Could microbes be both rapid degrader and key constituent of predator risk cues?

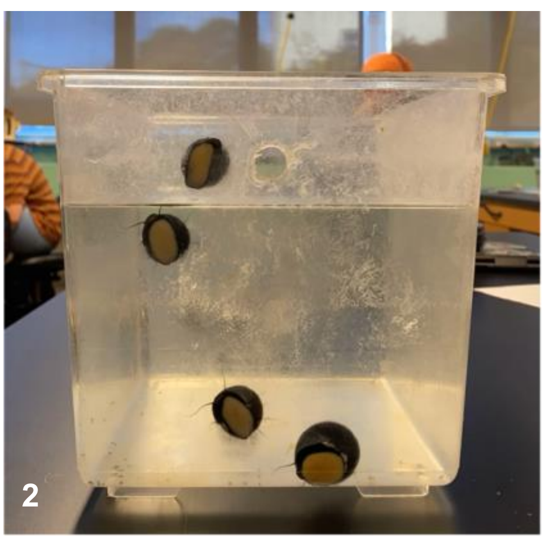
by Jake Vargas

(2022 PNWSC grant recipient)

The chemical scent of predators plays an important role in mediating the interactions between predators and their prey in marine food webs[1,2,3]. This is particularly true in aquatic environments where prey species have been shown to alter their behavior[4] in response to the water-borne chemical scent of predators. Whereas the prey's behavioral changes may increase its chance of survival in the presence of predators, these behavioral alterations may also carry a cost of reduced opportunities for feeding, growth, and reproduction[5]. Because behavioral responses by prey in response to predators often involve a reduction in feeding, they can have cascading effects on other species in marine ecosystems[4]. Therefore, understanding how prey respond to predator chemical 'risk cues' is critical to understanding how predators shape both prey populations, and how they affect communities as a whole.

While it has been postulated that microbes may be critical to breaking down predator risk cues, whether marine microbes influence predator-prey interactions is unknown. So, I set out with two objectives: (1) to investigate how predator risk cues degrade over time; and (2) to investigate whether marine microbes are in fact a key factor in risk cue degradation. To investigate these objectives, I conducted a laboratory experiment that used a well-documented, chemically-mediated interaction between the keystone predatory sea star *Pisaster ochraceous* and its marine gastropod prey, the black turban snail (*Tegula funebralis*; **Fig. 1**). In the chemical presence of *Pisaster* turban snails adaptively crawl out of the water where, once emerged, *Pisaster* no longer pursues them (**Fig. 2**). In the context of my study, I leveraged this crawl-out behavior as a proxy for the chemical presence of *Pisaster*.

I found that turban snails' crawl out response decreased by >50% within 24 hours, suggesting that the chemical risk cue from *Pisaster* degrades quickly. Further, I also found that *Pisaster* risk cue activity was preserved for up to 72 hours by freezing water containing the cue, which presumably slowed microbial degradation of the cue. Surprisingly, my results also suggested that microbes themselves may be a key component of the *Pisaster* risk cue that turban snails respond to, as the snails did not respond to cue water where microbes were removed. To build on these findings, and to study marine microbes' ability to degrade predator chemical cues, while also capturing the ability of microbes to activate these cues, I conducted a final laboratory experiment using a design that crossed microbial presence/absence and predator cue age.



To establish microbe presence/absence treatments, I used the antimicrobial, tetracycline, which has been shown to inhibit the growth of aquatic decomposer bacteria. By applying tetracycline to cue water, and then allowing this mixture of cue water and tetracycline to age over time, I was able to study the degradation of chemical cues through time in the presence and absence of marine microbes (i.e., (+) tetracycline and (-) tetracycline) (**Fig. 3**).

Each trial was conducted within a flow through seawater table at the Telonicher Marine Lab which contained individual (20cm x 20cm x 30cm) aquaria. In these aquaria, four small and four large *Tegula* were added to each aquarium and allowed to acclimate for 15 minutes, after which aquaria were dosed with 150ml of *Pisaster* chemical cue conditioned water, which was slowly poured into the center of the aquarium to ensure that *Tegula* were not disturbed. Previous experiments have shown that once introduced to *Pisaster* cue, *Tegula* crawl out within one hour (Vargas & Bourdeau, unpublished data); therefore, each trial was two hours long. Throughout the experiment, I took note of an individual snail's position in its aquarium every 20 minutes. Individual *Tegula* were considered in 'crawl-out' avoidance if they completely exit the water, or at a minimum; were 25% emerged.

Through this final experiment I found that microbes likely play a key role in the chemically-mediated predator-prey interaction between *Pisaster* and turban snails through the production and/or activation of the predator risk cues.

My research begins to provide evidence that microbes play important roles in limiting how aquatic prey chemically perceive their predators. Further study of the *Pisaster*-*Tegula* system and those like it is critical to addressing knowledge gaps in the total effects of predators on prey populations, and their indirect effects on communities. Finally, when considering the abundance of marine microbes and the importance of chemical signaling across many marine species (echinoderms, crustaceans, mollusks, etc.), it is likely that microbial influence on chemical signaling is conserved in many predator-prey interactions throughout the marine environment.

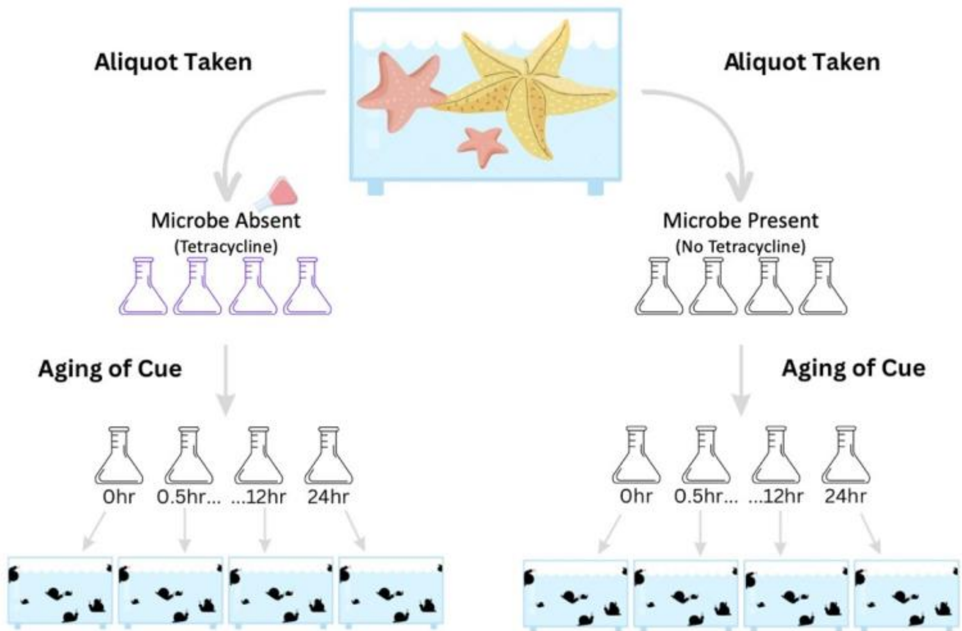


Fig. 3 - A flow chart showing cue water source, followed by the addition of tetracycline in pink, and subsequent aging and addition of cue to treatments.

References:

1. Matassa CM (2010) Purple sea urchins *Strongylocentrotus purpuratus* reduce grazing rates in response to risk cues from the spiny lobster *Panulirus interruptus*. *Mar Ecol Prog Ser* 400:283-288.
2. Mazorra-Alonso M, Tomás G, & Soler JJ (2021) Microbially mediated chemical ecology of animals: A review of its role in conspecific communication, parasitism and predation. *Biology* 10(4): 274.
3. Stabell OB, Ogbebo F, Primicerio R, Inducible Defences in *Daphnia* Depend on Latent Alarm Signals from Conspecific Prey Activated in Predators. *Chemical Senses*. Volume 28 (2): 141–153
4. Estes JA, Terborgh J, Brashares JS, Power ME, Berger J, Bond WJ, ... & Wardle DA. (2011) Trophic downgrading of planet Earth. *Science*. 333(6040): 301-306.
5. Chivers DP, Dixson DL, White JR, McCormick MI, Ferrari MC. Degradation of chemical alarm cues and assessment of risk throughout the day. *Ecol Evol*. 3(11):3925-34. doi: 10.1002/ece3.760. Epub 2013 Sep 17. PMID: 24198950; PMCID: PMC3810885.
6. Bourdeau PE (2010) Cue reliability, risk sensitivity and inducible morphological defense in a marine snail. *Oecologia* 162: 987–994. https://doi.org/10.1007/s00442-009-1488-5