

Genetic probes give new clues about the stunning diversity of comb jellies

September 1 2021, by Raúl Nava



Scientists have described around 200 comb jelly species so far. They come in an assortment of shapes, sizes, colors, and patterns. Some are small, while others like this giant comb jelly (Aulacottena sp.) can be quite large—growing larger than a football. All play an integral role in ocean ecosystems. Credit: MBARI



Comb jellies—known to scientists as ctenophores (pronounced "teen-ohfours")—mesmerize with their beauty, but these captivating creatures remain poorly studied due to their delicate nature. MBARI researchers have used the power of genetics to learn more about these animals.

In a study published online earlier this summer in *Molecular Ecology Resources*, MBARI researchers Lynne Christianson, Shannon Johnson, Darrin Schultz, and Steve Haddock examined a specific gene sequence in <u>comb</u> jellies. This sequence has revealed untold diversity within this group of animals.

"Using genetics, we discovered surprising diversity in some groups, including some more commonly-seen species that were previously considered a <u>single species</u>, but are now revealed to be multiple species," said Lynne Christianson, lead author on the study and a senior research technician at MBARI. This research also uncovered several comb jelly species that are new to science.

This work—funded by the David and Lucile Packard Foundation, the National Science Foundation, and the National Institutes of Health—opens the door for future research on comb jellies using eDNA, or environmental DNA. eDNA holds promise for detecting marine animals from the drifting bits of genetic material they leave behind in seawater. "Importantly, the genetic data we have shared to public databases will provide a valuable reference for others using eDNA to help reveal the complexity of ocean ecosystems," said Christianson.

Comb jellies live throughout the ocean, from the shallow depths to the deep seafloor and from warm tropical seas to chilly polar waters. Although they are an important part of marine ecosystems, the challenge of collecting intact specimens, especially from the deep sea, makes them difficult to study. "Most scientists are not able to collect comb jellies in any recognizable manner—they are just too fragile," explained MBARI



Senior Scientist Steve Haddock.

But MBARI is uniquely equipped to study these delicate drifters.

MBARI's research vessels offer a platform for blue-water diving—a form of scuba diving far offshore suspended in <u>open water</u>—and deployment of specialized robotic submersibles called remotely operated vehicles (ROVs).

Using both scuba and submersibles, MBARI researchers have been able to carefully collect comb jellies for research in the lab. For decades, they have amassed a collection of specimens representing nearly every known family of comb jellies.



Blue-water diving is one of several techniques used by MBARI researchers to collect delicate ctenophores. Credit: Steve Haddock © 2018 MBARI



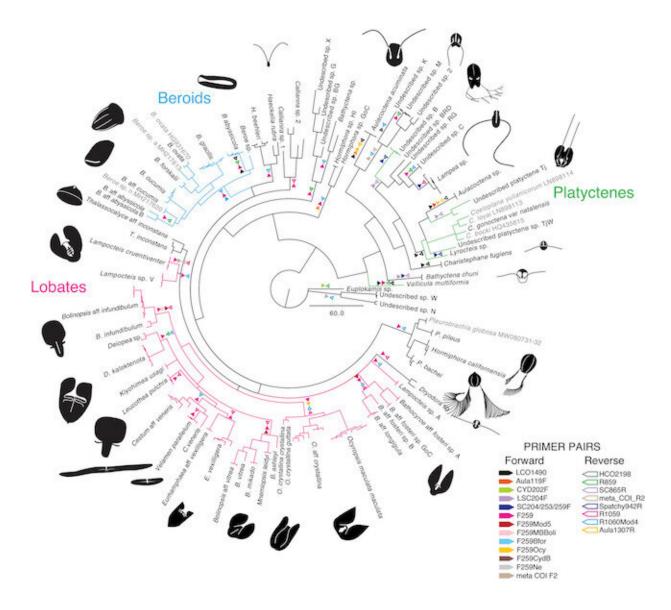
Studying these animals from their appearance alone has been valuable, but has at times been an imperfect science. Some specimens are damaged, some distinguishing characteristics between species are cryptic, and some tissues are too delicate for preservation. In addition to examining the appearance of live animals, MBARI researchers have turned to genetics to identify and catalog their specimens.

The mitochondrial gene cytochrome-c-oxidase subunit I (COI) is like a genetic fingerprint for animals. This gene is widely used in everything from bees to baboons for distinguishing species and even geographic subpopulations. Building a library of ctenophore "fingerprints," though, was no easy task.

To read these genes, researchers use primers—short, manufactured DNA pieces that complement segments of DNA found in a species' genome and serve as anchor points to start genetic sequencing. The DNA of comb jellies is so different from other animals that the standard primers do not work for most comb jelly species. The team set out to solve this problem. First, the team had to make primers that would work on comb jelly COI sequences. They did this by examining the genomes of comb jellies and testing hundreds of primer combinations. Then, they used those primers to generate the library of individual sequences from across hundreds of comb jelly specimens they had collected. These are the two most impactful outcomes of the work.

The team created the most complete library of DNA sequences from comb jellies, adding 72 species to the global database where only 15 had been represented before. By comparing sequences of the COI gene, they also built a more accurate family tree for comb jellies at the species level.





MBARI researchers prepared a "tree" of relationships using COI sequences to show species differences between comb jellies. They used this tree to help other researchers determine which DNA primers would work best for a group of interest. Major branches include the lobed comb jellies (pink), beroids (blue), and the seafloor-dwelling platyctenes (green). Credit: Christianson et al. 2021 Molecular Ecology Resources

This new library of genetic information helped clarify relationships between similar-looking species. The genetic probes have sparked



several new questions about common species.

MBARI researchers frequently observe the scallop comb jelly (Bathocyroe sp.) off the central California coast. But a closer look at its COI gene suggested three distinct species live off our coast. Even more intriguing? None seem to align with the three known species in the genus. Slight differences in appearance and depth distribution from the recognized species of Bathocyroe suggest the three species off California's Central Coast may be new to science.

MBARI's ROVs have been instrumental in revealing the untold diversity of the ocean's midnight zone. In 34 years of research, MBARI has documented more than 200 species previously unknown to scientists. One such find was the bloody-belly comb jelly (Lampocteis cruentiventer). MBARI scientists George Matsumoto and Bruce Robison contributed to the original description of this species in 2001. Researchers sometimes see individuals with the same body shape, but lacking the signature scarlet color. One variant has an amber appearance, while another deeper-dwelling one is brilliant magenta. Looking at the COI gene has confirmed scientists' suspicions that these are not unique color morphs but likely separate species.

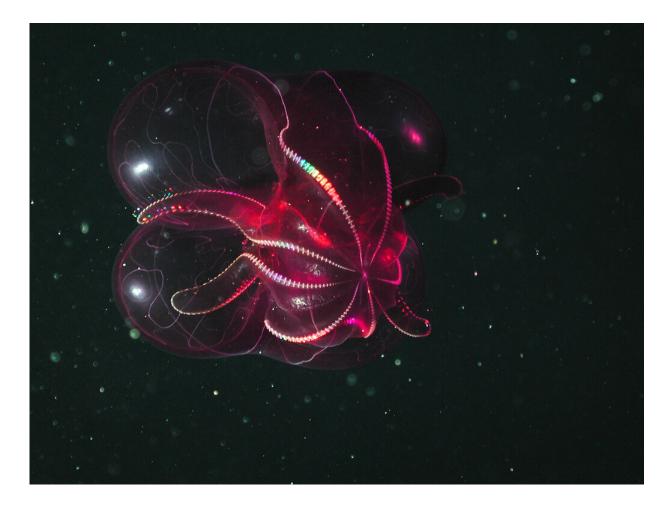
"Everywhere that we have been able to look in detail has revealed unseen diversity, suggesting there are even more species of ctenophores than our most optimistic estimates," said Haddock.





Comparing COI gene sequences revealed two branches of the family tree for the lobed comb jelly genus Bolinopsis. One branch consisted of four tropical species and the related warty comb jelly (Mnemiopsis leidyi), while another included two species from cooler temperate waters. Credit: MBARI





The bloody-belly comb jelly (Lampocteis cruentiventer) is one of the most stunning comb jellies in the depths of Monterey Bay. Sometimes MBARI's ROVs encounter an amber-colored variant (previous) and a deep-dwelling purplecolored variant (pictured). New genetic analysis has confirmed scientists' suspicions that these two are not strange color morphs of the crimson ctenophore, but in fact distinct species. Credit: MBARI

Examining comb jellies collected from more locations around the globe will be a critical next step for understanding relationships among these animals by comparing the sequences of potentially undescribed species to sequences from species formally recognized by scientists. The COI gene provides a useful starting point for researchers to focus their work.



This work has laid the foundation for scientists around the globe to read the genetic fingerprints of comb jellies left behind in eDNA and improve efforts to catalog the diversity of life in the ocean. Comb jellies have largely been excluded from the promising new field of rapid species identifications because of the scarcity of COI gene sequences for this group necessary to detect the eDNA they leave behind.

The MBARI research team has now provided tools for other scientists to begin rapid identifications of <u>comb jellies</u>. Samples from MBARI and our collaborators have added five times as many comb jelly species to the National Center for Biotechnology Information archives as have been previously sequenced. "We are providing genetic sequences for an unprecedented number of ctenophore <u>species</u> into public databases for all to use," said Christianson.

Who knows what new discoveries await now?

More information: Lynne M. Christianson et al, Hidden diversity of Ctenophora revealed by new mitochondrial COI primers and sequences, *Molecular Ecology Resources* (2021). DOI: 10.1111/1755-0998.13459

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