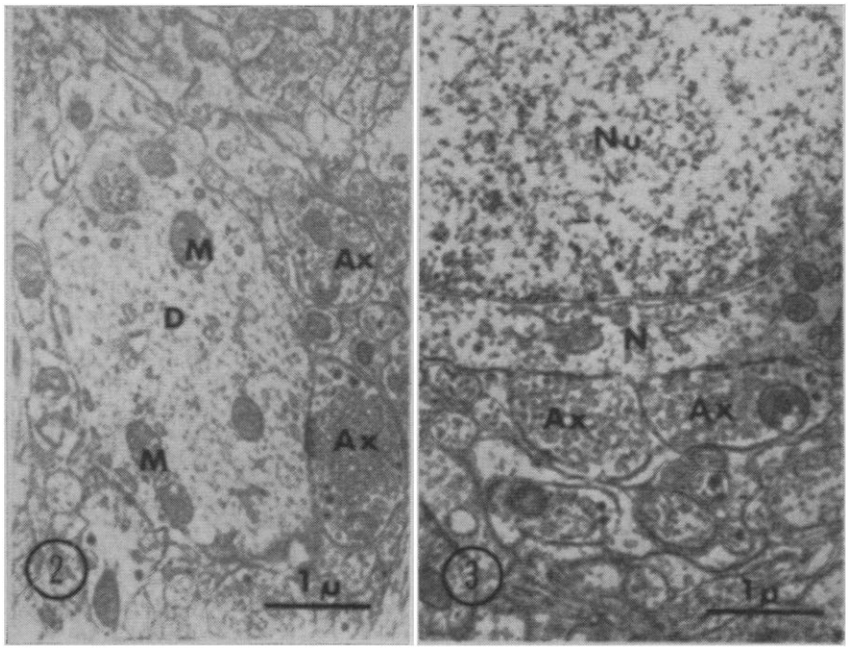

Fig. 2. Dendrite (*D*) in neuropil of median eminence of an MSG-treated adult rat. *Ax*, axon terminal; *M*, mitochondrion (× 15,000). Fig. 3. Neuron and neuropil in the arcuate nucleus of an MSG-treated adult rat. *Ax*, axon terminal; *M*, mitochondrion; *Nu*, nucleus (× 16,300).

brains of the infant rats killed 3 hours after a single injection of MSG failed to reveal any effects upon the lateral preoptic nucleus, arcuate nucleus, and median eminence. The possibility still existed that the single injection of MSG could have produced effects which were not obvious with the light microscope; therefore, the same neural regions from male and female adult rats were examined with the electron microscope. Electron microscopic observations of the lateral preoptic nucleus, arcuate nucleus, and median eminence region did not show any edematous swelling or disintegration of organelles in neurons or neuronal processes such as was illustrated by Olney (*2*) in the infant monkey. There was no indication that such damage had ever occurred (Figs. 1–3). These areas were also examined with respect to distribution of neurons and glia, distribution and size of axon terminals and dendrites, nucleolar size, and distribution of dense-core and clear vesicles. No differences were observed between MSG-treated and control animals.

Not only were no morphological lesions of the brain detectable after MSG treatment, but adult animals also did not manifest any pronounced disturbances with respect to reproductive function. The lack of MSG effect was determined by the following criteria: (i) adult MSG-treated females cycled normally and were capable of mating and producing normal litters; (ii) although the relative ovarian weights were significantly less than the controls (34.7 ± 0.9 versus 29.3 ± 1.4, $P < .01$), no differences were noted when the ovaries were examined histologically; (iii) the relative uterine weights were not significantly different; and (iv) no significant differences were observed in the weights of testes, seminal vesicles, and prostates of MSG-treated males compared to controls (Table 1).

These results fail to duplicate the findings reported in mice and monkey by Olney (*1*) and Olney and Sharpe (*2*). Since the experimental approaches used in the present study and reported by Olney are very similar, it is difficult to determine the cause of the discrepancies at this time. However, the possibility exists that the differences may be due to variation in species susceptibility.

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11 May 1970

Sterols in Recent Marine Sediments

Abstract. *Sterolic fractions have been isolated from Recent marine sediments representing two different environments. The fractions were characterized by infrared spectroscopy and thin-layer chromatography. The major sterols in the fractions were identified by gas chromatography and mass spectrometry. The demonstrated survival of these common plant and animal sterols for several thousand years suggests that these molecules will be useful geochemical indicators.*

Most organic matter produced by a marine community its quickly re-oxidized by the organisms or by inorganic processes. A small fraction escapes oxidation and is deposited in sediments. This small fraction is the object of study for organic geochemistry. The part of this preserved organic matter not bound into kerogen contains most of the preserved biochemicals (*1*).

The high degree of structural diversity of sterols suggests that the sterols might carry significant information with regard to the type of biological community associated with various sediments and perhaps even to specific organisms of the community (*2*). Using a colorimetric method, Schwendinger and Erdman (*3*) made a survey for sterols and reported positive tests for them in a series of Recent sediments, varying from freshwater to deep marine. They stated that high sterolic concentrations of 60 to 300 parts per million in their samples make sterols possible precursors for some of the compounds found in petroleum. Others (*4*) think that sterols or their degradation products are major contributors to the optical activity of petroleum because much of this activity is confined to that fraction of compounds having 27 to 30 carbon atoms. We now report the finding of several specific sterols from marine sediments from two different marine environments.

The bottom meter of a three-meter

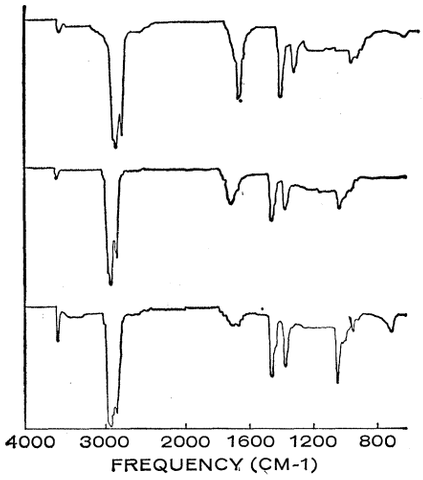

Fig. 1. Infrared spectra of sterol fraction from Baffin Bay sediment (upper), sterol fraction of sediment from San Pedro Basin (middle), and mixture of campesterol and β-sitosterol (lower). Measurements were made on solutions in carbon tetrachloride.

piston core from Baffin Bay, Texas, and a grab sample from San Pedro Basin, offshore from southern California, were analyzed. The section of core was estimated to be from 2000 to 3000 years old. The hydrocarbons, long chain alcohols, and fatty acids in these samples have been well studied (see *5*).

The samples were extracted with mixtures of distilled chloroform and distilled methanol by stirring at room temperature. The resulting extracts were saponified in methanolic KOH. The unsaponifiable fractions were extracted from the reaction mixtures in ethyl ether and chromatographed in a mixture of ethyl ether and heptane (1:9) on columns of silica gel (Woelm). Fractions were collected, and those in which sterolic materials were indicated by thin-layer chromatography, were combined and chromatographed again on fresh columns of silica gel. The results of thin-layer chromatography indicated that one fraction from each of the second columns contained primarily sterols. These fractions gave the characteristic lavender color of sterols when on thin layers of silica gel G. They were sprayed with 5 percent sulfuric acid in ethanol and heated at 95°C for 15 minutes and they exhibited R_F values corresponding to that of a mixture of authentic sterols.

The infrared spectra of the two sterolic fractions were measured in carbon tetrachloride (7 mg/ml). Figure 1 shows the infrared spectrum of the Baffin Bay and San Pedro sterolic fractions and the infrared spectrum of a mixture of campesterol and β-sitosterol. The band at 3580 cm^{-1} (oxygen-hydrogen stretching), the 1050 cm^{-1} (carbon-hydroxyl stretching), and the general similarity of the spectra suggested that the fractions were sterolic.

The lipid yield from the Baffin Bay sample was 400 ppm on a dry-weight basis, and the yield of sterolic material was 6 ppm. The San Pedro sample gave 10,000 ppm of lipids and 40 ppm of sterols which is near the range of concentrations reported by Schwendinger and Erdman (*3*).

Some of the components of these fractions were tentatively identified by comparative gas chromatography on columns (1.8 m by 0.3 cm) of 3 percent JXR on Chromosorb Q (80/100 mesh). The temperature was programed from 175° to 250°C at 1 deg/min. Helium, flowing at 35 ml/min, was the carrier gas. Dual columns with flame detectors were used. Four components of each sterolic fraction exhibited retention times the same as those of cholesterol, campesterol, stigmasterol, or β-sitosterol. Coinjection of the sterolic fractions with authentic samples of those four sterols indicated that retention times corresponded exactly.

The sterolic fractions were treated with refluxing acetic anhydride for 3 hours, and resulting steryl acetates were isolated by the method of Vishniac (*6*). Comparative gas chromatography of the acetyl derivatives and of authentic steryl acetates also indicated that four major components of each of the fractions were cholesterol, campesterol, stigmasterol, and β-sitosterol. Figure 2 shows the gas chromatogram of the sterolic fraction of the Baffin Bay sediment. Suggested identities of four of the components are shown. Figure 3 shows the corresponding gas-chromatogram of the sterolic fraction from the San Pedro sample. It has a pattern similar to that of the Baffin Bay sample; although it contains a major concentration of component *X* which, if present in the Baffin Bay sample, is a minor component.

Mass spectra data of the Baffin Bay sample indicate molecular ions at mass 386, 388, 400, 412, 414, 416, and 428. Ions of mass 386, 400, 412, and 414 correspond to cholesterol, campesterol, stigmasterol, and β-sitosterol, respectively.

When all components of these sterolic fractions are identified, it should be possible to assess the relative contributions of phytogenic and zoogenous

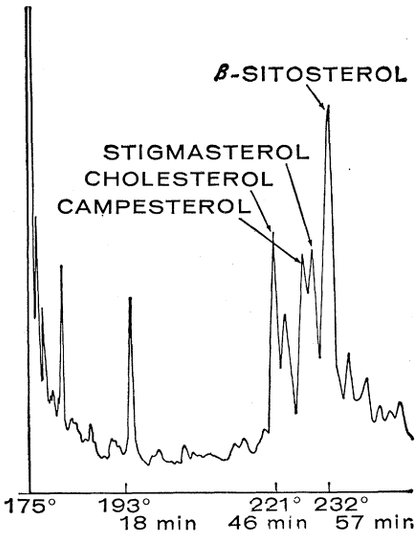

Fig. 2. Gas chromatogram of sterol fraction of Baffin Bay sediment.

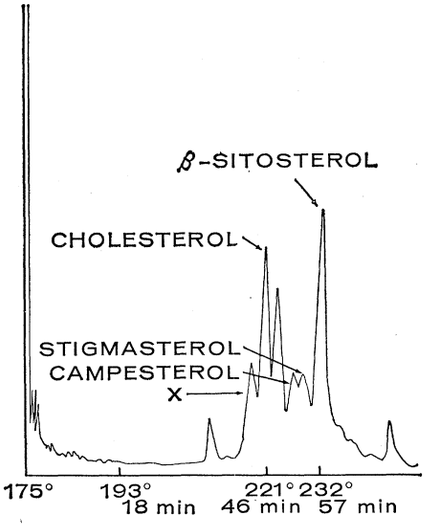

Fig. 3. Gas chromatogram of sterol fraction from San Pedro sample.

sterols and perhaps to suggest types of source organisms for them.

Since a triterpene alcohol, similar in structure to sterols, has been isolated (*7*) from the Messel oil shale (50 million years old) it is likely that intact sterols are also present in similar ancient sedimentary rocks. Further study of sterol geochemistry may yield clues to the nature of contributing organisms which in turn partly identifies the environments at times of deposition. For example, blue-green algae and bacteria lack sterols so that an environment dominated by these forms might have an unusual sterol pattern.

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Oil Spills: Method for Measuring Their Extent on the Sea Surface

Abstract. *It is difficult to estimate the area affected by an oil spill at sea, the degree of coverage by oil pollutants within the affected area, and the quantity of pollutants involved. Estimates of volumes and flow rates are based on estimated changes in areal extent of the spill. Uncertainties in measurement of area degrade the accuracy of estimating other parameters. To resolve this problem, available stock components have now been assembled into a system that yields repeatable, economical measurement of the areal extent of oil spills at acceptable levels of accuracy. The system comprises overflights with a thermal infrared imaging system, densitometric color enhancement of the infrared images, and automatic digital planimetry of the areas of specified image densities.*

Recurrent spillages of petroleum and petroleum products are becoming ever more serious sources of pollution of the marine environment. It has been difficult to estimate the area affected by a given oil spill, the degree of concentration of pollutants within the affected area, and the quantity of pollutants involved. Oil on the sea surface does not image well in the spectral bands recorded by conventional black-and-white or color photography. By use of a thermal infrared mapper and densitometric color enhancement of the resulting infrared image, in combination with automatic digital planimetry, it is now possible to map the areal extent of an oil spill at a given time and to obtain, automatically and economically, a repeatable and accurate measurement of the area of sea surface actually covered by oil pollutants. We have tested a fairly simple system that shows a good potential for breaking through the bottleneck of much uncertainty and considerable disagreement in the estimates of oil spill magnitudes.

The Santa Barbara oil spill is one of the events that stimulated our present, newfound awareness of the delicate environmental balance and its vulnerability to pollution. The Santa Barbara oil spill was touched off on 28 January 1969, when well No. 21 blew out of control on Union Oil Company's platform A (some 9 km south of Santa Barbara, on Federal Lease Parcel No. 402). Vast quantities of crude oil polluted the channel during the following 10½ days, until the well was plugged. Some 4 days after the well was cemented, vigorous oil leaks began to flow from the fractured bedrock beneath the sea floor adjacent to platform A (*1*). This flow has gradually decreased to the point where only about 1000 gallons (3000 to 4000 liters) per day are still leaking out at present.

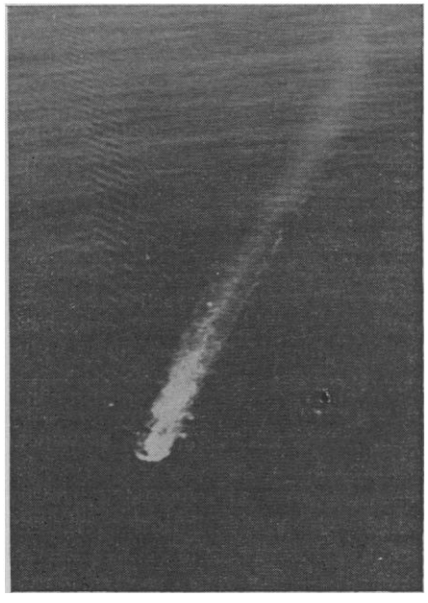

Fig. 1. Black-and-white rendition of a color photograph showing oil-covered sea surface except where underlying water is revealed in the boat's wake. Without the contrasting tone image of the boat's wake, this photograph and the color original from which it was made would be perceived as showing a normal sea surface.

Estimates of the amount of spillage vary: a low figure was proposed by Union Oil and the U.S. Geological Survey, and much higher estimates by independent researchers. Alan A. Allen of General Research Corporation computed that 2.2 million gallons (8⅓ million liters) of crude petroleum poured into the Santa Barbara Channel during the first 10 days and that spillage during the spill's first year was 3.3 million gallons (12½ million liters). Both Allen and the Union Oil Company now agree that present flow rates average about 30 barrels (5 m^3) per day. Allen's flow rate estimate of 220,000 gallons (830,000 liters) per day during the first 10 days is ten times that of the oil company's more reassuring figure of 500 barrels (80 m^3) daily for the same time period (*2*). On the assumption that an unbiased search for truth motivated all estimates, such a tenfold variability demonstrates the need for developing simple, standardized techniques that could be applied uniformly and that could materially increase the accuracy of estimating such basic parameters.

Floating oil on seawater does not photograph well on panchromatic emulsions. The contrast between an oil patch and the adjacent water is very low. High contrast occurs only where a favorable sun angle accentuates reflectance from the oil. Even then, such oil reflectance is difficult to distinguish from sun glitter on oil-free waters. Oil-free areas of upwellings, surface currents, and localized air flows show a streaked, alternately choppy and smooth sea surface, which is difficult to distinguish from patchy oil slicks. Color photography is similarly ineffective. The color contrast between the oil film and the adjacent water is again very low. Except for areas where thick ropy streamers of oil register as dark gray, the oil-polluted waters show a grayish-green color, which the eye readily accepts as a normal sea surface tone. It is, therefore, often impossible to tell whether a particular air photograph shows a completely oil-covered or completely oil-free sea surface. Figure 1, a black-and-white rendition of a color photograph, illustrates this condition; only the boat's wake revealing the oil-free water underneath shows that the surrounding sea surface is entirely covered by oil.