Examining *Daphnia* Fitness in Response to an Unknown Parasite

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Daphnia are planktonic crustaceans commonly found in ponds that play a crucial role in the aquatic food chain. These zooplankton are primary consumers of algae and a keystone species—making them the perfect species to study in terms of larger scale ecological effects. Additionally, *Daphnia* are commonly used as a model system in ecology and evolutionary biology. They can reproduce asexually which allows us a genetic constant while studying their traits (Ebert 2022). Their asexual reproduction also allows researchers to maintain populations of *Daphnia* from multiple sampling years. The Rogalski lab studies a population of *Daphnia ambigua* living in Sewall Pond (Arrowsic, ME). In previous years, the Rogalski lab has focused on salinity as a stressor on these microscopic organisms. My summer research focused on a different stressor—a parasite infecting the *Daphnia* gut.

At Sewall Pond we sampled the water collum twice a week, collecting *Daphnia ambigua* and data on the environmental conditions of the pond. Early this summer, we found a prevalent gut parasite in the *Daphnia ambigua* population. Around the halfway point of my 8-week fellowship, we saw a drastic decrease in the Sewall *Daphnia* population: which reached zero by the end of June. There were multiple stressors present in the pond that contributed to this rapid decline: the predation of Chaoborus larvae (an insect in the midge family), the predation of juvenile alewife fish, and this unknown gut parasite. Research fellow Eva Ahn (2026) and I wanted to focus our attention on identifying the parasite. We speculated that the parasite might be similar to *O. paujunii,* a parasite studied by Marcin Dziuba et al. (2024). We aimed to answer the following questions in order to identify the parasite and understand how it can be used in the lab:

1. How can we infect *Daphnia* with the unknown parasite in the lab?
2. How virulent is the unknown parasite?
3. How does the parasite affect genetically different *Daphnia*?

In late June and early July, we conducted two pilot trials to determine how we should approach infecting *Daphnia* in future experiments. We infected *Daphnia ambigua* with methods used by Dziuba's lab, which involved exposing healthy *Daphnia dentifera* to a 'slurry' of ground up highly infected *Daphnia* in a small concentration of pond water. In our first trial, we found this method to be too virulent in *Daphnia ambigua* under 24 hours old (at time of infection). Most of the *Daphnia* we infected with this treatment died before we could analyze the animals for infection status. However, in trial two, the 'slurry' method proved to be effective for our purposes of analysis when we infected older *Daphnia* (2 or 4 days old at time of infection). During our first pilot trial, we also tested infecting healthy *Daphnia* by exposing them to 'yucky water': water containing waste and spores from an infected animal. In our pilot trial, the 'yucky water' treatment infected animals and allowed us to study them prior to death.

Using what we learned from our pilot trials we designed an experiment to determine how genetically different *Daphnia* might differ in their reaction to infection. We used the 'yucky water' treatment to infect clones that were 24 hours old. Our preliminary results suggested that the spore density of our 'yucky water' treatment was likely higher and therefore more virulent when exposed to *Daphnia*. However, some infected *Daphnia* survived the duration of the experiment (12 days).

Overall, the preliminary work we did this summer will allow us to design future experiments involving our unknown gut parasite. We are continuing our work to identify this parasite by imaging it and sequencing its genome, and we will continue developing a way to standardize spore density to infect *Daphnia ambigua* in the lab.

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References

Ebert, D. (2022). Daphnia as a versatile model system in ecology and evolution. EvoDevo, 13. https://doi.org/10.1186/s13227-022-00199-0

Dziuba MK, McIntire KM, Seto K, Davenport ES, Rogalski MA, Gowler CD, Baird E, Vaandrager M, Huerta C, Jaye R, Corcoran FE, Withrow A, Ahrendt S, Salamov A, Nolan M, Tejomurthula S, Barry K, Grigoriev IV, James TY, Duffy MA.2024.Phylogeny, morphology, virulence, ecology, and host range of Ordospora pajunii (Ordosporidae), a microsporidian symbiont of Daphnia spp.. mBio15:e00582-24.https://doi.org/10.1128/mbio.00582-24