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The color of blood: Pigment helps stage symbiosis in squid

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New findings about the Hawaiian bobtail squid help reveal some of the hidden rules of symbiosis, processes that are also likely occurring in higher animals, including humans.

Photo: Chris Frazee, UW School of Medicine and Public Health

The small but charismatic Hawaiian bobtail squid is known for its predator-fooling light organ.

To survive, the nocturnal cephalopod depends on a

symbiotic association with a luminescent bacterium that gives it the ability to mimic moonlight on the surface of the ocean and, in the fashion of a Klingon cloaking device, deceive barracuda and other fish that would happily make a meal of the small creature.

The relationship between the squid and the bacterium *Vibrio fischeri* is well chronicled, but writing in the current issue of the journal Proceedings of the Royal Society B, a group led by University of Wisconsin-Madison microbiologists Margaret McFall-Ngai, Edward Ruby and their colleagues adds a new wrinkle to the story.

"The squid has seen an opportunity to recruit an organism to make light," explains McFall-Ngai, a UW-Madison professor of



Margaret McFall-Ngai



Edward Ruby

medical microbiology in the <u>School of Medicine and Public Health</u>. "But to do that you have to tame it. You have to train it to do what you want it to do."

In the case of the bobtail squid, it seems that the blood pigment hemocyanin plays a dual role in helping the squid recruit and sustain the bacterium it uses to avoid predation.

Like its human analog hemoglobin, hemocyanin is primarily responsible for transporting oxygen from the squid respiratory system to the rest of the body. But the hemocyanin protein also appears to be deployed in a way to help the squid recruit its population of *Vibrio fischeri*, which the squid flushes and replenishes on a daily cycle to enable its nocturnal defenses.

"In the early events of symbiosis, hemocyanin appears to have antimicrobial activity," says Ruby, also a UW-Madison professor of medical microbiology and a co-author of the new report. "We think it is part of the mechanism by which *Vibrio fischeri* become specific."

In essence, the squid is using the antimicrobial properties of hemocyanin to weed out competing bacteria so that only the glowing *Vibrio fischeri* can colonize the surfaces of the crypts that compose the squid's defensive light organ.

The new findings help reveal some of the hidden rules of symbiosis, processes that are also likely occurring in higher animals, including humans. How Vibrio fischeri
copes with the
antimicrobial properties
of the squid's blood
pigment is an
unanswered question,
but agents with
selective antimicrobial
activity are not
unknown: "People tend

to assume that whenever they encounter an antimicrobial, it is meant to kill everything," Ruby notes. "Here, we know some bacteria are being courted."

The second role played by the hemocyanin protein in helping to establish symbiosis is more in keeping with its traditional function of ferrying oxygen. The oxygen-transporting properties of hemocyanin are exploited by the squid as the symbiotic population of *Vibrio fischeri* requires lots of oxygen to fuel the chemical reaction that causes the microbe to light up in the dark. The animal seems able to direct its symbionts to modulate the acidity of the crypts where they take up residence, creating oxygen rich niches at night while suppressing the flow of oxygen during the day when the squid has no need of its companions' glow.

"At night the squid is creating an environment that is more acidic, where oxygen is more easily dumped," says Ruby.

"Oxygen is really pivotal," adds McFall-Ngai.

"There is a lot of energy that goes into making light."

The new findings, according to the Wisconsin biologists, help reveal some of the hidden rules of symbiosis, processes that are also likely occurring in higher animals, including humans, who also depend on microbes to perform critical services.

"There is a dynamic interplay in symbiosis," says McFall-Ngai, who designed and performed the study with UW-Madison post-doctoral fellow Natacha Kremer, the lead author of the Proceedings of the Royal Society B report. "In humans, there is an ecological succession in microbiota. What we are looking for in our model are the general themes."

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