. (American Association of Petroleum Geologists, Tulsa, Okla., 1980), p. 47.
99. W. K. Seifert, J. M. Moldowan, R. W. Jones, in *Tenth World Petroleum Congress* (Heyden, London, 1979), vol. 2, p. 425.
100. B. Durand and J. L. Oudin, in *ibid.*, p. 3; A. S. Mackenzie, unpublished results for oils from the Mahakam Delta, Indonesia.
101. J. C. Winters and J. A. Williams, *Am. Chem. Soc. Div. Pet. Chem. Prepr. 4(14)* (1969), p. E22.
102. H. P. Hansen, *Mar. Chem.* **3**, 183 (1975); C. W. D. Milner, M. A. Rogers, C. R. Evans, *J. Geochem. Explor.* **7**, 101 (1977); S. C. Brassell and G. Eglinton, in *Analytical Techniques in Environmental Chemistry*, J. Albaiges, Ed. (Pergamon, Oxford, 1980), p. 1.
103. J. Albaiges [in *Analytical Techniques in Environmental Chemistry*, J. Albaiges, Ed. (Pergamon, Oxford, 1980), p. 69] and unpublished results from our laboratory and from Masspec Analytical (Speciality Services) Ltd., Woodchester, Stroud, Gloucestershire, United Kingdom.
104. A. S. Mackenzie, W. K. Seifert, L. Ren-wei, J. R. Maxwell, J. M. Moldowan, in *Advances in Organic Geochemistry 1981*, M. Bjorøy *et al.*, Eds. (Heyden, London, in press).
105. N. J. R. Wright, *J. Pet. Geol.* **2**, 411 (1980).
106. D. McKenzie, *Earth Planet. Sci. Lett.* **40**, 25 (1978); *ibid.* **55**, 87 (1981); C. Beaumont, *Geophys. J. R. Astron. Soc.* **65**, 291 (1981); L. Royden, J. G. Sclater, R. P. van Herzen, *Bull. Am. Assoc. Pet. Geol.* **64**, 173 (1980); C. L. Angerine and D. L. Turcotte, *ibid.* **65**, 219 (1981).
107. For example, see L. C. Price, *J. Pet. Geol.* **4**, 89 (1980).
108. J. R. Maxwell, R. E. Cox, C. T. Pillinger, R. G. Ackman, S. N. Hooper, *Geochim. Cosmochim. Acta* **37**, 297 (1973).
109. M. De Rosa, S. De Rosa, A. Gambacorta, J. D. Bu'lock, *Chem. Commun.* (1977), p. 514; T. G. Tornabene and T. A. Langworthy, *Science* **203**, 51 (1978); M. Kates, B. Palameta, C. N. Joo, D. J. Kushner, N. E. Gibbons, *Biochemistry* **5**, 4092 (1966); M. Kates, in *Ether Lipids, Chemistry and Biology*, F. S. Snyder, Ed. (Academic Press, New York, 1972), p. 351; R. Anderson, M. Kates, M. J. Baedecker, I. R. Kaplan, R. G. Ackman, *Geochim. Cosmochim. Acta* **41**, 1381 (1977); T. G. Tornabene, R. S. Wolfe, W. E. Balch, G. Holzer, G. E. Fox, J. Oro, *J. Mol. Evol.* **11**, 259 (1978); T. G. Tornabene, T. A. Langworthy, G. Holzer, J. Oro, *ibid.* **13**, 73 (1979); G. Holzer, J. Oro, T. G. Tornabene, *J. Chromatogr.* **186**, 795 (1979).
110. T. K. Devon and A. I. Scott, *Handbook of Naturally Occurring Compounds*, part 2, *Terpenes* (Academic Press, London, 1972).
111. G. Ourisson, P. Albrecht, M. Rohmer, *Pure Appl. Chem.* **51**, 709 (1979).
112. J. R. Maxwell, C. T. Pillinger, G. Eglinton, *Q. Rev. Chem. Soc.* **25**, 571 (1971); W. E. Reed, *Geochim. Cosmochim. Acta* **41**, 237 (1977); B. R. T. Simoneit, *ibid.*, p. 463; R. E. Laflamme and R. A. Hites, *ibid.* **42**, 289 (1978); S. G. Wakeham, C. Schaffner, W. Giger, *ibid.* **44**, 415 (1980); J. W. Louda, S. E. Palmer, E. W. Baker, *Init. Rep. Deep Sea Drill. Proj.* **56/57** (part 2), 1391 (1980).
113. W. K. Seifert and J. M. Moldowan, *Geochim. Cosmochim. Acta* **43**, 111 (1979).
114. D. Attaway and P. L. Parker, *Science* **169**, 674 (1970); G. Mattern, P. Albrecht, G. Ourisson, *Chem. Commun.* (1970), p. 1570.
115. W. Henderson, W. E. Reed, G. Steel, M. Calvin, *Nature (London)* **231**, 308 (1971); G. Steel, W. E. Reed, W. Henderson, in *Advances in Organic Geochemistry 1971*, H. R. von Gaertner and H. Wehner, Eds. (Pergamon, Oxford, 1972), p. 353; G. Steel and W. Henderson, *Anal. Chem.* **44**, 1302 (1972); *Nature (London)* **238**, 148 (1972).
116. Unpublished sterol identifications of J. W. de Leeuw, J. K. Volkman, and S. C. Brassell.
117. M. T. J. Murphy, A. McCormick, G. Eglinton, *Science* **157**, 1040 (1967); W. Henderson, V. Wollrab, G. Eglinton, in *Advances in Organic Geochemistry 1968*, P. A. Schenck and I. Havenaar, Eds. (Pergamon, Oxford, 1969), p. 181; I. R. Hills, G. W. Smith, E. V. Whitehead, *J. Inst. Pet. (London)* **56**, 127 (1970).
118. E. J. Gallegos, *Anal. Chem.* **43**, 1151 (1971); B. Balogh, D. M. Wilson, A. L. Burlingame, *Nature (London)* **233**, 261 (1971).
119. I. Rubinstein, O. P. Strausz, C. Spyckerelle, R. S. Crawford, D. W. S. Westlake, *Geochim. Cosmochim. Acta* **41**, 1341 (1977).
120. G. van Graas, F. de Lange, J. W. de Leeuw, P. A. Schenck [*Nature (London)* **296**, 59 (1982)] report series of 5α(H)- and 5β(H)-A-nor steranes.
121. I. R. Hills, E. V. Whitehead, D. E. Anders, J. J. Cummings, W. E. Robinson, *Chem. Commun.* (1966), p. 752; I. R. Hills, G. W. Smith, E. V. Whitehead, *Nature (London)* **219**, 243 (1968).
122. E. V. Whitehead, *Chem. Ind.* (1971), p. 1116; A. Ensminger, P. Albrecht, G. Ourisson, B. J. Kimble, J. R. Maxwell, G. Eglinton, *Tetrahedron Lett.* (1972), p. 3861; E. V. Whitehead, in *Proceedings of a Symposium on Hydrogeochemistry and Biochemistry*, E. Ingerson, Ed. (Clarke, Washington, D.C., 1973), vol. 2, p. 158; B. Balogh, D. M. Wilson, P. Christiansen, A. L. Burlingame, *Nature (London)* **242**, 603 (1973).
123. A. Van Dorsselaer *et al.*, *Tetrahedron Lett.* (1974), p. 1349.
124. W. K. Seifert, J. M. Moldowan, G. W. Smith, E. V. Whitehead, *Nature (London)* **271**, 436 (1978); C. M. Ekweozor, thesis, University of Ibadan (1978); M. Bjorøy and J. Rullkötter, *Chem. Geol.* **30**, 27 (1980).
125. B. Corbet, P. Albrecht, G. Ourisson, *J. Am. Chem. Soc.* **102**, 1171 (1980).
126. J. Rullkötter and P. Philp [*Nature (London)* **292**, 616 (1981)] reported C_{36}–C_{40} series of 22*S*- and 22*R*-17α(H),21β(H)-hopanes.
127. M. Rohmer and G. Ourisson, *Tetrahedron Lett.* (1976), p. 3633.
128. M. Dastillung and G. Ourisson, in *Géochimie Organique des Sédiments Marins Profonds*, orgon I, *Mer de Norvège* (Editions du CNRS, Paris, 1977), p. 237; P. W. Brooks, G. Eglinton, S. J. Gaskell, D. J. McHugh, J. R. Maxwell, R. P. Philp, *Chem. Geol.* **20**, 189 (1977); J. J. Boon, J. W. de Leeuw, A. L. Burlingame, *Geochim. Cosmochim. Acta* **42**, 631 (1978); P. J. Barnes *et al.*, *Init. Rep. Deep Sea Drill. Proj.* **48**, 965 (1979).
129. I. Maclean, G. Eglinton, K. Douraghi-Zadeh, R. G. Ackman, S. N. Hooper, *Nature (London)* **218**, 1019 (1968); P. W. Brooks, J. R. Maxwell, R. L. Patience, *Geochim. Cosmochim. Acta* **42**, 1175 (1978).
130. J. M. E. Quirke and J. R. Maxwell, *Tetrahedron* **36**, 3453 (1980).
131. J. M. Moldowan and W. K. Seifert, *Science* **204**, 169 (1979).
132. I. Rubinstein and L. J. Goad, *Phytochemistry* **13**, 485 (1974); D. M. Orcutt and G. W. Patterson, *Comp. Biochem. Physiol. B* **50**, 579 (1975); J. K. Volkman, D. J. Smith, G. Eglinton, T. E. V. Forsberg, E. D. S. Corner, *J. Mar. Biol. Assoc. U.K.* **61**, 509 (1981).
133. J. A. Ballantine, K. Williams, B. A. Burke, *Tetrahedron Lett.* (1977), p. 1547; R. M. K. Carlson *et al.*, *Bioorg. Chem.* **7**, 453 (1978); C. Delseth, R. M. K. Carlson, C. Djerassi, T. R. Erdman, P. J. Scheuer, *Helv. Chim. Acta* **61**, 1470 (1978).
134. C. Delseth, L. Tolela, P. J. Scheuer, R. J. Wells, C. Djerassi, *Helv. Chim. Acta* **62**, 101 (1979).
135. A. Kanazawa, S. Teshima, T. Ando, S. Tomita, *Bull. Jpn. Soc. Sci. Fish.* **40**, 729 (1974); J. K. Volkman, E. D. S. Corner, G. Eglinton, in *Biogéochemie de La Matière Organique à l'Interface Eau-Sédiment Marin*, R. Daumas, Ed. (Éditions du CNRS, Paris, 1980), p. 185; J. K. Volkman, G. Eglinton, E. D. S. Corner, *Phytochemistry* **19**, 1809 (1980).
136. Y. Shimizu, M. Alam, A. Kobayashi, *J. Am. Chem. Soc.* **98**, 1059 (1976).
137. L. J. Mulheirn and G. Ryback, in *Advances in Organic Geochemistry 1975*, R. Campos and J. Goñi, Eds. (Enadimsa, Madrid, 1977), p. 173.
138. We thank our colleagues at Bristol for helpful discussions and, in particular, the following for their assistance: A. Gowar (GC-MS), N. Lamb (monoaromatic steroids and $\Delta^{4,22}$-steradienes), D. Smith (extract of *C. hyalinas*), S. Ji-yang (sterane data for Zhanahua Basin samples), C. Hoffman (bibliography and triaromatic steroids), and T. Lewis (figures). We thank T. Wardroper, S. Rowland, and J. McEvoy for access to unpublished results. Our work is supported mainly by the Natural Environment Research Council (GR3/2951, GR3/3419, and GR3/3758), by a NASA subcontract (NGL 05-003-003) from the University of California, Berkeley, and by British Petroleum Company Ltd.