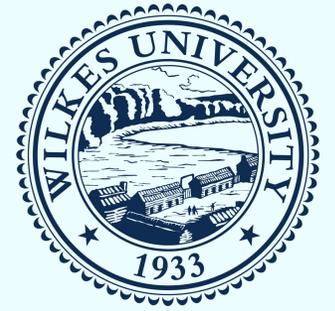


Effects of *Microcystis aeruginosa* on the Metamorphosis and Growth of the Polychaete Annelid *Capitella teleta*

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Introduction

The polychaete *Capitella teleta* is very opportunistic in nature due to their ability to inhabit and flourish in organically enriched marine sediments, which allows them to survive in harsh conditions and polluted environments, such as may be associated with algal blooms (Gamenick *et al.*, 1998). *Capitella teleta* uses chemical cues in order to know when and where to metamorphose. Many of these cues are found in organic materials that algal blooms may possess. (Cohen *et al.*, 1999) "Harmful algal blooms are globally increasing in frequency, magnitude and geographical extent" (De Rijcke *et al.*, 2015). In addition 75% of these blooms contain some form of toxin. Due to this the *Capitella teleta* is being exposed to the effects of algal blooms all around the world.

A prevalent cyanobacteria in many of these blooms is *Microcystis aeruginosa*. *Microcystis* contains the liver toxin microcystin, A cyclic peptide. The toxin is a potent inhibitor of serine & threonine protein phosphatases. When protein phosphatases are inhibited accumulation of phosphorylated proteins in the liver is caused. This can cause many effects to happen to liver cells (Hepatocytes) and to liver function overall. Some of these include cell necrosis, massive hemorrhaging and in severe cases death can occur to the organism due to complete liver failure.

This study aims to look at how the Microcystin liver toxin of *microcystis* effects the opportunistic *Capitella teleta* polychete worm. Due to the *Capitella teleta*'s abilities in other polluted environments it is expected to potentially be able to survive in a heavy *Microcystis* environment. This will be done by testing how *Capitella teleta* reacts to the *Microcystis* environment in all of its life cycle forms. From larvae through adult. As well as if it can reproduce in the toxic environment.

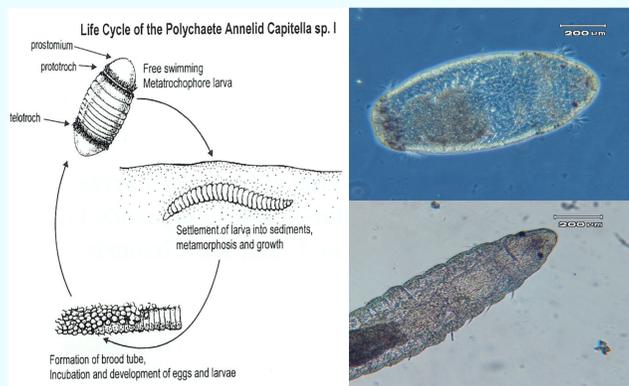


Figure 1. The polychaete annelid *Capitella teleta*. The image to the left shows the biphasic life style of *C. teleta*, showing both the swimming larval stage and the settled juvenile stage. The top right image shows the larval stage and the bottom right is a juvenile worm.

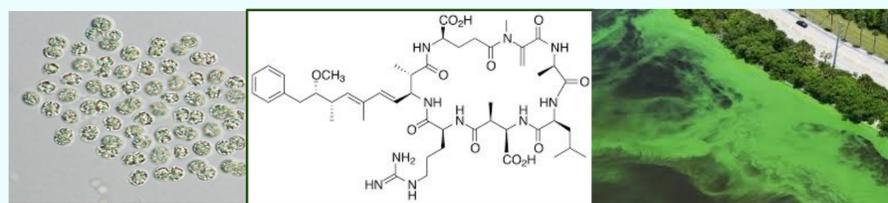


Figure 2. *Microcystis*. The image on the left depicts *microcystis*, under 1000x, which is a cyanobacteria that is known to produce *microcystin*, a toxin. The image in the middle depicts the chemical makeup of microcystin, which is a cyclic peptide. Lastly, the image on the right depicts *microcystis* creating an algal bloom.

Methods

- Cultures of adult *Capitella teleta* were maintained in a temperature and light controlled incubator in one gallon plastic containers containing sea sand and artificial seawater (ASW) at 18°C, with a photoperiod of 12 hrs. light/ 12 hours darkness. The cultures were fed Tetramin fish food flakes provided *ad libitum*.
- Brood tubes of *C. teleta* were isolated from the cultures, placed in glass Petri dishes, and checked daily for released metatrochophore larvae.
- *Microcystis aeruginosa* was obtained from Carolina Biological Supply Co. and cultured in Alga-Gro medium at room temperature (20°C) with ambient lighting.
- Larval settlement : 10 trochophore larvae were added to two plastic petri dishes with 10 mL of artificial seawater, control group contained 1 mL algal medium and experimental group contained 1 mL microcystis LB strain. Experimental group was left in contact with microcystis for 2 hours before adding marine mud.
- Measurements of *C. teleta* : growth of larvae were taken via a ruler every 2 days, as well as percentage of larval settlement and metamorphosis.

Results

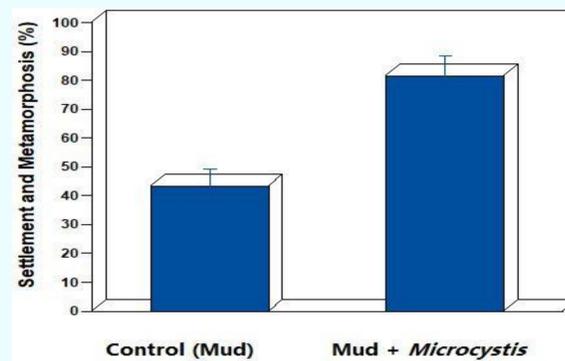


Figure 3. Effects of *Microcystis aeruginosa* on the rate of larval settlement of *Capitella teleta*.

C. teleta larvae were obtained from brood tubes and introduced to finger bowls containing 10 ml of ASW with tetramin or *Microcystis* along with marine mud (a known inducer of settlement), and the average percentage of larvae that were settled and had metamorphosed were recorded every two days. The results show that *Microcystis* caused 70% larval settlement of *Capitella* larvae compared to 30% with control larvae to which just tetramin was added.

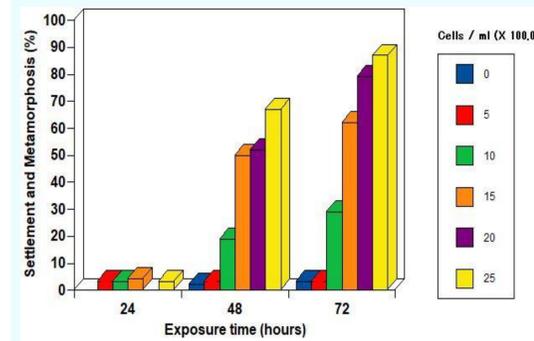


Figure 4. Effect of *Microcystis* on trochophore larval settlement and metamorphosis of *Capitella teleta*. Increased cell concentrations of *Microcystis* and increased time of culture were found to stimulate instead of inhibit larval settlement and metamorphosis. Individual bars represent the mean settlement and metamorphosis for three replicates of thirty larvae.

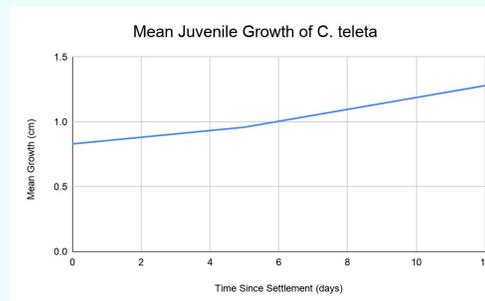


Figure 5. Effect of *Microcystis* Diet on Juvenile growth. 300,000 cells of *microcystis* Texas LB 2385 was added per 100 mg silica. The mean growth per day of juveniles is plotted with an average rate of about 1.0 mm per day being observed.

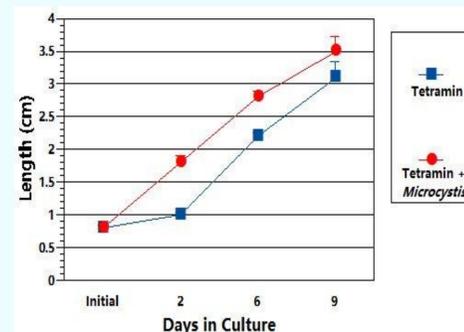


Figure 6. Effect of Organic Content on *C. teleta* Growth. The blue line representing the control group which was only fed tetramin had a growth rate of about 2.4 mm per day. The experimental group which was fed both tetramin and *microcystis* Texas LB 2385 is shown in red and that group had a growth rate of 3.1 mm per day.

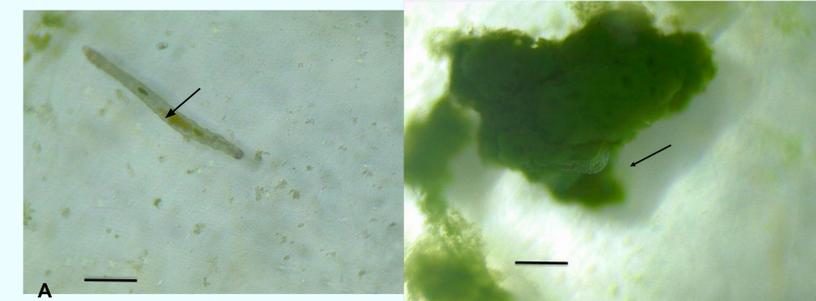


Figure 7. Consumption of *Microcystis* by *C. teleta* juveniles. *Capitella* larvae metamorphosed into juvenile worms upon exposure to *Microcystis* and the metamorphosed juveniles were seen to actively feed on the *Microcystis* and also made cyanobacterial conglomerates from the *Microcystis* in which *Capitella* colonies formed. (A) Juvenile *C. teleta* showing green *Microcystis* in gut and fecal pellet (arrow), scale bar = 1 cm. (B) Juvenile *C. teleta* (arrow) shown emerging from a cyanobacterial conglomerate. Scale bar = 1 cm.

Summary

- The presence of *Microcystis aeruginosa* in the water column appears to stimulate settlement and metamorphosis of *Capitella teleta* instead of inhibiting it.
- The presence of *Microcystis aeruginosa* in the water column stimulates settlement and metamorphosis of *C. teleta* both by itself and also in response to a marsh mud cue.
- Juvenile worms of *C. teleta* appear to grow for a short period of time using *Microcystis aeruginosa* as a food source, but for longer periods of time over 1 week, the diet of just *Microcystis* did not support further growth and mortality resulted.
- When juvenile worms of *C. teleta* were however grown in the presence of both Tetramin fish food flakes and *Microcystis* however, growth was increased compared to worms grown on just Tetramin alone.
- These results indicate that algal blooms of *Microcystis* may be beneficial for the development and growth of *Capitella teleta* in the marine environment, instead of being harmful as it is to many other species.
- We are further carrying out studies to determine if the presence of *Microcystis* inhibits ovarian maturation and reproduction of *C. teleta*.

References

- Cohen, Risa A., *et al.* "Relationship between Sediment Organic Content, Metamorphosis, and Postlarval Performance in the Deposit-Feeding Polychaete *Capitella* Sp. I." *Journal of Experimental Marine Biology and Ecology*, Elsevier, 9 Feb. 2000, www.sciencedirect.com/science/article/abs/pii/S0022098199000477.
- De Rijcke, M., *et al.* "Common European Harmful Algal Blooms Affect the Viability and Innate Immune Responses of *Mytilus Edulis* Larvae." *Fish & Shellfish Immunology*, vol. 47, no. 1, 2015, pp. 175-181.
- Ellen P. Preece, F. Joan Hardy, Barry C. Moore, Michael Bryan, A review of microcystin detections in Estuarine and Marine waters: Environmental implications and human health risk, *Harmful Algae*, Volume 61, 2017, Pages 31-45, ISSN 1568-9883, https://doi.org/10.1016/j.hal.2016.11.006.
- Miller, M.A., *et al.* "Evidence for a Novel Marine Harmful Algal Bloom: Cyanotoxin (*Microcystin*) Transfer from Land to Sea Otters." *PLoS One*, vol. 5, no. 9, 2010, pp. E12576.
- Toonen, Robert., *et al.* "Settlement of the gregarious tube worm *Hydroides dianthus* (Polychaeta: Serpulidae)." *Figure 2f from: Irimia R, Gottschling M. Biodiversity Data Journal 4: e7720. https://doi.org/10.3897/BDJ.4.e7720.*" 2001, doi:10.3897/bdj.4.e7720.figure2f.

Acknowledgement : This research was kindly supported by a Fenner Grant received from the Fenner Endowment to Wilkes University