# Leafy green "solar-powered" sea slugs begin to reveal their true colors

By Ferris Jabr, Scientific American 11.22.15

Pictured is the emerald sea slug, Elysia chlorotica. Photo: EOL Learning and Education Group, via Flickr

Nature is full of thieves. Instead of laboriously collecting pollen and nectar from flowers, robber bees raid the hives of other pollinators and steal the honey within. Some ant species routinely kidnap and enslave members of neighboring colonies, taking tens of thousands of pupae in a single season. And massive-winged frigate birds—also called pirate birds—swoop down on smaller seabirds and snatch the fish out of their beaks, or harass their victims until they regurgitate their latest catch.

Sea slugs dubbed sacoglossans are some of the most remarkable biological burglars on the planet. About the length of a postage stamp or two, these slugs feed on algae by sucking all the delicious gelatinous cytoplasm and crunchy protein nuggets out of the underwater plants. In the process, they slurp up algal chloroplasts, also known as plastids—green jelly bean-shaped organelles that perform photosynthesis, capturing the sun's energy and combining it with carbon dioxide and water to make food. Most sap-vacuuming slugs digest the chloroplasts right away, but some species store the plastids for weeks to months in large transparent digestive glands, turning the animals brilliant shades of green.

For the past several decades, scientists thought that the slugs hoarded chloroplasts in order to become solar-powered animal-plant hybrids. It made a lot of sense. Experiments confirmed that, when placed in the light, the slugs actively absorbed carbon dioxide, just like leaves. And some of the slugs could stay alive in a well-lit laboratory for months with no food. Besides, why else would the animals go to the trouble of preserving the chloroplasts, if not as a long-term energy source?

Two studies published in the past few weeks are forcing scientists to completely rethink solar-powered sea slugs, however. In the first study, Sven Gould of the University of Dusseldorf and his colleagues deprived six sacoglossans of food for 55 days: two enjoyed 12 hours of light followed by 12 hours of darkness each day; two lived with the same cycle, but received a drug that disrupted photosynthesis; and the remaining two slugs lived in continuous darkness. At the end of the starvation period, all the slugs were alive, equally pale and all had lost about the same amount of weight. Whether they could photosynthesize had not made a difference to their health and survival.

“It’s very appealing to think that here you have an animal slowly turning into plant,” Gould says. “But now we’re back at square one.” Gould has proposed that, rather than becoming part vegetable, the slugs are doing something lots of critters do: saving some of the food they collect for times of hardship. Some sacoglossans essentially hibernate in the winter, burrowing into sediment. Sunlight would not help them endure this subterranean slumber, but a digestive sac full of nutritious chloroplasts to munch on would do the trick.

That would also explain how the slugs in his experiment made it through nearly two months of perpetual night without any food and why all the slugs were pale and shrunken after the ordeal.

Even if these slugs are not relying primarily on photosynthesis for nutrition, Gould's recent experiment does not completely rule out the possibility that they get some small amount of energy in the form of secreted sugars from chloroplasts.

They might even use some of the oxygen produced by the organelles, the same way the spotted salamander evolved a symbiotic relationship with algae that live in its eggs: the salamander embryos breathe in the oxygen the algae release while the encapsulated plants enjoy the animal's exhaled carbon dioxide.

Setting aside the seductive idea of a plantimal suddenly brings into focus other explanations for the slugs' verdigris. Maybe some of the mollusks are camouflaging themselves—after all, they spend an awful lot of time hanging out on green plants. Or maybe green is to these slugs as pink is to flamingos: sex appeal. Flamingos get their red and pink pigments from the carotenoid proteins in the plankton they eat. Healthier, well-nourished flamingos are pinker and more attractive to mates. Perhaps sea slugs ostentatiously display chloroplasts in a similar come-hither way.

Equally mysterious to the benefit of storing chloroplasts is how the slugs delay digestion in the first place. Do their digestive enzymes identify and ignore chloroplasts, but break down all the other part of the algae? Or do the slugs have a kind of gut sanctuary that is physically separated from the rest of the digestive tract? Both Gould's experiment and earlier studies have affirmed yet another mystery: in the slugs' guts, the chloroplasts keep photosynthesizing—something that should not be possible.

More than 600 million years ago, chloroplasts were free-living photosynthetic cyanobacteria often swallowed and digested by larger cells. At some point, instead of eating the cyanobacterium right away, one of those larger cells allowed it to hang around as a kind of in-house chef. Eventually, cyanobacteria became permanent residents of the cell and lost a lot of their autonomy.

Even today, however, chloroplasts have their own DNA, though their genomes are not as large and complex as they once were. In order to photosynthesize, the chloroplasts inside an alga depend on many genes in the alga's own nucleus and the proteins for which they code. Tearing chloroplasts out of algal cells and asking them to make food inside a slug's gut is like expecting the bottom half of a blender to puree some carrots sans the blade and glass jar.

A few years ago, scientists thought they had started to crack this conundrum. In 2008, Mary Rumpho of the University of Connecticut and her colleagues discovered that an especially chartreuse species of sea slug, Elysia chlorotica, had taken at least one of the chloroplast-maintenance genes from the alga's nucleus and woven it into its own slug genome. This fascinating and rather unique instance of horizontal gene transfer could potentially explain how the slugs kept the chloroplasts running.

More thorough follow-up experiments snuffed out this spark of an idea, however. As described in a 2011 study, Gould and his teammates could not find any trace of active algal nuclear genes in the green sea slugs. Further investigations by Rumpho—including one with sea slug embryos that had never been exposed to algae, and therefore could not be contaminated with plant DNA—confirmed the absence of active algal genes in the slugs' genomes.

Perhaps her initial tests detected partially digested algal genes that were in the slugs' bodies, not incorporated into their own DNA. Or maybe a virus with which the slugs are almost always infected was harboring stolen algal genes. Either way, "I don’t think there's any gene transfer into the chromosomes of the animals," Rumpho says. And even if some DNA hopping has happened, researchers now agree that horizontal gene transfer does not account for the chloroplasts' longevity, adds Gould.

In another recent study, however, Gould and his colleagues think they have hit upon the beginnings of a new explanation. The fact that chloroplasts rely on genes in the nucleus of plant cells is true for those organelles in ocean algae and those in more recently evolved land plants—but it may be less true for the former. Gould and his team compared chloroplast DNA from some of the algae that green sea slugs eat to chloroplast genomes from land plants. Unlike the organelles in plants on terra firma, many of the algal chloroplasts contained a gene that coded for a very important protein named ftsH—a kind of onboard mechanic, as Gould puts it.

One reason modern chloroplasts depend on proteins encoded in nuclear DNA is to continually repair the molecular damage they sustain during the arduous process of photosynthesis. Because some algal chloroplasts can make at least one of these damage control proteins themselves, they could theoretically survive for a while even when ripped from algal cells.

Admittedly, Gould says, "It is not going to be this gene alone. There will be other mechanisms—and the slug will need to provide the right biological milieu, such as neutral pH—but this protein is a very very important player. Deleting this single gene in higher plants is lethal for the plastids."

If algal chloroplasts are in fact more independent than scientists realized—if, unassisted, they can make other proteins like ftsH that keep all their parts in working order—then the relationship between sacoglossans and chloroplasts may not be as straightforward a case of theft as it once seemed.

Perhaps, once consumed, the organelles continue to take care of themselves the way they would inside an alga—or reflexively trigger certain defenses that help them survive in a hostile environment—and the slugs take advantage of this self-sufficiency to enjoy a few weeks or months of complimentary sugar and oxygen. Alternatively, the chloroplasts might consent to keep busy because they are getting carbon dioxide or something else out of the deal, like the algae living in the salamanders' eggs—until the slugs double-cross them and digest the plastids properly. Or maybe the kidnapped organelles have developed Stockholm syndrome.

Ongoing research in Rumpho's lab further calls into question who really has the power in this hostage situation. E. chlorotica, the shimmering emerald slug species that Rumpho studies, seems to be so dependent on algae for survival that if baby slugs are not exposed to algae for at least seven days, they stop growing and die. It’s almost as if these sea slugs have evolved to self-destruct if there is no algae around in their youth, because without the plants and their chloroplasts they simply would not make it.

Rather than producing sugars, the chloroplasts E. chlorotica captures from its algae primarily make lipids, which provide a source of energy as well as building materials for cells. A young slug needs a lot of lipids to grow rapidly. What if this one sacoglossan has outsourced its lipid production to chloroplasts? If so, captive chloroplasts would be so much more than a postponed meal for the slugs—they would be more like a continually renewed vital organ.

Vocabulary

- Chartreuse: a variable color averaging a brilliant yellow green
- Verdigris: a green or greenish-blue poisonous pigment
- Terra firma: dry and solid ground as compared to air or water
- Milieu: in this case; environment. Also; the physical or social setting in which people live or in which something happens or develops

1. Based on Sven Gould's experiment (where he kept slugs in different conditions), provide an example of a hypothesis he may have come up with before doing the experiment. Provide an example hypothesis he may have come up with after the experiment (after he realized how the slugs reacted).
2. Why would it be advantageous for a cell to keep a photosynthetic cyanobacteria inside of itself instead of digesting it?
3. What is meant by "horizontal gene transfer"? How would you define vertical gene transfer?
4. Why would a young slug need sugars and lipids to develop?
5. Out of the several possible hypotheses presented to explain why the slugs contain chloroplast, which seems the most plausible?