exposed at 0-day stage and assayed at the 20-day stage. In contrast, with prothalli exposed at the 0-day stage and assayed at 12-day stage none of the eluates caused antheridium formation within the specified period of time. The results are interpreted to mean that the sections of the chromatogram which caused antheridium induction also caused the induction of the physiological state inhibitory to antheridium formation. Clearly, a substance inducing the inhibitory state was not separated from A_{An}. The two solvent mixtures used led to the chromatographic separation of the three native fern antheridiogens thus far demonstrated (*12*). The possibility cannot be excluded that the inhibitory state is induced by A_{An}.

Such a hypothesis seems paradoxical. The described phenomenon might be functional in preventing the precocious attainment of the sexual phase. However, the inducibility of the inhibitory state has already largely decayed at the 4-day stage. The prothalli developed from the germinated spores then comprised an average of 1.5 green vegetative cells (only cells seen outside the spore coat were counted). The first spores germinated 3 days after inoculation. Quite likely, the peak in the inducibility of the inhibitory state begins to decline even before the spore germinates. It may be stressed that A_3 (*5*) and medium harvested from *Anemia* cultures (*11*) not only cause antheridium formation but also can cancel the light requirement for spore germination. In all likelihood, the native factor that induces germination in the dark is identical with A_{An} (*11*). In nature, recently shed spores are probably exposed to A_{An} secreted by prothalli developed from spores shed earlier. Accordingly, I propose that the inducibility of a state antagonistic to antheridium formation may be functional in the prevention of "hybrid metabolism," that is, the simultaneous occurrence of physiological states leading to germination and to formation of antheridia. A consideration of the above situation is valid regardless of whether or not light promotes germination by causing the synthesis of A_{An}. At a genetic level such a phenomenon would have its parallel in the finding that activation at one gene site may lead to suppression at another.

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Gaseous Hydrocarbons in Sea Water: Determination

Abstract. *Low-molecular-weight hydrocarbons in the C_1 to C_4 range have been detected and measured in sea water. Methane shows by far the highest concentration, followed in order by the C_2, C_3, and C_4 fractions. In general, the concentration tends to decrease with depth, but there is a strong indication that a maximum may exist about 30 to 50 meters below the surface.*

We report what we believe to be the first determinations of C_1 to C_4 low-molecular-weight hydrocarbons in waters of the open ocean. The presence of methane is well established in freshwater lakes and marshes (*1*). The presence of methane was recently reported in Lake Nitinat (*2*), an anoxic fjord on Vancouver Island; it apparently occurred only in the oxygen-depleted zone where H_2S also was found. Similar findings have been reported in the anoxic areas of the Black Sea. However, nothing has been reported of low-molecular-weight hydrocarbons other than methane.

With the development of a new and and extremely sensitive gas-chromatographic technique (*3*), we have succeeded in identifying and measuring methane, ethane, ethylene, propane, propylene, *n*-butane, *iso*-butane, and some of the butenes in waters of the open ocean. Some of the chromatograms showed small peaks having longer retention times than the butenes had, indicating the presence of C_5 hydrocarbons. Our primary purpose was to evaluate the analytic method as applied to sea water, so that very few supporting oceanographic data were taken at the time of sample collection.

Samples were collected from two areas: the open Atlantic some 500 km west of Ireland, and the Gulf of Mexico just off the continental shelf, directly south of Mobile, Alabama. All sampling was with standard Nansen casts; upon retrieval, the sea water was immediately transferred to an all-glass 1-liter bottle by gravity flow, the bottle being filled to overflowing by means of a 6-mm copper tube extending to its bottom. A tapered ground-glass stopper was inserted in such a way so as to avoid entrapment of room air.

Analysis was by a technique in which the dissolved hydrocarbons are first stripped from solution by purging with helium, and then concentrated in cold traps containing appropriate adsorbents; they are subsequently released by increase in temperature and swept into the chromatograph by a second stream of helium carrier gas. With this technique, sample size is not restricted and very dilute solutions may be analyzed.

Two cold traps (−77°C) in series were used. In the first, activated alumina was used to trap all hydrocarbons except methane; in the second, activated charcoal was used to trap methane. When the stripping was complete, the traps were isolated by closure of appropriate valves, and their temperature was raised to approximately 90°C. Helium carrier gas was then used to strip each adsorbent, in turn, of the

Table 1. Concentrations ($\times 10^{-7}$ ml/liter) of hydrocarbons found in waters from the Gulf of Mexico (G) and the North Atlantic (A). Ethane and ethylene were not separable with the column used.

Depth (m)	Methane		Ethane and ethylene		Propane		Propylene		*iso*-Butane	
	G	A	G	A	G	A	G	A	G	A
0	676	474	45.0	99	2.4	1.9	9.3	5.9	5.9	2.9
30	2830	600	102	51.3	6.6	3.4	6.3	8.5	6.9	2.4
500	246	488	5.4	12.3	1.7	0.8	1.6	3.1	5.9	1.4

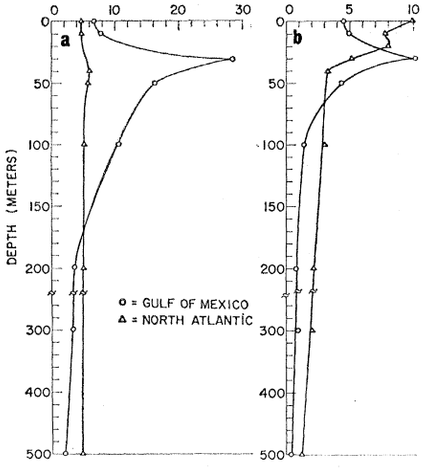

Fig. 1. Concentration-depth profile for methane (a; $\times 10^{-5}$ ml/liter) and the ethane-ethylene fraction (b; $\times 10^{-6}$ ml/liter). Gulf of Mexico samples from 28°59'N, 88°11'W; Atlantic samples from 52°35'N, 20°9'W.

adsorbed gases and to carry these gases into the chromatograph (*4*) for further separation and analysis (*3*).

For the low-molecular-weight hydrocarbons, the absolute sensitivity of the method is approximately 2×10^{-12} moles. On the basis of the 1-liter samples of water used by us, this sensitivity corresponds to about 1 part in 10^{13} by weight, or approximately 5×10^{-8} ml of dissolved gas per liter of sea water. At this lower limit, the precision of the method, on the basis of replicate measurements under laboratory conditions, is ± 10 percent (relative S.D.). For quantities of gas greater by a factor of 10 or more, the precision improves to ± 0.4 percent. Additional error incurred during field operations may increase this spread in uncertainty to as much as ± 1 percent. The chromatograph was calibrated with an artificial gas mixture containing known amounts of the hydrocarbons in question.

In most instances, analysis of our Atlantic samples at the Naval Research Laboratory entailed considerable time delay caused by shipment, some samples being analyzed from 2 weeks to 3 months after collection. The Gulf of Mexico samples, however, were analyzed within 2 to 6 hours of collection (*5*). Duplicate analyses were made of the Gulf samples: the first, immediately; the second, about 1 month later at the Naval Research Laboratory. All samples were stored in bottles having ground-glass stoppers. Duplicates did not differ significantly.

At the time of collection, several samples were treated with either sodium azide or mercuric chloride to retard bacterial action; a few samples were deliberately left untreated. Treatment with sodium azide had no noticeable effect, but mercuric chloride caused the complete disappearance of unsaturated hydrocarbons such as ethylene and propylene.

With all samples, concentrations of the C_1 to C_4 hydrocarbons generally tended to decrease with increase in depth, but in several samples the concentration profile showed a maximum at a depth of 30 to 50 m, where the concentration was from 4 to 15 times greater than at 500 m (Fig. 1). Figure 1a shows a dramatic increase in methane concentration near the surface for the Gulf water; the Atlantic samples showed only a slight increase in methane at the same depth. Figure 1b shows a similar trend for the ethane-ethylene fraction.

Some illustrative data for samples from both sources are shown in Table 1. In all instances, as one might anticipate, methane showed by far the highest concentrations. An interesting feature of these observations is shown by the propane-propylene data; in the 0- to 50-m range, the unsaturated-hydrocarbon concentration appears to exceed significantly the concentration of the saturated component. Individual ethane and ethylene concentrations, while not shown in Table 1, exhibit the same trend in the same range of depth.

In order to separate ethane from ethylene, a different chromatographic column was used, causing an increase in retention time of the other hydrocarbons; consequently, use of this column was limited during the period of production of these data. Data are still too few to permit conclusions concerning the possible significance of these values; the work continues.

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Symbiosis: Effects of a Mutualistic Fungus upon the Growth and Reproduction of Xyleborus ferrugineus

Abstract. Xyleborus ferrugineus *beetles developed from asymbiontic eggs through the adult stage on a sterilized meridic diet, but the resulting adults reproduced only when a mutualistic fungus was inoculated into the diet. Beetles with bacterial symbionts still required the fungus for reproduction on a second meridic diet.*

As a nutritional substrate for insects, wood is especially deficient in vitamins, sterols, and related growth factors. Because species of insects successfully attack and reproduce in the wood of most trees, their sources of vitamins, sterols, and related growth factors in such situations is of interest. Wood-infesting beetles in the families Scolytidae and Platypodidae are closely associated with microbes in their brood galleries. The associated microbes have been strong suspects as sources of the nutrients that are absent in wood but that are required by these beetles.

Francke-Grosmann (*1*) recently reviewed the extensive knowledge of the interrelationships among ambrosia beetles (Platypodidae and certain species of Scolytidae), their microbial symbionts, and their woody substrate. This knowledge lacked proof that microbes produce specific nutrients essential to the beetles.

In studies of the nutritional interrelationships among the ambrosia beetle *Xyleborus ferrugineus* (Fabricius), its microbial symbionts, and the woody substrate, we have found that the insects developed from asymbiontic eggs through the adult stage on a sterile meridic diet (I) similar to that described by Chippendale and Beck (*2*), but eggs produced by these asymbiontic adults were not viable.

The females tunneled and fed extensively in the sterile medium, but oviposited viable eggs only after a mutualistic fungus (*Fusarium solani*) was inoculated into the diet. This fungus was isolated consistently from the oral mycangia (organs for the transport and