The vertical migration behavior of *Trichocoleus* sp. in the absence of light

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ABSTRACT

Trichocoleus sp. is a blue-green algal species that forms mats in the sandy sediments of Douglas Lake. The alga forms mats in order to gain protection from UV radiation, and the alga sacrifices productivity in the process. Once removed from light and UV exposure, *Trichocoleus* sp. interestingly did not migrate upward in the sediments to seek higher nutrient concentrations and light levels. A combination of mechanisms such as positive and negative chemotaxis and factors such as shading and anoxic conditions could have influenced the behavior of *Trichocoleus* sp. in such a way that no change in mat depth was evident at the conclusion of the experiment. The thickness of the mat did increase slightly, however. The function of mat thickness is unclear but might represent mat stability.

INTRODUCTION

UV irradiation has many negative effects on photosynthetic organisms. Destruction or conformational changes of molecules used in the photosynthetic pathway, such as proteins, can reduce photosynthetic activity. The nucleic acids that constitute DNA can also suffer from photodamage, preventing proper cell division (Holzinger and Lütz, 2006). Cyanobacteria employ a number of methods to increase survival in environments with high UV exposure, including DNA repair mechanisms (Holzinger and Lütz, 2006) and anti-oxidant and UV-absorbing pigment production (Ehling-Schulz and Scherer, 1999).

Filamentous cyanobacteria capable of gliding movements use an additional tactic to gain protection from UV radiation. The cyanobacteria can move through and under sand grains and other algae to seek shading from harmful UV radiation (Whale and Walsby, 1984). The vertical movements of filamentous cyanobacteria results in the formation of mats in sediments (Bebout and Garcia-Pichel, 1995).

Although the cyanobacteria experience less stress induced by UV radiation while living within an algal mat, the algal community is less productive. The position of the cyanobacteria deep within the mat might decrease exposure to wavelengths of light used in photosynthesis and increase the distance by which nutrients would have to diffuse to reach the algal cells (Bebout and Garcia-Pichel, 1995). For this reason, the position of gliding cyanobacterial filaments within an algal mat most likely represents a tradeoff between gaining protection from damaging UV radiation and experiencing lower productivity levels.

Blue-green algal mats form in Douglas Lake in sandy sediments in the shallow littoral zone. Because of the morphology and size of the trichomes comprising the algal filaments, the alga forming the Douglas Lake mats is most likely a member of the genus *Trichocoleus.* Available resources did not allow the identification of the species of the alga. If the algal mats in Douglas Lake form due to UV stress, the removal of UV radiation from the environment might allow the cyanobacteria to migrate vertically in the sediments where nutrients and photosynthetic wavelengths are more available for primary production.

In this experiment, I tested the hypothesis that *Trichocoleus* sp. forming mats in Douglas Lake near Grapevine Point did so in order to escape intense UV radiation exposure and would move to the top layer of the mat to seek more nutrients and light if stress from UV radiation were removed. I eliminated light exposure, and thereby eliminated UV exposure, to algal mat specimens taken from Douglas Lake and tracked the movement of the *Trichocoleus* sp. after a two week period. At the conclusion of the experiment, I looked for vertical migration of the blue-green algal layer and changes in the thickness of the layer to see if *Trichocoleus* sp. reacted behaviorally to the removal of UV radiation from the environment.

METHODS

I used five treatment aquaria and four control aquaria. Each aquarium had a capacity of 10 gallons. All nine aquaria were filled with a three centimeter layer of sand that was removed from the area surrounding the original location of the algal mats in Douglas Lake. I filled the aquaria with water from Douglas Lake and allowed the sediments to settle for 24 hours. I placed the control aquaria at the end of a pier in order to best simulate natural photoperiod and light levels. The aquaria were covered with transparent material to prevent lake water displacement by rainwater. I placed the treatment aquaria indoors in a dim area and put a blanket over the aquaria to block out any remaining light.

The algal mat was collected from Douglas Lake, on the west side of Grapevine Point. The area was sandy with no visible rocks or macrophytes. At the time of the collection, the skies were overcast and the winds were moderate. The location of my collection was approximately

45.569487, -84.680321. I harvested one large mat at a depth of 0.82 meters. With a razor, I divided the mat into nine different samples of approximately the same size and randomly placed each sample into the top half of a petri dish, each numbered 1-9.

I measured the thickness and depth of the *Trichocoleus* sp. layer in the mat before running the experiment using a ruler. I measured the thickness by qualitatively determining the extent of blue-green color and recording the thickness of the layer. The layer depth in the mat was determined by measuring the distance from the surface of the mat to the upper limit of the layer. I took three depth measurements and three thickness measurements along the cross section of each sample, spaced 1 centimeter apart, and averaged the three numbers in order to generate one average thickness and one average depth value for each sample. All measurements were rounded to the nearest quarter millimeter before the average was taken.

I placed samples 1-4 into the control aquaria and dishes 5-9 into the treatment aquaria and let the experiment run for two weeks. At the end of the two-week period, I removed the petri dishes from the aquaria, took a fresh cross-section of each sample, and repeated the measurement regime outlined in the previous paragraph.

I compared the thicknesses before and after the two-week period, and I compared the depths of the algal mats before and after the two-week period. I used t-tests to determine if any significant differences the measurements existed between the samples before and after the completion of the experiment.

RESULTS

The thickness and depth measurements of each sample taken before I ran the experiment showed some variation (Table 1). The average thickness ranged between 1.17 mm and 3.33 mm. The average depth ranged between 1.08 mm and 1.83 mm. At the completion of the experiment, variation in average mat thickness and depth still existed among the nine samples (Table 2).

I also used t-tests to determine if any change in mat depth or thickness occurred during the experiment. According to the statistical tests, the thicknesses of the control samples did not significantly change over time (Figure 1). The p-value for the control mat thickness before and after the experiment was 0.6939. A p-value of 0.0563 supported that the thicknesses of the treatment samples did show a nearly statistically significant change, increasing slightly over time

(Figure 1). A change in mat thickness might represent a behavioral reaction of the *Trichocoleus* sp. to the conditions in the treatment aquaria. The statistics also showed that the depths of the control and treatment samples did not change over the course of the experiment (Figure 2). The p-value for the control mat depth before and after the experiment was 0.8977. The depths of the treatment samples stayed nearly the same over time, as shown by a p-value of 0.9743.

I made three additional observations about the algal mats. First, at the completion of the experiment, a new, soft algal mat formed in the transplanted sediments of the control aquaria, but no such mat formed in the treatment aquaria. Second, I noticed that at the end of the experiment, a noticeable layer of green algae formed over the surface of the sediments in the treatment aquaria. Finally, the algal mats were denser higher in the sediments, and denser mats were able to withstand wave action in Douglas Lake.

Table 1. Initial average thickness and depth of the algal mat on nine samples.

Mat piece #	Thickness (mm)	Depth (mm)
1	1.92	1.25
2	2.33	1.33
3	1.92	1.08
4	3.33	1.83
5	1.33	1.67
6	2.22	1.25
7	1.17	1.17
8	1.5	1.42
9	2	1.67

Table 2. Final average thickness and depth of the algal mat on nine samples. Grey boxes indicate treatment pieces, and white boxes indicate control pieces.

Mat piece #	Thickness (mm)	Depth (mm)
1	1.5	1.48
2	2.17	1.25
3	2.08	.58
4	3	2
5	2.25	1.33
6	2.75	1.5
7	2.17	1.5
8	1.83	1.25
9	2.25	1.58

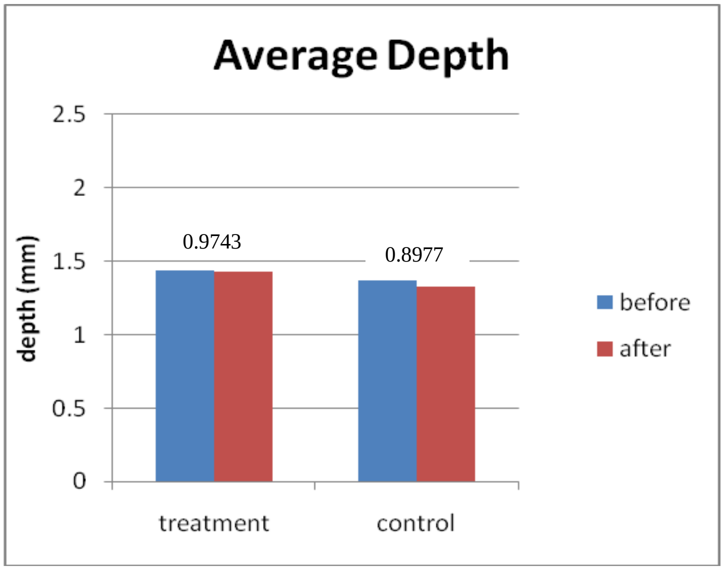

Figure 1 shows the average depths of the control and treatment samples before and after the experiment. There is no significant change in the average depth in either the control or treatment samples. The p-values are shown aove the columns.

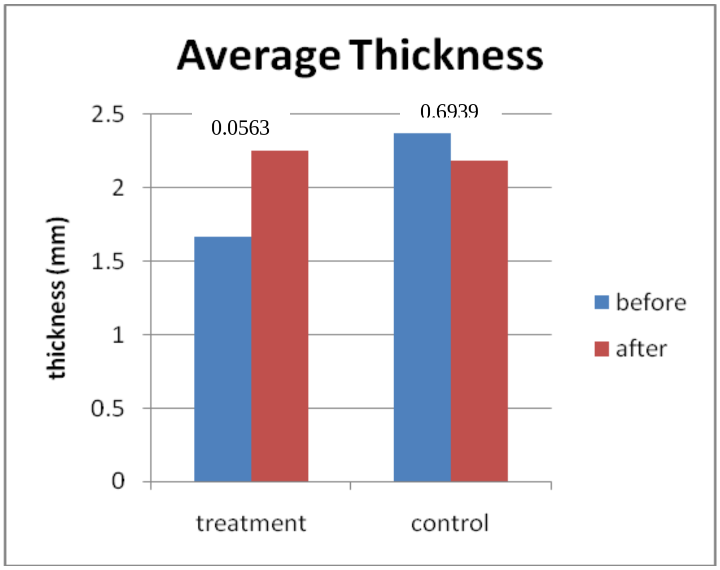

Figure 2 shows the average thicknesses of the control and treatment samples before and after the experiment. There is no significant change in the average thickness of the control samples. There was a weakly significant change in the average thickness of the treatment samples. The p-values are shown above the columns.

DISCUSSION

Many biotic and abiotic factors conflicted over the course of my experiment, influencing the behavior of *Trichocoleus* sp. and preventing clear results. Originally, I thought the complete removal of light exposure from the treatment samples affected the alga's ability to migrate vertically in the mat during my experiment. The fact that new mats formed in the control aquaria but not in the treatment aquaria provided some evidence that light was necessary for mat formation. Evidence from past studies suggested that cyanobacteria use phototaxis to some degree for migration (Whale and Walsby, 1984). If movement was governed exclusively by phototaxis, the absence of a light source could have prevented any vertical migrations in my treatment and explained why there was no significant change in the depth of the mat layer after the experiment. However, a previous study of vertical migration in cyanobacteria determined that the related *Microcoleus chthonoplastes* did not require a light source to move upward in sediment and instead migrated based on chemical influences such as oxygen or acid concentrations (Whale and Walsby, 1984).

The same study suggested that *M. chthonoplastes* used chemotaxis to migrate, either moving toward aerobic conditions near the surface of the sediment or moving away from unfavorable acid concentrations (Whale and Walsby, 1984). Relevant information about *Trichocoleus* sp. migration was lacking, so for the purpose of my study, I assumed that *Trichocoleus* sp. and *M. chthonoplastes* migrated by the same mechanisms. If there was no significant concentration gradient between the sediments and the open water in the aquaria, *Trichocoleus* sp. might not have had a chemical signal toward which to move. The absence of light in the treatment aquaria probably reduced photosynthesis rates dramatically, preventing further production of oxygen, while microbial metabolic activity and algal respiration could have consumed any remaining oxygen in the treatment aquaria (Dodds, 2002). A study of *Oscillatoria agardhii* showed that the cyanobacterium could continue growing in dark conditions by use of aerobic glycogen degradation (Stal, 1995). The use of glycogen by cyanobacteria as an energy source under aerobic conditions endorsed the mechanism of positive oxygen chemotaxis in *Trichocoleus* sp. Alternatively, production of acids by bacteria living above or below the mat in the sediments could have influenced the position of *Trichocoleus* sp. in the sediments. Sulfide is an acid that inhibits motility and interferes with respiratory and photosynthetic electron transport in cyanobacteria (Stal, 1995). High sulfide concentrations in either the control or the treatment aquaria could have arrested the movement of *Trichocoleus*, locking the filaments in the position in which I found the filaments (Stal, 1995). The alga could have also been actively and successfully avoiding high sulfide concentrations in the sediments, settling where the algal cells were most protected from the sulfide. If I were to repeat this study using aquaria, I would measure dissolved oxygen and acid concentrations before and after the duration of the experiment to see if oxygen or acids influenced the behavior of *Trichocoleus*.

The growth of the green algae on the surface sediments in the control aquaria but a lack of movement in response to the presence of the green substance suggested that perhaps another factor that altered the habitat of the blue-green algae was preventing the upward migration of *Trichocoleus* sp. in the mat. The fact that a large quantity of green algae was able to grow on the surface of the sand in the control aquaria represented a substantial shift in the conditions in the aquaria from the conditions in Douglas Lake. In general, green algae prefer high light levels (Wehr and Sheath, 2003), and the green algae growing in the control aquaria might have required slow currents or stagnant waters to proliferate on sand. The same factors that affected the growth of green algae in the control aquaria could have affected the vertical migration of *Trichocoleus* sp. in the control aquaria. The aquaria were much shallower than the areas from which I removed the algal mat. The decreased water depth of the samples could have increased the samples' exposure to UV radiation because the UV radiation did not have to travel through as much dissolved organic carbon before reaching the algal mat (Morris, 1995). The depth of the treatment mats at the end of the experiment could have represented a balance between shading compensation, light level compensation, and UV exposure compensation. The presence of the green algae might have directly affected the vertical migration of *Trichocoleus* sp.

One might predict that increased shading by a green algae layer at the surface of the sand would stimulate *Trichocoleus* sp. to migrate upward to regain normal light exposure (Dodds, 2002). However, the t-test comparing the depths of the control samples before and after the experiment show that depth did not change in the control aquaria despite increased shading. Oxygen production by green algae at the surface might have also influenced positive chemotaxis in *Trichocoleus* sp. Mat density is also an important characteristic of mat structure.

Higher filament density might greatly reduce the productivity of filaments lower in the mat by creating a physical barrier through which nutrients and light could not easily pass.

Although high filament density at the top of the mat might have decreased the productivity of filaments lower in the mat, higher density might be important to mat survival. At Douglas Lake I observed that the densest mats withstood wave action. Perhaps this observation was evidence that high filament density was important to protection from disruption by currents. I am unsure why mat structure did not change in the current-free conditions within the aquaria.

Mat thickness most likely had implications for the productivity of *Trichocoleus* sp. filaments in the mat. The uppermost individuals in the mat probably experienced the most favorable light and nutrient conditions at the expense of the productivity of the filaments deeper in the mat. I believe the change in mat piece thickness in the treatment aquaria was simply a result of the death and sinking of some filaments in the sediments after the two weeks. Because photosynthesis continued in the control aquaria, I assume that relatively few filament deaths occurred in the control aquaria, preventing a noticeable increase in mat thickness. Further studies could determine which factors, other than filament death, affect mat thickness, but my experiment did not appear to test the behavior associated with mat thickness.

Ultimately, my experiment did not directly test the affect of UV radiation removal on motile cyanobacteria. The absence of light exposure or the removal from the natural habitat in Douglas Lake most likely altered too many factors important to the vertical migration behavior of *Trichocoleus* sp. The utilization of a more quantitative than qualitative method for determining the location of *Trichocoleus* sp. in the sediments would also be beneficial to the study. Further experiments would reveal which specific factors affect the migration of the blue-green algae in Douglas Lake.

LITERATURE

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