A POCKETFUL OF GLOW

Luminous bacteria team up with deep sea fish. Biologists try to fathom the mutual benefits.

BY JULIE ANN MILLER

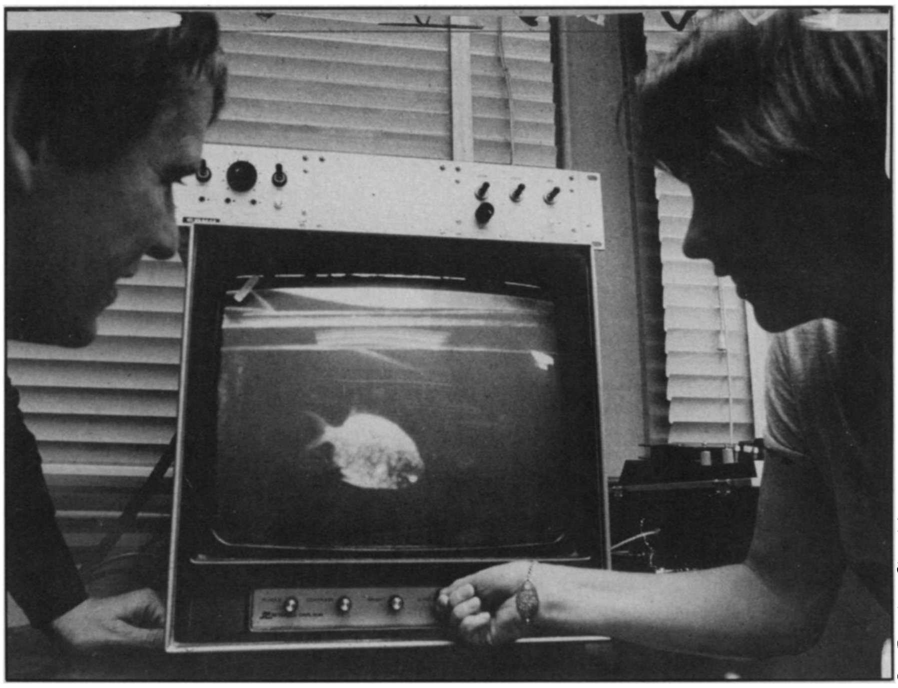
A thrifty miner might consider saving bulbs and batteries by mounting a jar of fireflies on his helmet. Perhaps if history had gone differently, and all people lived in dark underground passages, we would have struck up an evolutionary partnership with such glowing organisms. Bacteria in our intestines help digest our food, so why not packets of luminescent organisms riding on our foreheads?

Such a symbiotic situation is common among ocean fish. Much light in the depths originates from bacteria specially nurtured in fish structures called light organs. For example, the "flashlight fish" swims above reefs in the Indian Ocean and Red Sea beaming light like an automobile on a country road. The French divers who first saw it called it "le petit Peugeot."

In symbiotic associations both the host and the well-entrenched guest must enjoy some benefits. For the fish the advantages of the partnership appear numerous. The blue bacterial glow serves not only as a headlight, but also as a beacon and a shield. Multiple uses of light organs prompted scientists to title one report on the flashlight fish's behavior, "Light for All Reasons" (SCIENCE 190: 74-76).

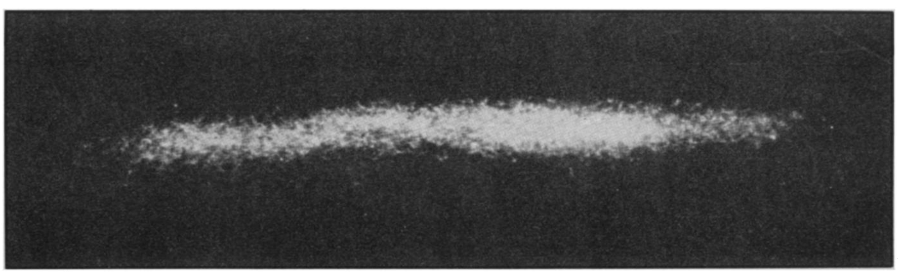
The styles of light organs among fish species vary more than fashions in car headlights. The light organs of some fish gleam like chrome along their sides or extend across the fish's belly. The light organs of the flashlight fish lie directly below each eye and a layer of skin can opaquely curtain them. The 1781 scientific name given to the flashlight fish was *Sparus palpebratus,* "the porgy with an eyelid."

One use fish have for their light organs was demonstrated in 1973 when a group of biologists studied the flashlight fish in the Red Sea and in laboratories of the Hebrew University in Elat, Israel. James G. Morin, now at the University of California in Los Angeles, and colleagues observed that in an otherwise dark laboratory, flashlight fish could capture prey by the light of their own luminous organs. Another luminescent fish, *Anomalops kaloptron,* also has light organs beneath each eye. Some specimins of this species were sent to John E. McCosker at the Steinhart Aquarium in San Fransico, but arrived in weakened condition with their lights extinguished. Those darkened fish could not visually locate brine shrimp put in their tank, until room light was added to approximate the fish's normal illumination. Then the shrimp were eaten immediately.



San Francisco Chronicle

Kenneth Nealson and Margo Haygood intensify image of glowing pinecone fish.



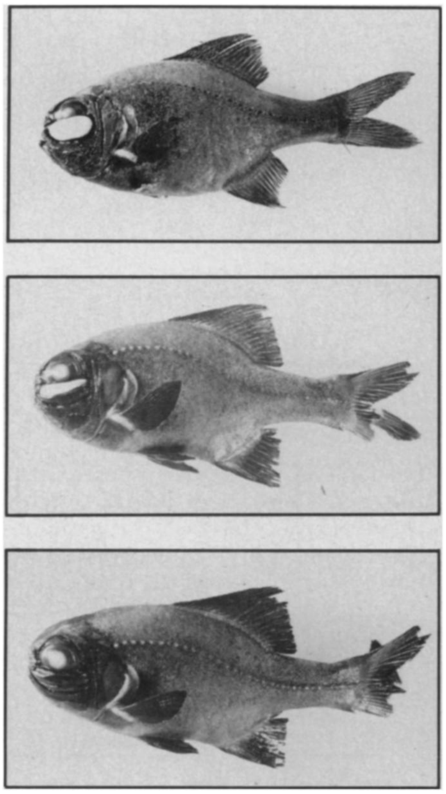
Steinhart Aquarium, San Francisco

Night view of a school of about 30 flashlight fish in the Red Sea.

A fish's light may also act as a beacon, both to attract prey and to communicate with fish of the same species. Most small crustaceans are attracted to light, so Morin and colleagues speculate that the brilliance of large aggregations of fish can attract dinner from a considerable area. The biologists also have evidence that fish respond to each other's lights. Two flashlight fish in adjacent tanks, or one fish exposed to a mirror, blink their lights more rapidly than a solitary (unmirrored) fish or a fish neighboring a non-luminous species.

The other apparent use of light is as a blinding shield. A female flashlight fish will turn off her light, swim up to an intruder and then emit a startling flash. The fish may also confuse predators by evasive swimming coordinated with the blinking light. Morin calls such behavior "blink and run." Other fish seem to use a glow as a disguise. Light of the proper intensity on a fish's belly prevents a silhouette that may attract predators from below.

The design of light organs is complex, and important aspects are not yet understood. "With canals and blood vessels and reflectors and shutters, they are as highly evolved as you can imagine," says Kenneth H. Nealson of the Scripps Institution of Oceanography in La Jolla. "Bacteria that we can't keep alive for a week in the lab, can live 20 years in the fish." Light organs are surprisingly specific as to what bacteria they will host. So far only three of the six known species of luminescent bacteria have been found in the light organs of more than 100 specimens of fish, representing 33 species. Although a sample of seawater might contain all six of these bacterial species, and seawater is continuous with the interior of the organs, each organ (and all light organs of one species of fish) contain only one species of bacteria. "Insofar as is known, there is ab-



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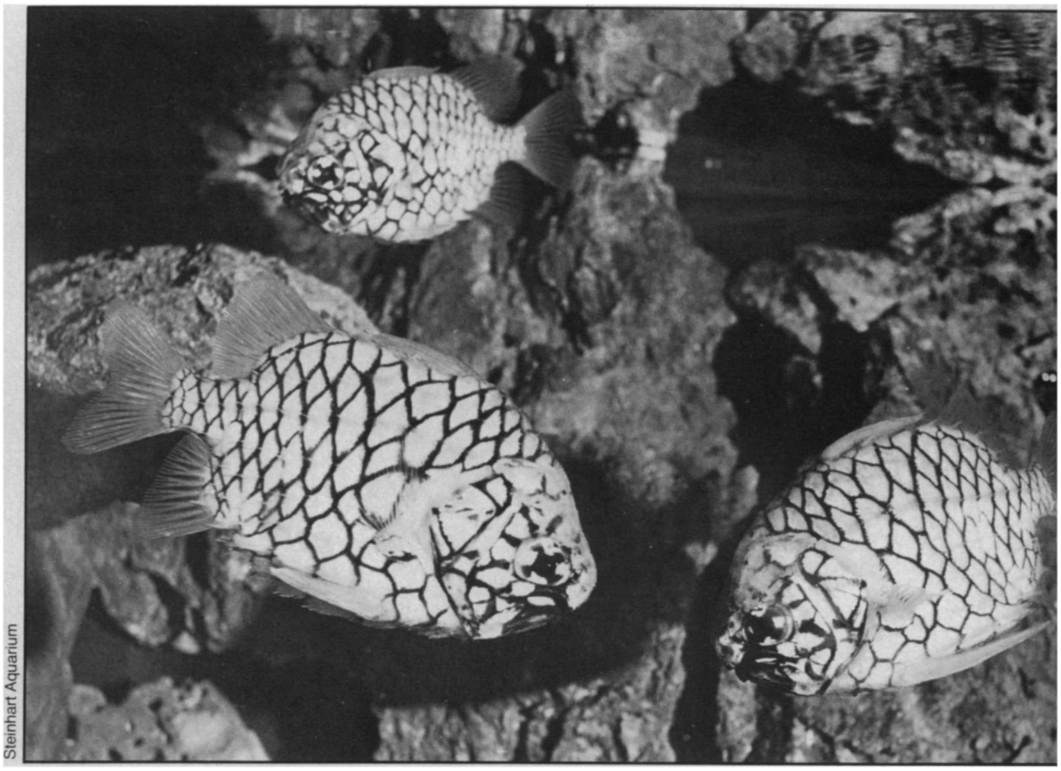
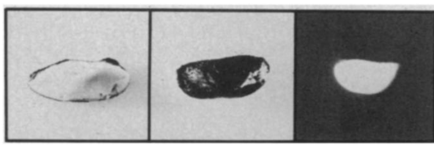


The port-and-starboard lightfish beams light from organ in its lower jaw.



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Flashlight fish controls its light by covering the continuously glowing organ with an opaque lid. Below, the outer surface of the light organ is cream-colored; the inner surface is black so as not to blind the fish. Lower right, flashlight fish photographed by the light of its own luminescent organ.

solute specificity with regard to the bacterial species cultured in the light organs of a given species of fish. Also, closely related species of fish, even when living or maintained in very different locations, appear to carry the same bacterial species," Nealson and J. Woodland Hastings of Harvard University say in a recent review (ANNUAL REVIEWS OF MICROBIOLOGY 1977, 31:549-595).

How does a bacterial species maintain exclusive rights to its comfortable quarters? At first, researchers thought that each type of luminescent bacteria might spew an antibiotic that wiped out competitors. In the first species of bacteria Nealson examined as a postdoctoral trainee in Hastings's laboratory, he found a potent inhibitor of other bacteria. This satisfying triumph was short-lived — in no other luminescent bacterium has an antibiotic been found, so the means of territorial defense remain an open question.

Answers have solidified, Nealson explains, on some other aspects of the luminescent life. For example, clues have emerged as to the benefits the bacteria receive from teaming up with fish. Nealson and co-workers have recently identified glucose as the nutrient that the bacteria receive from the fish. The fish may also utilize, and prevent accumulation of, pyruvic acid which the luminous bacteria excrete.

Nutrient exchange is not an entirely satisfactory explanation of bacterial luminescence. Many of the bacteria also live freely in the ocean. There a glow would be likely to attract predators. Outside the fish, what good could light do?

Nealson suggests two possible solutions to that puzzle. For some free-living bacteria, being eaten may be an important goal. No one yet knows how a light organ obtains its bacteria. But by becoming an attractive meal for a fish, bacteria may establish a colony in the gut, a seemingly comfortable, although less selective, niche. All six species of luminescent bacteria are frequently found in fish intestines.

Alternatively, Hastings and Nealson propose that some bacteria are simply not luminous when they are on their own in the ocean. "Chemically they are pretty smart," Nealson says. "When they get away from their host, they shut off the luminescent system. That way they don't waste energy."

A unique control mechanism seems to rule the on-off switch. Nealson calls it "autoinduction." The switch guarantees bacteria will glow only in a crowd, as in the light organ where as many as 10 billion bacteria cram a milliliter of fluid. "They turn on in a test tube, but not if you take away the walls," Nealson reports. Widely dispersed in the ocean, the "luminescent" bacteria, therefore, remain unlit.

Nealson and co-workers have discovered that the bacteria release an "inducer" chemical into the surroundings. When the concentration of that substance builds up, production of the enzyme responsible for light production begins in every bacterial cell. "Thus, autoinduction could be viewed as an 'environmental sensing' mechanism, which functions to turn off the synthesis of the luminescent system when cell density is low, when bioluminescence may have no functional importance," Hastings and Nealson explain.

Each species of bacteria that Nealson has examined has a different inducer. One mutant bacterium that is 10 times brighter than normal makes 10 times as much of the compound. Nealson and colleagues are now identifying those substances (which appear to be indole derivatives) and synthesizing them chemically. "This molecule might be viewed as a bacterial pheromone: a small molecule with a specific biological target acting from without at very low concentrations," say Hastings and Nealson.

The basic reaction that produces light in bacteria is similar to that in the firefly. (An enzyme called luciferase combines hydrogens from a flavin compound with oxygen to make water and transform an aldehyde into an acid. The energy released by the reaction is emitted as light.) Except for a possible evolutionary adaptive function, light production is not essential for the metabolism of the bacteria.

The maximum glow of a bacterium is 1,000 to 100,000 photons per second. Light from about 10^{20} bacteria would equal the

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brightness of a 60-watt bulb. This light production requires a major commitment of the cell's energy; it involves about 10 percent of the oxygen consumed by the bacterium.

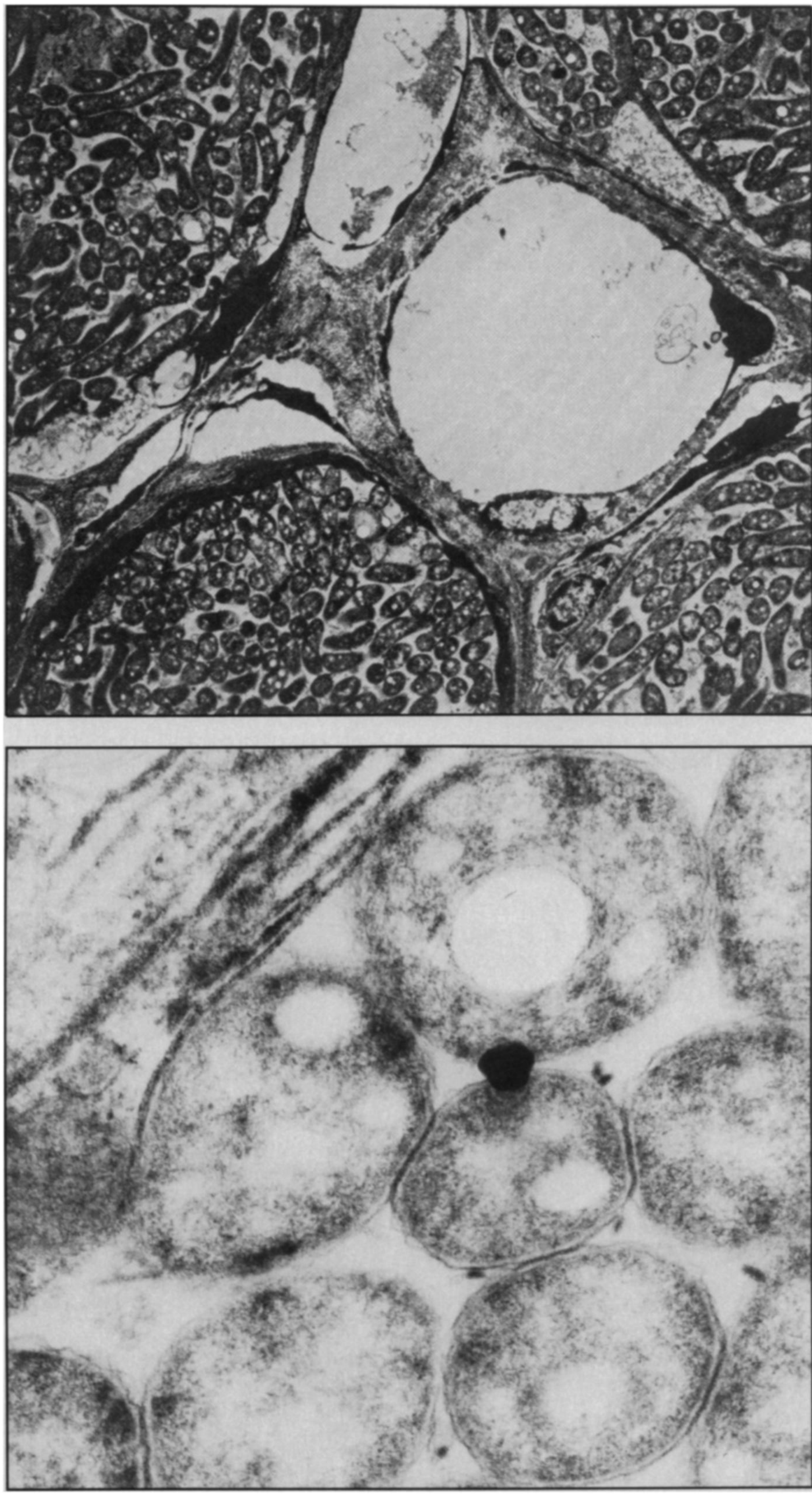
Although any of the luminescent bacteria could survive in any of the light organs, each has different requirements for making that special glow. Some of the bacteria only emit light in the presence of large or small amounts of acid. Some have high or low oxygen requirements. From examining the characteristics of each fish's organ, scientists can predict which of the bacteria would be luminescent there—and therefore which is the natural inhabitant.

Blue is the color of light given off by the several hundred thousand bacteria that have been isolated from fish and from seawater. So scientists were amazed to learn last winter of a bacterium of a different color. Edward G. Ruby, a graduate student in Nealson's laboratory, discovered a yellow-emitting bacterium while sampling seawater off the coast of San Diego. This bacterium, named Y-1, is a member of a known luminescent species, *Photobacterium fischeri,* and it produces the autoinducer typical of that species. The reactants and products of the light-producing reaction appear to be the same as in other bacteria. Although they recognize several possibilities, Ruby and Nealson believe there is an added step to the light-emission process in Y-1. Energy from the luciferase reaction, instead of being emitted as blue light, excites yet a different molecule that then emits yellow light.

The researchers suggest that perhaps all bacteria transfer energy from the reaction products to some special emitter molecule. That could explain the small variation in the exact hue of blue light emitted among bacteria.

So far no one knows whether the oceans contain fish whose light organs emit yellow light instead of blue. If such fish are found, the whole concept of symbiotic bioluminescence may have to change. Blue light travels best through water and is best seen by fish. If yellow light were commonly emitted, it would suggest either that the light has no practical use for the fish or that some fish actually can see yellow light. "This is an exciting possibility," Nealson says. "The light would be invisible to competitors. The fish would just use it for intraspecies communication." One deep sea fish, which produces red light by its own chemical mechanism, without bacteria, has set a precedent. When scientists examined those fish they found that the particular species could see red light.

Future work on these bacteria will move from biochemistry into genetic and ecological problems, Nealson predicts. "Now we have answers, but no mechanisms," he says. The discovery of marine viruses that attack one type of luminescent bacteria, and that can package bacterial DNA and carry it from one cell to another, already has allowed location of a few bacterial genes. The researchers plan eventually to identify all the genes and control mechanisms involved in bioluminescence. On a more general level, the researchers are turning enthusiastically to the relationship between the bacteria and their hosts. "There are several years of work yet on the molecular mechanisms," Nealson says, "but everyone wants to work on symbiosis." □



Steinhart Aquarium

Luminous bacteria crowded in the organ of a flashlight fish. The central vessel provides nourishment from the fish to the bacteria. The magnification, by electron microscope, is 5300. Below, a close-up view of light-emitting bacteria, Photoblepharon palpebratus.