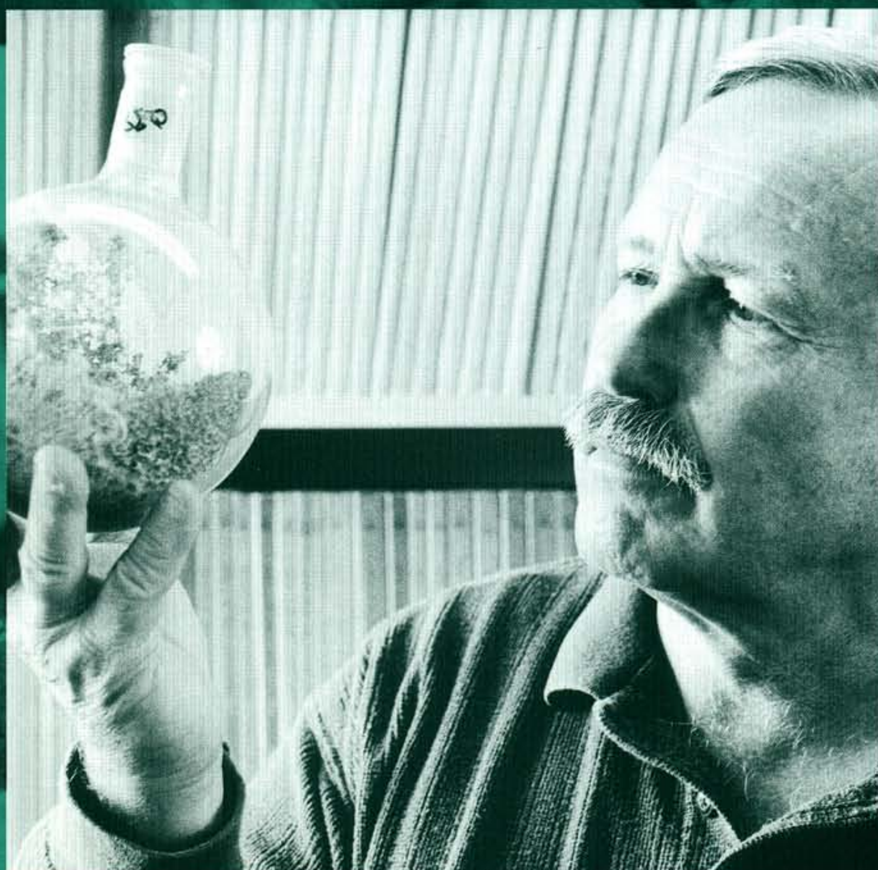
MARINE MEDICINE MEN

Searching Tropical Oceans for New Cures

BY JANET HOWARD



WILLIAM FENICAL

DESPITE MORE THAN 25 years of diving around the world in search of unique marine creatures, William Fenical was baffled when he came across a strange, yellow coral growing on an underwater boulder in the Indian Ocean.

"Soft corals just don't look like that," said the Scripps professor of marine chemistry. "They looked like fingers sticking out all over the rock. I had no idea what they were when I collected them."

The coral, named *Eleutherobia*, turned out to be a rare species of soft coral that shows great promise as a potential drug to fight breast and ovarian cancer.

Fenical discovered *Eleutherobia* in 1993 while diving off the northwest coast of Australia. The research team had decided to explore a shallow area of water known as Bennett's Shoal, which serves as home to a wide variety of marine life. Finding Bennett's Shoal, however, was not easy.

"We eventually found a guide—this very colorful, aging Aussie bloke," Fenical said, staring out his office window overlooking the Scripps Pier and chuckling. "He was one of the only guys around there, sort of a folkloric character out of the middle of the Aussie outback. The guy was riddled with bullet hole scars and had survived some kind of boat fire."

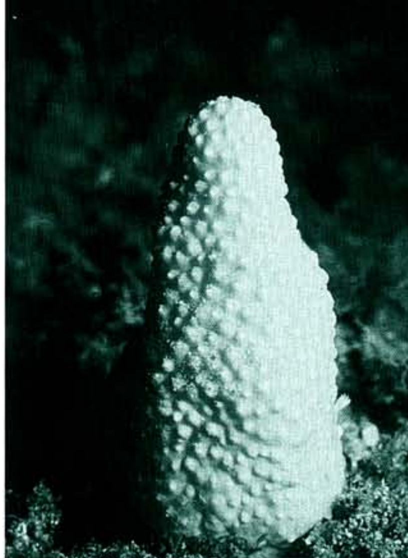
While regaling the team with shark stories, including how one had turned over his boat, the guide, who went by the name of Pancho, led the crew to Bennett's Shoal.

"He knew how to get to it perfectly," said Fenical. "I don't know how, but he did. So we jumped over and it was a real spooky place. It was very murky and there were things swimming around all over the place. You couldn't tell what was going on. Visibility was only a few feet."

As Fenical approached a large boulder, he noticed some small organisms that resembled cheese puffs sticking out from the rock.

"I've been working on soft corals for 25 years and I never saw this species before," he said. "I never saw anything like it."

Recently patented and licensed to Bristol-Myers Squibb, a chemical called eleutherobin, extracted from the coral, appears to function similarly to taxol in preventing cancer cells from dividing. Heralded as a breakthrough treatment for breast and ovarian cancer, taxol is found in the bark of the Pacific yew tree. Because extracting the product



Left, *Eleutherobià*, a soft coral discovered by William Fenical, produces a chemical compound effective in fighting cancer. Scripps researchers explore tropical reefs throughout the Indo-Pacific and Caribbean in search of marine organisms that may yield natural compounds with biomedical potential.

> "THE OCEAN CAN CONTRIBUTE ENORMOUSLY TO THE CURE AND UNDERSTANDING OF HUMAN DISEASE. OUR GOAL IS TO TAKE ADVANTAGE OF THAT VAST RESOURCE."
>
> **WILLIAM FENICAL**

resulted in the death of the slow-growing tree, Bristol-Myers Squibb began producing a semisynthetic version of the drug from a precursor to taxol found in the trees' needles. While it is a potent weapon against cancer, taxol is difficult to administer and has serious side effects, including immune system suppression, nausea, and hair loss.

One of Fenical's students, Thomas Lindel, stumbled across eleutherobin's taxol-like potential while studying the unusual coral in the lab. After extracting the chemical, Lindel tested it in a standard bioassay used to determine whether substances show activity against human colon cancer cells. He was stunned by what he saw.

"The stuff was so extraordinarily potent that it was dangerous to handle," said Fenical, director of the Scripps Center for Marine Biotechnology and Biomedicine. "You could dilute it a million-fold, and it still killed cells very powerfully."

Further tests showed that eleutherobin mimics taxol's very unusual method of blocking cell division. Like taxol, it binds to cellular structures called microtubules, which are part of the mitotic spindle and play a key role in cell division. Once eleutherobin has attached to the microtubules, they become extremely rigid and prevent the cancer cells from dividing.

"Taxol is important because it is the only drug discovered that acts by this very specific mechanism," Fenical said. "People have looked for ten years and rarely found molecules anything like it, and we just stumbled across it in the middle of this place off the coast of Australia."

While eleutherobin will have to go through years of testing to determine its effectiveness in humans, Fenical points to it as an example of the potential the oceans hold as a source of new pharmaceuticals.

"The ocean can contribute enormously to the cure and understanding of human disease," he said. "Our goal is to take advantage of that vast resource."

Fenical and colleague John Faulkner, also a Scripps professor of marine chemistry, are considered pioneers of the relatively new field of marine natural products chemistry. The field, which did not come into its own until the 1970s, is garnering more attention as scientists find it progressively more difficult to find new drugs from terrestrial sources.

"Everybody is saying, 'We need new antibiotics, where are we going to get them?'" said Fenical. "And here is this vast resource out there in the ocean that has been completely overlooked despite the fact that the oceans form the majority of the surface of Earth."

With so much to choose from, how do scientists know which marine organisms to collect? The key, said Faulkner, is to look for organisms that appear to defend themselves chemically rather than rely on the protection of such things as shells and spines or the mobility to run away and hide.

"There is an inverse correlation between physical protection and chemical protection," said

Faulkner. "So if we go out onto the reef and we see something that looks like a large chunk of food—poorly protected, soft-bodied, and easy to grab—and nothing is eating it, then we assume it has chemical protection."

The underlying assumption is that some of the chemicals that help protect marine organisms also may ward off disease in humans.

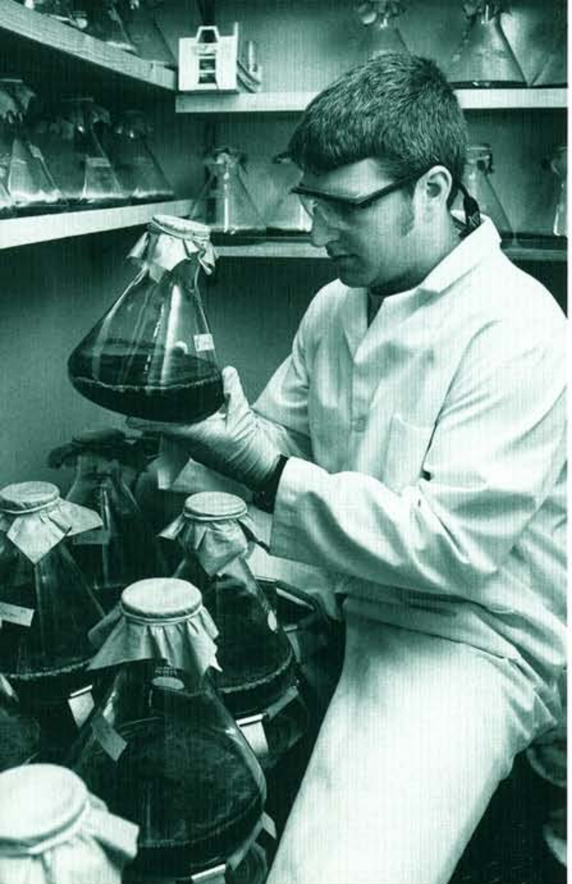
One of the first places scientists look for chemically defended species are tropical reefs, where marine animals must fiercely compete for space and nutrients.

Faulkner's team of researchers, for example, often travels to such remote places as Palau in the western Pacific to explore the island's reefs, mangrove-lined channels, and underwater caves in search of new organisms. They are particularly interested in collecting such things as sponges, tunicates, and opisthobranch mollusks (mollusks without shells).

Immediately after each dive, Faulkner's team sorts, numbers,

"IF WE GO OUT ONTO THE REEF AND WE SEE SOMETHING THAT LOOKS LIKE A LARGE CHUNK OF FOOD—POORLY PROTECTED, SOFT-BODIED, AND EASY TO GRAB—AND NOTHING IS EATING IT, THEN WE ASSUME IT HAS CHEMICAL PROTECTION."

JOHN FAULKNER



"TO BEGIN WITH IT WAS REALLY PURE ACADEMIC INTEREST IN HOW THE OCEAN WORKS,... THEN WE STARTED TO GET ALL OF THESE WEIRD AND WONDERFUL COMPOUNDS AND WE SAID, 'WHY AREN'T WE LOOKING AT THESE THINGS IN THE CONTEXT OF DOING SOME MEDICAL RESEARCH?'"

WILLIAM FENICAL

Top left, postdoctoral researcher Asfia Qureshi is a marine chemist working in the laboratory of John Faulkner (right). **Clockwise from top right,** research associates Chris Kauffman and Stephanie Lewis and graduate students David Rowley and Helene Vervoort conduct research in the Fenical laboratory.

classifies, photographs, and records each specimen in field journals. They then perform a crude screening of the organisms to determine if they contain novel chemicals that are biologically active.

Once samples are brought back to Faulkner's lab at Scripps, a small portion of each specimen is extracted using an organic solvent such as methanol. This extract is then tested in a variety of bioassays to determine whether it is effective against bacteria, fungi, or viruses. Other tests examine whether the agent shows potential for inhibiting inflammation or cancer-cell growth.

If an extract shows desired activity, the remainder of the sample is extracted and separated into its component chemicals, which are again put through a battery of tests to determine their potential against disease. These chemicals are then identified using a nuclear magnetic resonance spectrometer that gives scientists information about a chemical's physical structure.

One sponge discovered by Faulkner in Palau was found to contain a substance named manoalide, which inhibits the action of an enzyme called phospholipase A2. This enzyme plays a key role in the biochemical processes that lead to pain and swelling from inflammatory conditions such as arthritis, psoriasis, and poison oak.

Fenical also has discovered a potent anti-inflammatory agent, called pseudopterosin. The compound, which is extracted from a Caribbean sea whip, already has been incorporated into a skin cream currently being marketed as a protection against sun damage. Pseudopterosin also has been licensed to a pharmaceutical firm, which is testing it as an anti-inflammatory drug against such conditions as contact dermatitis.

The compound, developed in conjunction with Robert Jacobs, a professor of pharmacology at UC Santa Barbara, is among the University of California's top ten royalty producing inventions. It generated $686,000 in royalties in fiscal year 1994-1995.

Fenical's and Faulkner's works on marine-based pharmaceuticals stemmed from their earlier interests in the roles chemicals play in the ecology of the ocean. The scientists wanted to learn more about how marine organisms use chemical compounds to signal each other, to ward off predators, and to mate.

"To begin with it was really pure academic interest in how the ocean works, especially in highly competitive tropical environments where everything is trying to eat everything else," said Fenical. "Then we started to get all of these weird and wonderful compounds and we said, 'Why aren't we looking at these things in the context of doing some medical research?'"

The research of Fenical and Faulkner is funded by the National Cancer Institute and the California Sea Grant College System. In addition to teaming up with pharmaceutical companies, the two scientists also collaborate with Stephen Howell, a cancer pharmacologist at UC San Diego's Cancer Research Center. Howell originally conceived the idea of working with Scripps to develop new marine-based cancer drugs. While Scripps researchers would collect and screen novel marine compounds, the cancer center would perform preclinical biological research and ultimately conduct clinical trials at UC San Diego Medical Center.

"It would be a first for a college campus to develop a drug from start to finish," Fenical said. "Academic scientists traditionally do not have the capability to discover, develop, and clinically evaluate a cancer drug within the same facility."

It is hoped that the Center for Marine Biotechnology and Biomedicine will be able to expand its current working relationship with pharmaceutical companies by encouraging them to invest more heavily in marine natural products research.

Fenical believes that such a bridge between marine science and medicine could ultimately lead the way to new cures for the diseases that still plague us, such as cancer and AIDS.

"The oceans are the next great biomedical frontier—there is no question in my mind about that," he said. "The degree to which we will find cures for diseases in the world's oceans, however, will be equivalent to our ability to invest in exploring the resources they offer."

...BACK IN THE LAB

When Sir Alexander Fleming discovered penicillin in bread mold in 1928, he sparked a revolution in drug development.

Researchers began diligently sifting through soil and plants in search of new weapons to fight disease. Indeed, many of today's pharmaceuticals originated in terrestrial sources. Morphine comes from the opium poppy. Aspirin is found in the bark of a willow tree. Digitalis, a heart medication, comes from the foxglove plant. Bacteria, fungi, and other soil microbes also give rise to an army of antibiotics and antifungals used to treat everything from strep throat to pneumonia.

A rise in the number of infectious agents that are resistant to known antibiotics, however, has scientists beginning to look to the tiniest organisms in the sea as new sources of drugs.

"For 60 years, people have spent huge amounts of money scrounging through dirt to find microorganisms to produce drugs, and there are more than a hundred such drugs on the market right now," said Bill Fenical. "It is clear, however, that we need to find new resources to supplement those upon which we have traditionally relied."

While Fenical continues to direct a laboratory that concentrates on finding novel compounds in larger marine organisms, such as soft corals, he established a new lab about six years ago that focuses exclusively on microbial agents found in the ocean.

The lab, soon to be expanded and renamed the Charmaine and Maurice Kaplan Cancer Drug Discovery Laboratory, centers around two walk-in refrigerators stacked to the ceiling with large glass flasks filled with sometimes scary-looking microbes. The flasks, many of which are continually shaken on oscillating shelves to provide adequate oxygenation, contain organisms that have been singled out as showing enough potential to warrant future study. They have been selected from hundreds of specimens that are grown in the culture plates that fill the lab's shelves.

Researchers extract compounds from the flasks and pour them into tall, column-shaped beakers containing organic solvents in order to separate them into their various components. These purified chemicals are then put through a variety of tests, including a bioassay to determine if they are effective in killing human cancer cells. Those showing promise are identified using nuclear magnetic resonance spectrometry, a method that allows a scientist to identify each atom in a molecule's structure.

The enthusiasm of the dozen graduate students, postdoctoral fellows and researchers who work in the lab is unmistakable.

"We're bioprospectors," said Gil Belofsky, a postdoctoral research scientist, taking a break from working over five giant flasks filled with

Above and right, UC San Diego undergraduate Chelsea Clough at work in the Fenical Lab.

goopy-looking mold. "We're moving into the next generation of marine natural products."

Fenical stated that one of the greatest advantages of working with marine microbes rather than larger marine organisms is that microbes can be cultured and grown in the lab rather than having to be continually collected from the sea. Because pharmaceutical companies already operate large fermentation plants where they grow terrestrial microbes, it is also much easier for them to incorporate marine microbial agents into their drug development programs.

"The beauty of it is even though the terrestrial source for microorganisms is becoming very scarce, the pharmaceutical companies don't have to dramatically change what they are doing," Fenical said. "You can hand them a batch of marine microorganisms and some table salt and say, 'Just grow it in seawater and you can do everything with these the same way you have been doing it for the last 25 years.'"

OCEAN'S

Scientists have isolated many chemicals from marine organisms that hold promise as future drugs or as molecular probes, which are used in scientific research. These include

Bryostatin • isolated from a U.S. west coast bryozoan (a moss animal), it holds promise in the treatment of various types of cancer.

Cyclomarin A • extracted from bacteria found in sediments in the Torrey Pines Estuary in California, this chemical is a powerful anti-inflammatory agent.

Debromohymenialdisine • found in a very common sponge from Palau, this compound shows promise for the treatment of osteoarthritis.

Didemnin B • found in a Caribbean tunicate, it has been tested in human clinical trials for the treatment of cancer, but there are concerns that it is too toxic.

Discodermolide • taken from a Bahamian sponge, this is a powerful immunosuppressive agent that could play a role in suppressing organ rejection following transplant surgery.

Eleutherobin • isolated from a soft coral found off the coast of Australia, it shows potential as a taxol-like drug to treat breast and ovarian cancers.

A Caribbean gorgonian coral.

Manoalide • found in a sponge, this compound inhibits the action of an enzyme that plays an important role in biochemical processes that cause pain and swelling associated with inflammatory conditions such as arthritis and poison oak. It is being used as a molecular probe in biomedical research.

Pseudopterosin • found in a Caribbean gorgonian coral, this chemical is used in a cream that protects the skin from weather damage. It also is being investigated as an anti-inflammatory for use in conditions such as psoriasis and contact dermatitis.