6. B. Richter, *Nucl. Instrum. Methods* **136**, 47 (1976).
7. ———, in *11th International Conference on High Energy Accelerators*, W. S. Newman, Ed. (Birkaüser, Basel, 1980), p. 168.
8. A. L. Robinson, *Science* **216**, 1395 (1982).
9. ———, *ibid.* **217**, 814 (1982).
10. D. Ayres *et al.*, "A 1000 GeV on 1000 GeV proton-proton colliding beam facility," unpublished Fermilab/Argonne report (1976).
11. E. Fisk and J. A. MacLachlan, in *Proceedings of the 1982 DPF Summer Study on Elementary Particle Physics and Future Facilities*, R. Donaldson, R. Gustafson, F. Paige, Eds. (Fermilab, Batavia, Ill., 1982), p. 347.
12. L. C. Teng, Ed., *Proceedings of the Workshop on Possibilities and Limitations of Accelerators and Detectors* (Fermilab, Batavia, Ill., 1978); U. Amaldi, Ed., *ibid.* (CERN, Geneva, 1979).
13. R. Diebold *et al.*, in *Proceedings of the 1982 DPF Summer Study on Elementary Particle Physics and Future Facilities*, R. Donaldson, R. Gustafson, F. Paige, Eds. (Fermilab, Batavia, Ill., 1982), p. 307.
14. R. Huson *et al.*, in *ibid.*, p. 315.
15. L. W. Jones, in *ibid.*, p. 345.
16. S. van der Meer, CERN/ISR-PO/72-31, unpublished report (1972).
17. "Design report Tevatron 1 project," unpublished Fermilab report (1982).
18. G. R. Lambertson and C. W. Leemann, in *Proceedings of the 1982 DPF Summer Study on Elementary Particle Physics and Future Facilities*, R. Donaldson, R. Gustafson, F. Paige, Eds. (Fermilab, Batavia, Ill., 1982), p. 338.
19. M. Tigner *et al.*, *Report of the 20 TeV Hadron Collider Technical Workshop* (Cornell University, Ithaca, N.Y., 1983).
20. R. Diebold, in *Particles and Fields—1982*, W. Caswell and G. Snow, Eds. (American Institute of Physics, New York, Conference Proceedings *98*, 1983), p. 89; see also S. C. Loken and P. Nemethy, Eds., *Proceedings of the 1983 DPF Workshop on Collider Detectors* (LBL-15973, Lawrence Berkeley Laboratory, Berkeley, Calif., 1983).
21. R. Palmer *et al.*, in *Proceedings of the 1982 DPF Summer Study on Elementary Particle Physics and Future Facilities*, R. Donaldson *et al.*, Eds. (Fermilab, Batavia, Ill., 1982), p. 90.
22. M. Tigner, in *ibid.*, p. 50.
23. R. R. Wilson, in *ibid.*, p. 330.
24. A. L. Robinson, *Science* **221**, 350 (1983).
25. P. Carruthers and R. Slansky, Los Alamos National Laboratory, unpublished reports.
26. W. Dieterle *et al.*, University of Arizona, unpublished reports.
27. C. Norman, *Science* **220**, 392 (1983).
28. M. M. Waldrop, *ibid.*, p. 809.
29. A. L. Robinson, *ibid.* **214**, 769 (1981); W. J. Broad, *ibid.* **218**, 551 (1982).
30. J. D. Bjorken *et al.*, "Fermilab dedicated collider," Fermilab report (1983).
31. S. Wojcicki *et al.*, *Report on the 1983 Subpanel on New Facilities for the U.S. High Energy Physics Program*, (DOE/ER Report, Department of Energy, Washington, D.C., 1983).
32. M. M. Waldrop, *Science* **221**, 344 (1983).
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Biotechnology in the Marine Sciences

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Summary. Genetic engineering applied to the production of fish, molluscs, algae, algal products, and crustaceans in natural environments and hatchery systems is still at the rudimentary stage. Cloning systems for producing commercially important chemicals, pharmacologically active compounds, and metamorphosis-stimulating substances present in marine organisms are being sought. Attempts are being made to develop useful drugs from the sea, including antineoplastic, antibiotic, growth-promoting (or -inhibiting), analgesic, and antispasmodic agents. Immediate commercial applications can be expected from engineered systems involving polysaccharide and specialty chemical production, with marine microorganisms as the source of genetic material.

Genetic engineering holds extraordinary promise for the marine sciences. The potential of the world oceans to feed and sustain humankind has been addressed during the past few decades, including reports of a huge food source represented by krill in Antarctic waters and by fishery stocks in offshore waters of the world's continents (*1*).

Genetic engineering is being applied to develop the products of fish, molluscs, and crustaceans in natural environments and hatchery systems, although real results are still scant. Streisinger *et al.* (*2*) have produced clones of homozygous diploid zebra fish (*Brachydanio rerio*). Successful aquaculture of many species of invertebrate animals and large populations of shellfish at the larval and intermediate stages has made it possible to clone their genes, so that the stage is set for the realization of genetic engineering's potential in the marine sciences.

A dramatic example of the potential that biotechnological application offers is that of marine pharmaceuticals. In a 1977 conference, "Drugs and Food from the Sea: Myth or Reality," investigators described cardiotonic polypeptides from sea anemones, an adrenergic compound from a sponge, and potential antitumor agents from Caribbean gorgonians and soft corals (*3*). More recently, Rinehart *et al.* (*4*) described antiviral and antitumor depsipeptides from a Caribbean tunicate. Extracts prepared from the Caribbean tunicate, an ascidian or sea squirt of the family Didemnidae, inhibit growth of DNA and RNA viruses, as well as L1210 murine leukemic cells. These depsipeptides—termed didemnins after the name of the tunicate family, Didemnidae, from which they are isolated—are closely related but vary in activity. The discovery indicates that the subphylum Tunicata or Urochordata (phylum Chordata) may be an abundant source of bioactive compounds of pharmaceutical interest (*4*). Another tunicate, of the genus *Tridemnum*, when extracted with a mixture of methanol and toluene (3:1), showed activity against herpes simplex virus, type 1, grown in CV-1 cells (monkey kidney tissue), indicating that the extract inhibited the growth of the virus. This antiviral activity may also include antitumor activity. When tested against other viruses, essentially all extracts of the tunicate collected at a number of sites showed activity in inhibiting both RNA and DNA viruses. The suggestion that the extracts might also have antitumor properties was evidenced from their high potency against L1210 leukemic cells. The novelty of the didemnins results from a new structural unit for a complex of depsipeptides, hydroxyisovaleryl propionate, and a new stereoisomer of the unusual amino acid statine (*4*).

In a review of compounds from the sea that act on the cardiovascular and central nervous system, Kaul (*5*) pointed out that drugs of high pharmacologic activity from nature have, in fact, been unsurpassed by synthetic compounds. Drugs from nature, predominantly from plants, include morphine, atropine, and digitalis glycosides. Marine animals and plants have yielded cardiovascular-active sub-

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stances, and these include histamine and *N*-methylated histamines of the sponge *Verongia fistularis* (*6*), asystolic nucleosides from the sponge *Dasychalina cyathina*, and the nucleoside spongosine, isolated from *Cryptotethya crypta*, leading to useful drugs as a result of the activity of spongosine.

Several marine organisms already provide useful compounds: fish liver oil provides an excellent source of vitamins A and D; insulin has been extracted from whales and tuna fish; and the red alga *Digenia simplex* has long been used to prepare an antihelminthic agent. Bacteriologists, for many years, have incorporated agar and alginic acids into laboratory media. The fact that it has been uneconomical to extract and purify material from organisms that have to be captured in large quantities from remote corners of the world, coupled with the general lack of knowledge concerning the basic chemistry of many marine natural products, has limited these sources for the development of useful drugs. Genetic engineering can change this situation dramatically, by revealing the vast and diverse genetic composition of marine life for pharmacological application.

Marine Toxins

Of particular interest are toxins produced by marine organisms. A toxin is a substance having a specific functional group arranged in the molecule or molecules and showing strong toxic physiological activity (*7*). Many toxins have potential applications as a drug or pharmacological agent. Where direct use as a drug is not feasible, because of potent or harmful side effects, some toxins can still serve as models for synthesis or improvement of other drugs. Drugs produced by marine organisms are being screened for antibiotic, growth-promoting (or -inhibiting), hemolytic, analgesic, antispasmodic, blood-pressure regulating, antiviral, and anti-inflammatory activities.

Two successes demonstrate the potential. Tetrodotoxin, which can paralyze peripheral nerves, is valuable because it inhibits the sodium permeability of nerve membranes and has been useful for elucidating the excitation mechanism. Both saxitoxin and tetrodotoxin have been used to study structure of the sodium channel of membranes (*8*).

A second success is an insecticide developed from nereistoxin and widely marketed in Japan since 1966. Fishermen are familiar with the fact that flies die when they come into contact with the dead specimens of the marine annelid *Lumbriconereis heteropoda* commonly used as bait. The toxin was first isolated in 1934, and once its structure was determined, a new insecticide, PADAN, was developed from nereistoxin (*9*). Cartap hydrochloride is one of the synthesized derivatives. Active against the rice stem borer and other insect pests, it does not appear to be toxic to warm-blooded animals, and resistant strains of insects do not readily develop (*7*).

Hashimoto (*7*) has compiled an extensive literature summarizing information about marine organisms causing food poisoning or having the capacity to produce a toxic sting or bite or are poisonous, as in the case of the toxic marine flagellates, for example, *Gonyaulax* and *Gymnodinium* spp. The vast array of marine animals and plants covered in the compendium includes algae, coelenterates (that is, jellyfish), echinoderms, corals, sea anemones, molluscs, algae, flagellates, nemertines, annelids, and other invertebrates, and fish (*10–17*). Research on marine toxins or bioactive substances in marine organisms has increased in recent years, and a number of monographs and reviews are available, including those of Russell and Saunders (*13*); Baslow (*14*); Martin and Padilla (*15*); Scheuer (*16*); Marderosian (*17*); and Youngken and Shimizu (*18*). Recently ciguatoxin, palytoxin, and halitoxin have also been investigated and provide interesting new information (*19*).

Hybridoma technology offers a valuable tool for characterizing the marine toxins (*20*). It was used in our laboratory to produce monoclonal antibodies to the enterotoxin of *Vibrio cholerae*, a brackish water and estuarine bacterium causing cholera and apparently related to the *Vibrio* sp. producing palytoxin (*21, 22*). Monoclonal antibodies against cholera toxin were produced to obtain highly specific antiserums to cholera toxin (*23*). Halitoxin, a toxic mixture from several marine sponges of the genus *Haliclona*, has been isolated, partially purified, spectrally characterized, and chemically degraded, yielding a proposed chemical structure for the toxin (*24*). The toxin has proved to be a complex mixture of high molecular weight and can be isolated from the sponges *Haliclona rubens*, *H. viridis*, and *H. erina*. The sponge extracts are detectably toxic for fish and mice (LD_{50} of approximately 275 mg/kg) and also inhibit the growth of Ehrlich ascites tumors (*14, 25*). Thus, the halitoxins may prove to be an antitumor agent or agents. The hybridoma approach to characterizing these compounds should permit wider screening for the distribution of the toxin among various sponges and lead to subsequent isolation and testing (*21*).

Similarly, lophotoxin, a new neuromuscular toxin from several Pacific gorgonians of the genus *Lophogorgia*, has been purified (*26*). Originally discovered during a search for chemical defense adaptations of marine organisms, the toxin occurs in horny corals or gorgonians (sea fans and whips, phylum Cnidaria, order Gorgonacaea) in tropical or subtropical waters, and exhibits cytotoxic, ichthyotoxic, and antibacterial activities. Lophotoxin inhibits nerve-stimulated contraction without affecting contraction evoked by direct electrical stimulation of the muscle. The data suggest that the epoxylactone and furanoaldehyde groups in the molecule may be responsible for the potent biological properties of lophotoxin.

Palytoxin, an extremely poisonous, water-soluble substance from zoanthid coral of the genus *Palythoa* (*27*), has been described and its structure elucidated (*21, 22, 28–30*). *Palythoa* species are abundant in most tropical waters and many *Palythoa* species have been reported to be moderately toxic (*28*). Palytoxin was used by natives on Maui to tip their weapons as a defensive advantage against invaders from the island of Hawaii. The toxic nature of zoanthids of the genus *Palythoa* was discovered by Ciereszko and Attaway in 1961 (*31*). The component responsible for the toxicity, palytoxin (PTX), was isolated several years later independently by two groups of investigators (*24, 32*).

The molecular size of the toxin is 2681 daltons, with three nitrogens in the molecule (*29*), and an elemental composition of $C_{129}H_{223}N_3O_{54}$. The complete stereochemical structure has been described, with the toxin representing an entirely new class of natural products. Its biogenesis is not clear, but the structure of this complex compound has been determined (*27, 30*). Palytoxin is a linear arrangement of 115 carbon atoms with 8 methyl groups and 40 hydroxyl groups. As was pointed out by Fenical (*19*), unusual features of palytoxin are two cyclic ketal constellations which are part of 11 ether linkages.

Palytoxin contains no repeating units, as is common in medium-sized molecules in nature. It is neither a peptide nor a terpene, but, rather, more like the polyether antibiotics. Some of the nonadjacent carbons along the chain are interconnected by ether, ketal, and hemiketal linkages, with no carbocyclic rings in the molecule. Palytoxin is similar in potency to ricin, a polypeptide toxin

found in castor beans. Although not quite as deadly as the toxin of *Clostridium botulinum*, it exerts its lethal effect when administered intravenously in low concentrations to laboratory animals. Palytoxin influences calcium and potassium ion transport in nerves and the heart. Animals undergo paralysis and heart failure. The mechanism of the cardiovascular effects of palytoxin has been reviewed (*33*).

A fascinating aspect of palytoxin is that it is synthesized by a marine *Vibrio* species growing symbiotically with the coelenterate *Palythoa* and apparently related to *Vibrio cholerae*. This vibrio reportedly is only mildly pathogenic for humans (*21, 22*), causing an influenza-like illness, and is rapidly attenuated in laboratory culture, losing its ability to make toxin. The *Palythoa* species containing the toxin grows in a tide pool along the coast of Maui not far from Hana. By land, the tide pool is virtually inaccessible, and native fishermen avoid that part of the coast (*21, 22*). Thus, palytoxin represents an interesting and highly challenging candidate for cloning of the gene (or genes) synthesizing the toxin [see (*34*)].

A very interesting compound, bryostatin, which has antitumor properties, has been isolated from marine animals of the phylum Ectoprocta, specifically *Bugula neritina*. The extremely low dose for antineoplastic activity suggests that bryostatin may have other potentially useful pharmacological or microbiological activities, sufficient to merit its use as a biochemical probe (*35*).

The applications of marine toxins are limited, and it is mainly in the area of understanding functions that the toxins have an interest. At the present time, strategies are needed for collecting, culturing, and screening marine organisms from which bioactive agents can be isolated and characterized. The need to study the fundamental chemistry, to determine the structures or new compounds, as has been done for bryostatin (*35*) and palytoxin cannot be overemphasized (*30*). The immediate successes are most likely to occur with discoveries of novel antineoplastic agents, antibiotics, and anti-inflammatory agents produced by marine bacteria, plants, and animals.

Industrial Chemicals

In the short term, marketable products from the sea may come first from marine polysaccharides, carotenoids, and specialty chemicals; these would include unusual sugars, enzymes, and algal lipids. Seaweeds are more important economically than is generally realized, since they are used as human and animal food, in medicine and agriculture, and as a source of raw materials for numerous industries. Carrageenin is prepared from the red seaweeds and is widely used as an extender in foods and related products, from evaporated milk to toothpaste. Agarose, another polysaccharide, is widely used in electrophoresis and chromatography analyses in the laboratory. Applications of genetic modification in seaweeds to commercial utilization and culture has been reviewed (*36*). Abbott and Cheney (*37*) have estimated that most people living in developed maritime countries today, directly or indirectly consume or come into contact with some form of algae daily. In general, the application of genetic modification or improvement techniques to seaweed culture is recent, but somewhat limited, the most common being simple strain selection, such as screening of wild plants for desirable traits such as fast growth. Protoplast fusion–somatic hybridization techniques are now being applied to agar-producing seaweeds (*35*). Specialty chemicals from salt-tolerant microbial systems, notably the polysaccharides, as well as enzymes and lipids, offer great potential in the immediate future.

In addition to alginates, chitin, and agarose, other compounds and metabolites have been reported, including spatene diterpenoids from the tropical marine alga *Stoechospermum marginatum*, and many other unusual algal products not observed from terrestrial sources (*38*). Fenical (*19*) has provided a useful discussion of natural products chemistry in the marine environment.

A new polyether derivative of okadaic acid, a C_{38} fatty acid, has been isolated from *Halichondria* (syn. *Reniera*) *okadai* Kadota, a black sponge commonly found along the Pacific coast of Japan, and *H. melanodocia*, a Caribbean sponge found in the Florida Keys (*39*). The structural features of okadaic acid suggest that it belongs to the class of compounds known as ionophores, hitherto known only from terrestrial microorganisms. It is likely that okadaic acid is a metabolite of an epiphytic microorganism, rather than of *Halichondria* spp. Also, however, structural studies of the marine toxins palytoxin and ciguatoxin indicate that the two compounds have many structural features normally associated with ionophores. In fact, okadaic acid has been isolated from the dinoflagellate *Procentrum lima*, a likely progenitor of okadaic acid (*39*).

Sponges and gorgonians have been useful sources of biologically active metabolites because they are frequently abundant, permitting pursuit of trace metabolites. These unusual compounds may be pathway intermediates and also offer potential sources of new chemicals. An antimicrobial sesterterpene-1, called palauolide, has been described (*40*).

Sea hares offer the advantage of being rich sources of interesting metabolites, but the ultimate source of such metabolites is not always clear, often proving to arise from algae. Recent work on pharmacologically active substances associated with sea hares, coelenterates, and sponges has been summarized by Schmitz *et al.* (*24*). Notable are the extracts of the sea hare *Aplysia dactylomela* which show both cytotoxicity and in vivo antitumor activity, as well as a variety of halogenated metabolites.

Recently the cloning and expression of sea urchin histone genes have been reported (*41*). The molecular biology of the sea urchin embryo has also been extensively studied (*42*). Thus, the cloning of genes of marine animals has begun.

The developments in molecular genetics in propelling industrial microbiology into new dimensions of activity have been described by Demain (*43*). Directed search for biologically active natural products (other than antibiotics) in the marine environment, especially those described as being confined to marine organisms (*28*), can open an entirely new source of industrial chemicals. What is needed immediately are new and novel screening strategies for such products (*44*).

Biotechnology Applications in Aquaculture

In the United States, most of the traditional fisheries are being harvested at or near maximum sustainable yields. Approximately 60 percent of the fisheries products consumed in the United States are imported, representing a trade deficit in excess of $2.5 billion. Mariculture offers good potential for reducing this deficit. Exploiting microbial sources of protein at the larval stages and during larval metamorphosis and growth can reap enormous benefits and profit.

Thus, interest in aquaculture is on the rise, and that interest has meant increases in our knowledge of marine biology and the technical expertise to apply discoveries in marine biology to aquaculture. The National Sea Grant College Program has been at the forefront of many of this country's advances in aqua-

Table 1. Chemical inducers of specific settlement of invertebrate larvae.

Invertebrate	Inducer	Reference
Barnacles	Arthropodins (proteins)	(*59*)
Coelenterates	Iodinated proteins	(*60*)
	Lag-phase bacterial products	(*66, 67*)
Sponges	*Pseudomonas* lectin	(*56*)
Oysters	Iodinated proteins	(*61*)
	Thyroxine	(*61*)
	L-Dopa	(*68*)
	D-Dopa or melanin	(*69*)
	Isochrysis	(*70*)
Bivalves	γ-Aminobutyric acid	(*62*)
	Algal Phycoerythrobilins	(*62*)
	Coral mucus	(*58*)
	Chlorine compounds	(*58, 71*)
Sea urchin	Low molecular weight bacterial by-product	(*72*)

culture and marine biomedical research. Research has been under way for more than a decade on marine shrimp, freshwater prawn, crawfish, blue crab, brine shrimp, salmon and other finfish, oysters, clams, abalone, and scallops. A major problem of mariculture is disease, predominantly microbially mediated infections and epidemics. Agents that are common hatchery complaints include infectious pancreatic necrosis (IPN), Egtved and other viruses, and *Vibrio* and *Aeromonas* among the bacteria (*45*). Many causes of disease and loss of hatchery stocks are still not yet known, nor are controls of epizootics yet available.

On the Atlantic coast, work on diseases in salmon has focused on IPN. The antigenic relationships of various IPN isolates are being studied to develop polyvalent antiserums and effective vaccines against the virus. Temperature-sensitive mutants of the virus have also been isolated to provide genetic information and possible sources of attenuated virus strains for use in live vaccines.

An extract from *Ecteinascidia turbinada* (Ete) has been shown to enhance the hemocyte function of invertebrates, such as blue crabs (*Callinectes sapidus*), crayfish (*Procambarus clarkii*), and prawns (*Macrobrachium rosenbergii*), possibly rendering the animals more resistant to infection (*46–50*). Interestingly, intraperitoneal injection of Ete renders eel strongly resistant to *Aeromonas hydrophila* and appears to create the potential for phagocytic activity. Ete also causes changes in the concentration of peripheral blood leucocytes (*46–50*).

Many species of shellfish and finfish are available in culture, providing excellent opportunities for selection and gene manipulation. Production, stabilization, and delivery of vaccines, whether by hybridoma technology or genetic engineering, will enhance productivity from the egg through the larval stages, at present a high-risk portion of the life cycle.

Stock assessment of migrating fish and species identification remain unresolved issues in fishery management. A method for comparison of mitochondrial DNA (mtDNA) from different individuals offers an opportunity for mapping the mtDNA genome and is being explored (*51*). The method of using restriction digests to analyze mtDNA has permitted investigation of evolutionary relationships among species and conspecific populations. When applied to marine fishes and invertebrates, analysis of mtDNA offers a possible mechanism for assessing the population structure of marine fishes.

The use of genetic marking, by introducing selected genetic traits into fish, as opposed to mechanical tags, is yet another avenue for marine genetic engineering. Fish wander over long distances without barriers to their movements, but now there can be a way to detect subpopulations or specific independent stocks by the very precise methods that yield unequivocal results.

Marine plants also offer special opportunities, and genetic engineering of osmoregulation is being studied (*52*). Plants that are halophytes can be useful in agricultural areas where the soil has become too salty for conventional agriculture, and application to marine and estuarine grasses can prove beneficial in managing erosion and shoreline losses.

Biofouling

Fouling of surfaces in the marine environment is a costly burden for any operation carried out in or near seawater. Biofouling occurs in progressive steps from the initial primary film to attachment of invertebrate animals capable of boring and digesting the surface (*53*). The ability of bacteria to find, attach, adhere, elaborate specific film-forming substances, and regulate expression of all these functions is fundamental to the fouling process. Fortunately, the tools of genetic engineering are well-suited to analysis of the properties of bacterial cell-surface components, since specific genetic elements determining the structure of specific polysaccharides and polypeptides can be isolated and the mechanism of cell-surface association probed so that appropriate steps for intervention can be taken.

Another facet of the biofouling issue should also be considered. Larvae of invertebrates prefer to settle on surfaces coated with microbial films (Table 1). Graham *et al.* (*54*) have shown that settlement and metamorphosis do not occur in the absence of microbial films, in the case of the tube-forming polychaete, *Janua* (*Dexiospira*) *brasiliensis*, which is a small (2 to 3 mm), hermaphroditic polychaete abundant on various surfaces, especially on *Zostera* (eelgrass).

Planula larvae of the medusa *Cassiopeia andromeda* settle on a substrate, attach, form a pedal disk and undergo metamorphosis, that is, they elongate, segregate stalk and calyx, and develop a hypostome and tentacle anlagen (*55*). Pedal-disk formation and metamorphosis are initiated by a substance or substances produced by a marine *Vibrio* species. The inducing factor is released into the culture medium by the bacteria.

For commercially important shellfish, such as the American oyster *Crassostrea virginica*, enhanced larval set and metamorphosis can be critical to a successful hatchery operation. A few years ago, a unique bacterium, LST, was isolated from tanks containing oyster larvae at the mariculture unit in Lewes, Delaware. It appears to be a member of a new genus, especially since its DNA does not hybridize with that of *Vibrio*, *Pseudomonas*, or other related strains. LST has since been repeatedly isolated from oysters and oyster beds, where it is believed to be associated with induction of settlement and metamorphosis of the oyster. The strain adheres very strongly to cultch and other hard surfaces, forming microcolonies on the cultch. In sufficient numbers, notably during the decline phase of growth, the bacterium produces a pigment product which attracts oyster larvae and may effect larval development and metamorphosis. Such relationships between bacteria and invertebrates are

known to occur in the marine environment, as for example, the sponge *Halichondria panicea* and *Pseudomonas insolita* (*56*).

The LST pigment appears to be melanin and it, or its precursors including L-dopa, may act as a metamorphosis-inducing agent for oyster larvae (*57*). The pigment, concentrated and partly purified by centrifugation, precipitation, and dialysis, is heterogeneous, with molecular sizes ranging from 12,000 to 120,000 daltons.

Planktonic larvae of benthic invertebrates settle and metamorphose in response to specific substances or conditions in their environment (*58*). In the absence of such substances, metamorphosis can be delayed indefinitely. In only a few cases has the metamorphosis-stimulating substance been isolated and identified. Barnacle larvae metamorphose in response to contact with a species-specific arthropodin, a soluble, tanned protein present in the cuticle of adult barnacles (*59*). Iodinated proteins are active on coelenterates (anthozoan planulae and scyphozoan scyphystomae) (*60*). The same class of compounds has been isolated from the mantle cavity of adult oysters and stimulates metamorphosis of oyster larvae (*61*). Bivalves and gastropods (larvae of the pink abalone) can be induced to metamorphose in the presence of gamma-aminobutyric acid a peptide almost identical to one of the main neurotransmitters in the human brain (*62*). A fragment of the molecule induces larvae of the abalone to metamorphose to the juvenile form. Thereafter, early growth sets in, triggered by insulin and growth hormone produced by the animal. The findings of Morse and associates (*62*) provide exciting new results and the promise of cloning the genes controlling growth and synthesis-accelerating peptide hormones.

Bacteria mediate metamorphosis of coelenterates (*Hydractinea planulae* larvae) and the sea urchin—in the latter case, in association with a 1- to 5-kilodalton protein. A carbohydrate produced by *Pseudomonas marina* stimulates invertebrate development via lectin interaction (*63*). Thus, several specific chemical inducers have been described to date, which trigger invertebrate morphogenesis.

Another fascinating aspect of the molecular biology of marine invertebrates is protochordate allorecognition. Colonial tunicates, unlike vertebrates, undergo transplantation in nature. Rejection or acceptance of colonies has been shown recently by Scofield *et al.* (*64*) to be controlled by a single gene locus with multiple alleles. The same genetic region apparently maintains polymorphism by preventing fertilization between gametes sharing alleles. Marine invertebrates also have a histocompatibility system that is involved in immune recognition of tissue antigens and rejection of transplanted allogenic organs, tissues, and cells, a system comparable with a mammalian major histocompatibility complex with its controlling gene system (*64, 65*).

Conclusion

The few examples offered here illustrate an immense potential of biotechnology for the marine sciences, but they scarcely begin to reveal opportunities which lie ahead. There exists a strong foundation for exploitation of biologically active compounds already known to occur in the sea and for further exploration into the recesses of the world oceans for compounds and food sources as yet undiscovered.

The greatest opportunity of all is represented by the applications of genetic engineering to the marine sciences to pursue the untapped gene pool representing transport systems for minerals, metal concentration, novel photosynthetic systems, and marine pheromones by marine animals. The advent of genetic engineering may bring to reality the potential of the oceans to serve as a significant source of protein on a scale not heretofore predicted. The scale of management and stock breeding for domestic animals can now be duplicated for fish and shellfish—providing an even greater advantage, compared with domestic livestock, since the generation time and maturation cycle is significantly shorter for these marine species.

References and Notes

1. F. Nast, *Meeresforschung* **29**, 154 (1982); H. Klendt, H. Thiel, F. Nast, *Arch. Fischereiwiss.* **33**, 85 (1982); V. Siegel, *ibid.*, p. 113.
2. G. Streisinger, C. Walker, N. Dower, D. Knauber, F. Singer, *Nature (London)* **291**, 293 (1981).
3. P. N. Kaul and C. J. Sinderman, Eds., *Drugs and Food from the Sea. Myth or Reality* (Univ. of Oklahoma Press, Norman, 1978).
4. K. L. Rinehart, Jr., *et al.*, *Science* **212**, 933 (1981).
5. P. N. Kaul, *Fed. Proc. Fed. Am. Soc. Exp. Biol.* **40**, 10 (1981).
6. K. H. Hollenbeak, F. J. Schmitz, P. N. Kaul, *Proceedings of the Food and Drugs from the Sea Conference* (Marine Technology Society, Washington, D.C., 1976), p. 282; also see P. N. Kaul, *Mar. Technol. Sci. J.* **29**, 123 (1979).
7. Y. Hashimoto, *Marine Toxins and Other Bioactive Marine Metabolites* (Japan Scientific Societies Press, Tokyo, 1979).
8. D. D. Doyle, M. Wong, J. Tanaka, L. Barr, *Science* **215**, 1117 (1982).
9. T. Okaichi and Y. Hashimoto, *Agric. Biol. Chem.* **26**, 224 (1967).
10. B. W. Halstead, *Poisonous and Venomous Marine Animals of the World* (Government Printing Office, Washington, D.C., 1965).
11. ———, *ibid.* (1967), vol. 2.
12. ———, *ibid.* (1970), vol. 3.
13. F. E. Russell and P. R. Saunders, Eds., *Animal Toxins* (Pergamon, New York, 1967).
14. M. H. Baslow, *Marine Pharmacology* (Williams Wilkins, Baltimore, 1969); republished with update (Krieger, Huntington, N.Y., 1977).
15. D. F. Martin and G. M. Padilla, *Marine Pharmacognosy* (Academic Press, New York, 1973).
16. P. J. Scheuer, Ed., *Marine Natural Products: Chemical and Biological Perspectives* (Academic Press, New York, 1978–1981), vols. 1 to 4.
17. A. D. Marderosian, *J. Pharm. Sci.* **58**, 1 (1969).
18. H. W. Youngken, Jr., and Y. Shimizu, in *Chemical Oceanography*, J. P. Riley and G. S. Kirrow, Eds. (Academic Press, New York, ed. 2, 1975), vol. 4, p. 269.
19. W. Fenical, *Science* **215**, 923 (1982).
20. M. D. Scharff and S. Roberts, *J. Tissue Culture Assoc.* **17**, 1072 (1981).
21. R. E. Moore, P. Helfrich, G. M. L. Patterson, *Oceanus* **25**, 54 (1982).
22. J. L. Fox, *Chem. Eng. News* (4 January 1982), p. 19.
23. E. F. Remmers, R. R. Colwell, R. A. Goldsby, *Infect. Immun.* **37**, 70 (1982).
24. F. J. Schmitz *et al.*, *Pure Appl. Chem.* **51**, 853 (1981).
25. P. Turlapaty, S. Shibata, T. R. Norton, M. Kashiwagi, *Eur. J. Pharmacol.* **24**, 310 (1973).
26. W. Fenical, R. K. Okuda, M. M. Bandurraga, P. Culver, R. S. Jacobs, *Science* **212**, 1512 (1981).
27. R. E. Moore and P. J. Scheuer, *ibid.* **172**, 495 (1971).
28. J. T. Baker and V. Murphy, *Compounds from Marine Organisms* (CRC Press, Cleveland, Ohio, 1976).
29. R. E. Moore, F. X. Woolard, G. Bartolini, *J. Am. Chem. Soc.* **102**, 7370 (1980).
30. R. E. Moore and G. Bartolini, *ibid.* **103**, 2491 (1981).
31. D. H. Attaway, thesis, University of Hawaii, 1968.
32. ——— and L. S. Ciereszko, *Isolation and Partial Characterization of Caribbean Palytoxin*, in *Proc. Sec. Internat. Coral Reef Symp.* (Brisbane, Australia, 1974), p. 497.
33. S. K. Kulkarni, W. G. Kirlin, P. K. Kaul, in *Drugs and Food from the Sea*, P. M. Kaul and C. J. Sindermann, Eds. (Univ. of Oklahoma Press, Norman, 1978), pp. 73–80.
34. R. W. Old and S. B. Primrose, *Principles of Gene Manipulation: An Introduction to Genetic Engineering* (Univ. of California Press, Berkeley, 1980).
35. G. R. Pettit, C. L. Herald, D. L. Boubek, D. L. Herald, *J. Am. Chem. Soc.* **104**, 6846 (1982).
36. D. P. Cheney, *Proceedings of the Sea Grant Conference*, Massachusetts Institute of Technology, Cambridge, 1983 (Wiley, New York, in press).
37. I. A. Abbott and D. P. Cheney, in *Selected Papers in Phycology*, R. Rosowski and B. Parker, Eds. (Phycological Society of America, Lawrence, Kans., 1982), vol. 2, p. 799.
38. W. H. Gerwick, W. Fenical, M. U. S. Sultanbawa, *J. Org. Chem.* **46**, 2233 (1981).
39. K. Tachibana, P. J. Scheuer, Y. Tsukitani, H. Kikuchi, D. Van Engen, J. Clardy, Y. Gopichand, F. J. Schmitz, *J. Am. Chem. Soc.* **103**, 2469 (1981).
40. B. Sullivan and D. J. Faulkner, *Tetrahedron Lett.* **23**, 907 (1982).
41. J. S. Kapstein and G. C. Fareed, *J. Supramol. Struct.* **8**, 7 (1979).
42. E. H. Davidson, B. R. Hough-Evans, R. J. Britten, *Science* **217**, 17 (1982).
43. A. L. Demain, *ibid.* **214**, 987 (1981).
44. S. Revah-Moiseev and A. Carroad, *Biotechnol. Bioeng.* **13**, 1067 (1981).
45. W. Ahne, *Fish Diseases* (Springer-Verlag, New York, 1980).
46. M. M. Sigel, L. L. Wellham, W. Lichter, L. E. Dudek, J. L. Gargus, A. H. Lucas, *Anticellular and Antitumor Activity of Extracts from Tropical Marine Invertebrates* (Marine Technology Society, Washington, D.C., 1969).
47. M. M. Sigel, *Ann. N.Y. Acad. Sci.* **234**, 198 (1974).
48. W. Lichter, D. M. Lopez, L. L. Wellham, M. M. Sigel, *Proc. Soc. Exp. Med. Biol.* **150**, 475 (1975).
49. W. Lichter, M. M. Sigel, D. M. Lopez, L. L. Wellham, *Inhibition of DNA Synthesis by* Ecteinascidia turbinata *Extract (Ete)* (Marine Technology Society, Washington, D.C., 1976).
50. M. M. Sigel, L. J. McCumber, J. A. Hightower, S. S. Hayasaka, E. M. Huggins, Jr., J. F. Davis, *Am. Zool.* **23**, 221 (1983).

51. D. Power, personal communication.
52. D. W. Rains and R. C. Valentine, *Genetic Engineering of Osmoregulation* (Plenum, New York, 1980).
53. G. W. Smedes and L. E. Hurd, *Ecology* **62**, 1561 (1981).
54. S. Graham, D. Kirchman, R. Mitchell, *Biol. Bull. (Woods Hole, Mass.)* **159**, 160 (1980).
55. R. Neumann, *Marine Ecology, Prog. Ser.* **1**, 21 (1979).
56. W. E. G. Muller, R. K. Zahn, B. Kurelec, C. Lucu, I. Muller, G. Uhlenbruch, *J. Bacteriol.* **145**, 548 (1981).
57. R. Weiner, in preparation.
58. M. G. Hadfield, *Settlement and Metamorphosis of Marine Invertebrate Larvae* (Elsevier, New York, 1978), p. 165.
59. D. J. Crisp, in *Chemoreception in Marine Organisms*, P. T. Grant and A. M. Mackie, Eds. (Academic Press, New York, 1974), p. 177.
60. D. B. Spangenberg, *J. Exp. Zool.* **178**, 183 (1971).
61. F. P. Veitch and H. Hidu, *Chesapeake Sci.* **12**, 173 (1971).
62. D. P. Morse, H. Duncan, N. Hooker, A. Belour, G. Young, *Fed. Proc. Fed. Am. Soc. Exp. Biol.* **39**, 3237 (1980).
63. D. Kirchman, S. Graham, D. Reish, R. Mitchell, *J. Exp. Mar. Biol. Ecol.* **56**, 153 (1982).
64. V. L. Scofield, J. M. Schlumpbergere, L. A. West, I. L. Weissman, *Nature (London)* **295**, 499 (1982).
65. V. L. Scofield and I. L. Weissman, *Dev. Comp. Immunol.* **5**, 23 (1981).
66. A. Bourdillon, *C. R. Acad. Sci. Paris* **239**, 1434 (1954).
67. W. A. Müller, *Wilhelm Roux's Arch. Dev. Biol.* **173**, 107 (1973).
68. S. Coon, personal communication (1983).
69. R. Weiner and R. R. Colwell, in preparation.
70. R. Mitchell and L. Young, *Tech. Rep. No. 3*, U.S. Office Naval Research Contract No. N00014-67-A-0298-0026 NR-306-025 (1972).
71. D. B. Bonar and M. G. Hadfield, *J. Exp. Mar. Biol. Ecol.* **16**, 227 (1974).
72. R. A. Cameron and R. T. Hinegardner, *Biol. Bull. (Woods Hole, Mass.)* **146**, 335 (1974).
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RESEARCH ARTICLE

Mineralogic Information from a New Airborne Thermal Infrared Multispectral Scanner

Anne B. Kahle and Alexander F. H. Goetz

Abstract. *A new six-channel aircraft multispectral scanner has been developed to exploit mineral signature information at wavelengths between 8 and 12 micrometers. Preliminary results show that igneous rock units can be identified from their free silica content, and that carbonate as well as clay-bearing units are readily separable on the digitally processed images.*

In the past decade, research in geologic remote sensing (*1*) has focused mainly on the development of techniques to exploit data obtained in the visible and near-infrared region of the spectrum extending from wavelengths between 0.4 and 2.5 μm (*2*). This region contains spectral features associated with electronic transitions in the transition metals in minerals and overtone vibrations in minerals containing hydroxyl and carbonate ions (*3*). Airborne and spaceborne multispectral scanners obtain images in this region by measuring reflected solar radiation, and the spectral information has been used extensively in geologic mapping (*2, 4*).

Now, a new airborne instrument, the thermal infrared multispectral scanner (TIMS), has been developed. With this instrument it is possible to obtain spectral emittance data in the thermal infrared, that portion of the electromagnetic spectrum which is dominated by thermal radiation from the earth's surface. In particular, the 8- to 13-μm region contains diagnostic spectral emission features for silicates. In silicate rocks there is a broad minimum in emissivity between 8 and 11 μm, the reststrahlen band, which results from interatomic stretching vibrations of silicon and oxygen bound in the crystal lattice (*5*); the depth and position of the band is related to the crystal structure of the constituent minerals. The band center has been shown (*5*) to move to longer wavelengths and decrease in intensity with decreasing quartz content and the concomitant increase in mafic mineral content. Sheet silicates (clays) also have an aluminum-oxygen-hydrogen bond bending mode feature in addition to the silicon-oxygen stretching features. Carbonate emittance spectra are essentially featureless in this wavelength region.

Early attempts to use multispectral image data in the thermal infrared (*6*) met with limited success because only two channels of data were available and the radiometric sensitivity of the instrument was inadequate. A sensitive instrument is required because the contrast in spectral emittance among rocks is usually less than 0.15, while the visible and reflective infrared regions exhibit contrasts of 0.5 or greater. Kahle and Rowan (*7*) were successful in obtaining data in six thermal infrared channels with an aircraft scanner over the East Tintic Mountains, Utah. While results were promising, the scanner was dismantled shortly thereafter and the experiment could not be repeated.

The TIMS is a six-channel scanner with high radiometric sensitivity that consists of a 19-cm diameter Newtonian reflector telescope mounted behind an object-plane 45° flat scanning mirror and followed by a Czerny-Turner spectrometer (*8*). In the focal plane, the entrance slit to the spectrometer acts as the field stop and defines the instantaneous field of view of 2.5 mrad. The total field of view scanned is 80°. A six-element mercury-cadmium-telluride dector array, cooled by liquid nitrogen, is mounted at the exit of the spectrometer. The nominal spectral bands covered are 8.2–8.6, 8.6–9.0, 9.0–9.4, 9.4–10.2, 10.2–11.2, and 11.2–12.2 μm. The sensitivity ranges between 0.1° and 0.3°C noise-equivalent temperature change at 300 K. This sensitivity is comparable to a noise-equivalent change in spectral emissivity of 0.002 to 0.006.

Some of the first flights with the TIMS have been over sites previously studied with other remote sensing instruments: Death Valley, California, and the Cuprite mining district in Nevada (Fig. 1). The image data are highly correlated from one spectral channel to the next, mainly because the radiance measured has a strong dependence on surface temperature. The diagnostic information in the multispectral data, however, lies in the emissivity variations as a function of

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