Causes and consequences of sex in a natural *Daphnia* population
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Daphnia are a genus of freshwater crustacean zooplankton that are both crucial to food webs in natural lake ecosystems and serve as prevailing model organisms in the lab. They have evolved a system of dual-reproduction called *cyclical parthenogenesis* that allows them to strike a beneficial balance of energy efficiency and survival as the seasons change around them. In the spring and summer, *Daphnia* reproduce asexually, with mothers producing large clutches of eggs which are all genetic clones. These clones develop quickly and can then asexually clone themselves, rapidly expanding *Daphnia* populations in a lake[1]. Because environmental conditions are fair during these seasons, populations can survive without possessing the genetic diversity that comes with sex. However, as it shifts to fall and winter, it becomes crucial that the population adapt to and survive the worsening conditions. *Daphnia* moms begin producing a mix of females and males and switch to a mode of sexual reproduction, which introduces greater genetic variability and ensures that at least *some Daphnia* offspring will be hardy enough to survive[2] (Figure 1).

In the lab, 3 stressors are known to reliably induce *Daphnia* sex: decreasing day (daylight) length, food limitation, and overcrowding of animals[3]. Moving to the field, these dynamics become much less clear, and other additional factors seem to have various impacts on sexual reproduction depending on the specific species and ecosystems examined[4]. During my 10-week summer fellowship, I monitored a population of the understudied *Daphnia ambigua* in local Sewall Pond (Arrowsic, ME) to better understand their reproductive cycle and the environmental triggers that may underlie it.

This work involved the collection of live *Daphnia* samples twice a week and a visual assessment of their reproductive status, infection status, and fitness. Each sampling visit, I also collected quantitative data on the changing environmental conditions of Sewell Pond (temperature, dissolved oxygen levels, light penetration, salinity, pH, algae biomass, etc.). I observed that from mid-May to mid-June, a significant portion of the *D. ambigua* population produced males and shifted to sex (Figure 2). During this time period, there were no correlating trends in day length, food limitation, or crowding (Figure 2); instead, I examined 4 additional stressors that might have been at play.

Over the summer season, we observed disease epidemics of the bacterial parasite *Spirobacillus* and an unidentified microsporidian gut parasite[5]. The peaking in prevalence of the gut parasite lines up with the shift toward sex in our population (Figure 3), and preliminary lab experiments (conducted by Eva Ahn '26 and Sage Garver '27) suggest that the parasite has negative impacts on fitness. This parasite possesses the traits required to warrant an evolved *Daphnia* sex response; we are continuing to perform lab experiments this fall to determine whether chronic parasite exposure can reliably induce sex[6].

While the gut parasite seems to be a likely culprit of our sex trends, I am continuing my analysis this fall of the 2 remaining stressors: the predation of Daphnia by alewife larvae and by *Chaoborus* (a fly larvae). Alewife moms are released into Sewall Pond each spring, and when they reproduce, their larvae are known to feed on *D. ambigua*. I plan to connect with local samplers to better estimate the timing of this process, especially as it relates to our observed sex trends. Finally, larvae of the midge fly *Chaoborus* are known to feed on *Daphnia*, although the timing of the *Chaoborus* life cycle in Sewall seems to be out-of-synch with our observed *Daphnia* sex trends. *D. ambigua* are known to develop spiky, helmet-like morphologies as a deterrent in response to this predation, and the size of these helmets can be used as a proxy for the strength of predation pressure. I am currently measuring our preserved *Daphnia* samples to quantify *Chaoborus* predation pressure and statistically support the lack of correlation that we observed.

In addition to work on predatory stresses, I am continuing work this fall with Eva Ahn '26 and Sage Garver '27 to identify and characterize the gut parasite, including sequencing its genome and running lab experiments to quantify its negative impact on *Daphnia* fitness. I hope that my resulting honors thesis can elucidate both the characteristics of this undiscovered microsporidian and the broader impacts that disease stressors can have on the function of complex life cycles in *Daphnia*.

Figure 1. Schematic of *Daphnia* life cycle. Not noted in my in-text summary is that during sexual reproduction, *Daphnia* moms produce resting eggs (*ephippia)* that experience a period of diapause before hatching instead of live-hatching like asexual eggs.

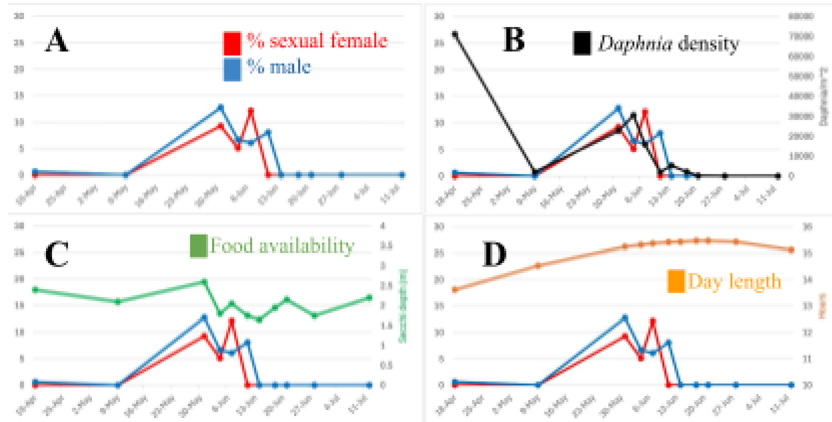

Figure 2. Sexual *Daphnia* females and males emerge in mid-May, peaking in late-May/early-June (A). In natural populations, shifts toward sex are known to "max out" at around 10-20% of the population[1]; in other words, this shift towards sex is significant. There is observed correlation between sexual reproduction and increasing *Daphnia* density (B), decreasing food availability (C), or decreasing day length (D).

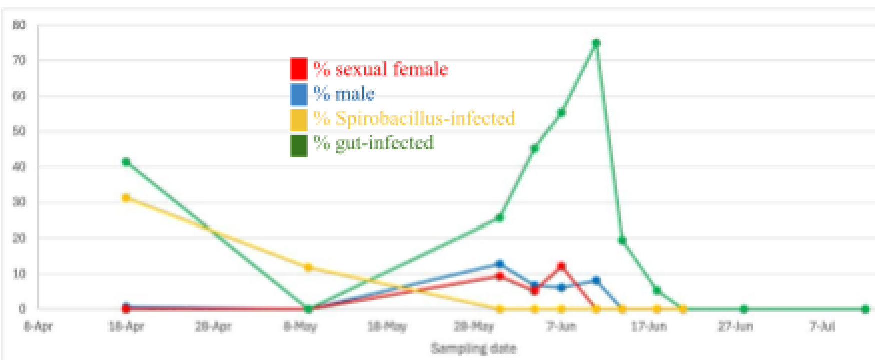

Figure 3. Observed correlation between sexual reproduction and prevalence of the unidentified microsporidian gut parasite.

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