

Migration and Microbiome in *Botrylloides diegensis*

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Botrylloides diegensis are marine invertebrate ascidians of the Tunicata taxon, in the order *Stolidobranchia*, family *Styelidae*. Ascidians can be colonial or solitary. Colonial ascidians, like *Botrylloides diegensis*, may consist of up to several hundreds of adults, called zooids, which organize into groups of interconnected animals known as systems, the set of systems is called a colony. Each zooid is an independent animal with its own organs, including a heart, a digestive system, and a neural gland. Colony of *B. diegensis* share an external vascular system, which is embedded in the colony's tunic. These external haemolymphatic vessels are lined by a layer of epithelial cells and terminated by contractile blind-ends called ampullae. *B. diegensis* have a biphasic life cycle, as a free-living larva and as a sessile colony attached to some substrate. During their larval stage, ascidians have a tail, a notochord, and a neural tube, which all disappear during their metamorphosis into an adult. These characteristics are typical of chordates, which places the tunicates among the invertebrates most closely related to vertebrates.

Although *B. diegensis* are described as sessile, we have nevertheless suspected movements of colonies in our laboratory, which is why I decided to study their migration in more details. During my experiments, I also observed that colonies cultured in freshly prepared artificial seawater quickly underwent a stress reaction known as regression. I suspected that the absence of a suitable microbiome in the seawater was blocking their development. *B. diegensis* have internal and external bacteria that can interact with them and seems to influence their survival. The bacteria floating in their environment can also have an influence on their survival, as they filter the water to feed themselves. This is why I decided to study the environmental microbiome of *Botrylloides diegensis* as a second topic.

Overall, in my master thesis I use macrophotography and timelapse combined with image analysis to demonstrate that *B. diegensis* migrate consistently upwards, against the direction of gravity, at an average speed of 1.15 mm/d. I used derivatives of the first experiment and found clues as to why and how *B. diegensis* migrate. Then I use DNA/RNA extraction and Sanger sequencing to determine the type of bacteria found in the microbiome of *B. diegensis*. At the end I inoculate some aquaria containing colonies of *B. diegensis* to show that the environmental microbiome seems to have a positive or negative influence on the survival of the animals, depending on the concentration and the specie for the bacteria.

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