Evaluating the effects of differing levels of UV radiation on periphytic diatom communities in a river

Crystal Cole

University of Michigan Biological Station
Limnology
16 August 2014
Dr. Paul Moore

Abstract

Release of ozone-depleting emissions into the atmosphere has resulted in the thinning of the stratospheric ozone layer. Higher levels of UV radiation are able to impact the biosphere, and the impacts on ecosystems, specifically aquatic ecosystems, could be significant. An experimental stream set up was designed to test the effects of different levels of UV radiation on periphytic diatom communities. Species diversity, differences in community composition, and changes in relative abundances of genera compared before and after manipulation of UV levels showed that UV radiation has a strong impact on diatom communities. Diversity is limited more by limited UV radiation than by limited photosynthetically active radiation. Populations are more significantly different from control populations when UV radiation is limited, compared to when PAR is limited. Changes in community composition as a result of different UV levels can have impacts throughout the food web as diatom predators selectively feed on certain diatom genera, further altering the diatoms' relative abundances, and as higher trophic levels are impacted by the changes seen in predator community composition. These results should not be extrapolated to indicate long term impacts of increased UV radiation, as other studies have shown that initial UV inhibition in the short term reverses and results in an increase in algal biomass long term (Bothwell et al. 1993). Additional study should be conducted to assess the long term impacts of increased UV radiation on freshwater lotic periphytic diatom communities.

Introduction

Depletion of the stratospheric ozone layer allows for the transmittance of higher levels of solar radiation than is currently found on Earth. Anthropogenic emissions that include elements like bromine (Yung et al. 1980), chlorine (Farman, Gardiner, and Shanklin 1985; McElroy et al. 1986; Molina and Rowland 1974), nitrogen and hydrogen oxides (Bates and Nicolet 1950; Crutzen 1970, 1971; Farman et al. 1985; Johnston 1971), and carbon (McElroy et al. 1986; Tung et al. 1986; Wofsy, McElroy, and Yung 1975; Yung et al. 1980; Zepp et al. 2007), increase natural levels of these gases in the atmosphere and lead to thinning of the ozone layer (Solomon 1999; Yung et al. 1980). The ozone "hole" over Antarctica is the most widely known example of the impacts of human emissions on the ozone layer. However, the ozone layer is thinning worldwide (Madronich et al. 1998).

The thinning of the ozone layer causes an increase in transmittance of harmful solar radiation, notably UV-A and UV-B radiation (Madronich et al. 1998; Gies et al. 2004). UV rays damage DNA in organisms from algae to humans (Rajeshwar and Hader 2002). UV radiation, because of its ability to damage cells, would likely have a larger effect on single cell organisms than larger multicellular organisms; damage to one cell of a single celled organism is much more significant than damage to one cell of a multicellular organism. Buma and colleagues (2001) discovered that DNA damage was more pronounced in smaller organisms than in larger. In addition, damage to communities of single celled autotrophs compared to single celled heterotrophs would have a greater impact on food webs and ecosystems, because the autotrophs form the base of the food web, especially in aquatic ecosystems. Aquatic ecosystems are often based on populations of algae as the main autotrophs. Therefore, if the populations of algae are greatly impacted by environmental changes, that impact will echo throughout the food web.

Diatoms are algae highly favored by heterotrophs because of their high nutrient and fat content, and are vital in aquatic systems for heterotrophs like fish. Many studies have been done examining the impacts of increased UV radiation on diatom communities in marine environments (Buma, De Boer, and Boelen 2001; Häder et al. 2007; Litchman and Neale 2005; Sundbäck et al. 1996; Zudaire and Roy 2001) and freshwater lentic environments (Pienitz and Vincent 2000; Vinebrooke and Leavitt 1996, 1999). Few studies have been done on diatom communities in freshwater lotic systems (Bothwell et al. 1993), however. Many fish species, like salmon and trout, rely on rivers and streams for stages of their life cycle, and diatoms are rich sources of nutrients, oils, and calories. A significant impact on diatom communities from increased UV radiation could have broad consequences as a bottom up cascade throughout the food web (Häder et al. 2007). More investigations should be conducted into possible consequences of increased UV radiation on algal populations in freshwater lotic systems.

This study aims to fill this need for more information regarding the impact different levels of UV radiation has on diatom communities in streams and rivers by exploring the impacts of different levels of UV radiation on diatom communities in a controlled stream setting. It is hypothesized that there will be a difference in diatom communities as a result of differing exposure to UV radiation. This hypothesis will be explored by way of examining the relative abundance of different diatom genera in a controlled experimental stream that retains as many characteristics of a natural river as is practical.

Methods

The experiment was conducted at the Stream Research Facility (SRF) at the University of Michigan Biological Station (UMBS), Douglas Lake, Michigan. Nine flow through vinyl flumes (8 cm width x 6 cm height) were fed by water pumped from the East Branch of the Maple River (EBMR) using a Monarch pump with 2.54-cm holes in the impeller. The water passes through headwater tanks and is distributed

by garden hose (19 mm diameter) to the flumes, and afterwards the water is collected and returned to the EBMR. EBMR drains from Douglas Lake, an alkaline glacial lake, and flows through second-growth forest before emptying into Burt Lake, another glacial lake. EBMR also receives groundwater input from the surrounding forest. The surrounding forest is owned by UMBS and experiences minimal human impact, and EBMR receives substantial groundwater input from that area. The combination of ground water and surface water results in a mixture of nutrients and established flora and fauna in the river, creating an optimum habitat to test algal communities in a controlled setting while still utilizing available nutrients and organisms in the EBMR ecosystem.

Algal communities were colonized on ceramic tiles (4.7 cm x 4.7 cm x 0.5 cm) for 13 days in a pool created by cinderblocks and plastic and fed by EBMR at 8 cm depth. The colonized tiles were then moved to the flumes for an additional 3 days of colonization before treatments were applied. Two tiles were placed in each stream, one at 1 m downstream of the flow source, and an additional meter downstream from the first tile. Two uncolonized tiles were placed directly upstream of each colonized tile in order to mimic turbulent flow present in the natural river system and to eliminate any stress the algal communities may have experienced from direct flow. An endcap was placed upstream of the flow source in each stream to stop backflow of water. A screened endcap (height of dam = 4.5 cm) was placed at the end of the flumes in order to increase water depth farther upstream. Nylon stockings were placed on the main hose at the headwater tanks in order to limit the amount of detritus flowing through and settling within the flumes.

Treatments were applied to the flumes following the total 16 days of colonization of tiles. The nine flumes were divided into sets of three, one set having one treatment. SOLACRYL® SUVT acrylic sheeting, which transmits 98% of all UV rays and 90% of photosynthetically active radiation (PAR), was used on

three streams. Plexiglass MC UF-5, which transmits 2% of all UV rays and 90% of PAR, was used on three streams. Three control streams had no acrylic sheeting, thus experiencing 100% of all UV rays and 100% of PAR.

One colonized tile was sampled randomly from each stream at 0 days by assigning the upstream and downstream tiles numbers and using a random number generator to determine which tile would be sampled. At least one tile was sampled from upstream and one from downstream. The tiles were cleaned and scraped into a glass high rim bowl using a toothbrush in order to gather the entire diatom population. Water from the Maple River was used to rinse the tile, toothbrush, and bowl into a Nasco Whirl-Pak®. Tiles were returned to the streams to maintain equal turbulence flow between streams. Samples were returned to the lab and refrigerated until processing. Tiles at 11 days were processed identically to tiles sampled at 0 days.

Samples were analyzed in the lab by oxidization to isolate diatom frustules, following the methods of APHA in *Standard Methods for the Examination of Water and Wastewater* (APHA, AWWA, and WEF 1999), replacing nitric acid with hydrogen peroxide in the cleaning process and restricting the rinsing process to only two times, due to limitations on time. Cleaned samples were mounted using Naphrax mounting medium. Diatom samples were examined under oil immersion at a magnification of 100x and diatoms were identified to genera, with a minimum of 250 valves counted per sample.

A Tukey multiple proportions test was used with calculated relative abundances to compare diatom genera between treatments in order to test for significant differences between treatments and over time. A Shannon-Weiner diversity index was calculated for each treatment and time to allow comparisons between treatments and over time. A chi square analysis was completed to compare

community composition differences between treatments and over time. Relative abundances of each genera in each sample were calculated.

Results

Relative abundance of each diatom genera in each sample was calculated and is displayed in Figure 1. Several genera increased over time, for example, *Synedra* increased from day 0 to day 11 in all three treatments (significant, $P < 0.05$, d.f. 6), but the increase was most pronounced in control streams. At day 11, UV streams and control streams had approximately equal relative abundances of *Synedra* but the UV-shielded stream had a lower relative abundance. *Achnanthidium* showed the same trend but the increase was only significant ($P < 0.05$, d.f. 6) in control streams. At day 11 control streams had the highest relative abundance of *Achnanthidium,* followed by UV and then UV-shielded streams. In the case of *Melosira*, there was a significant increase in relative abundance in UV streams ($P < 0.05$, d.f. 6).

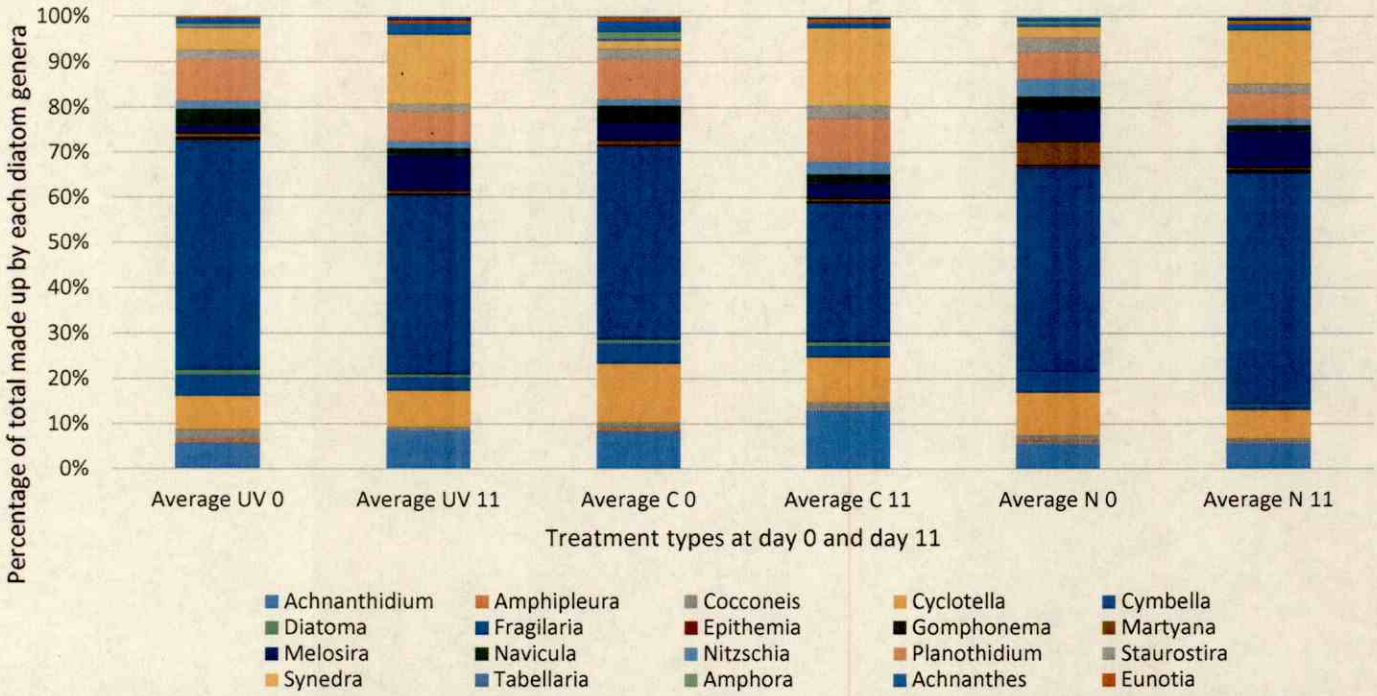

Figure 1. Each diatom genera found is depicted as the percentage of the total population sampled, otherwise called relative abundances. UV samples experienced 90% PAR and 85-90% UV radiation, Control samples were controls and experienced 100% PAR and UV radiation, and UV-shielded samples were shielded from UV radiation and experienced 90% PAR and 2% UV radiation. Treatments are compared between samples at 0 days and samples at 11 days.

Some genera had significant decreases over time. *Fragilaria* decreased in control and UV streams (sig. $p<0.05$, d.f. 6) but had an insignificant increase in UV-shielded streams ($P > 0.05$, d.f. 6). *Martyana* and *Nitzschia* had a significant decrease in UV-shielded streams ($P < 0.05$, d.f. 6; $P < 0.05$, d.f. 6). *Planothidium* had an insignificant decrease in UV streams ($P > 0.05$, d.f. 6), and *Cyclotella* exhibited insignificant decreases in UV-shielded and control streams ($P > 0.05$, d.f. 6; $P > 0.05$, d.f. 6).

UV 0 and UV 11	UV-shielded 0 and UV-shielded 11	Control 0 and Control 11	UV-shielded 11 and Control 11	Control 11 and UV 11	UV 11 and UV-shielded 11	Control 0 and UV-shielded 0	UV 0 and UV-shielded 0	UV 0 and Control 0
210	190	197	136	72	54	108	152	77

Table 1. Chi square values comparing different treatments and different times. The critical chi square value with degrees of freedom 23 is 35.17 ($p<0.05$). All calculations return significant results. UV samples experienced 90% PAR and 85-90% UV radiation, Control samples were controls and experienced 100% PAR and UV radiation, and

Chi square analysis between treatments at day 0, between treatments at day 11, and within treatments at days 0 and 11 returned significant differences for all comparisons (all $P < 0.05$; Table 1). Some comparisons are more similar or more different than others, and this is the best way to compare community composition. UV streams had the largest difference between day 0 and day 11, while UV-shielded streams had the smallest difference.

Chi square values at day 0 between different streams were very high. UV and UV-shielded had the highest difference, followed by control and UV-shielded, and the smallest difference belonged to the comparison between control streams and UV streams.

After treatments the comparisons changed dramatically. The UV-shielded streams compared to the control streams had the largest difference at day 11, followed by the control streams compared to UV streams, and the smallest difference was between UV and UV-shielded streams at day 11. The chi square value comparing UV and UV-shielded streams on the same day dropped from 152 at day 0 to 54 at day 11. Other comparisons remained relatively similar: control and UV-shielded stream comparison rose from 108 to 136, and control and UV stream comparison dropped from 77 to 72.

UV-shielded 0	UV-shielded 11	Control 0	Control 11	UV 0	UV 11
2.35	1.79	2.31	2.16	2.23	2.03

Table 2. Shannon diversity indices were calculated for each sample at the beginning and end of the trial. UV samples experienced 90% PAR and 85-90% UV radiation, Control samples were controls and experienced 100% PAR and UV radiation, and UV-shielded samples were shielded from UV radiation and experienced 90% PAR and 2% UV radiation.The control sample exhibited the least reduction in diversity from beginning to end, followed by the UV sample, and the greatest reduction in diversity was found in the UV-shielded treatment.

The Shannon-Weiner diversity index was used to calculate the diversity of each set of samples before and after treatment, and showed decreases over time in all streams (Table 2). The UV-shielded stream experienced the greatest decrease, from 2.35 to 1.79. UV streams and control streams decreased as well, but not as dramatically.

Discussion

The differences in relative abundance between day 0 and day 11 between treatments are intriguing and show how different genera respond to differing levels of PAR and UV radiation. *Synedra* and *Achnanthidium* increased in all streams but most predominantly or most significantly in control streams.

Other genera exhibited decreases, such as the significant decreases of *Martyana* and *Nitzschia* and the insignificant decreases of *Cyclotella* in UV-shielded streams. These genera did not have significant

increases in streams with higher levels of UV radiation, the control and UV streams, but their relative abundances decreased when UV radiation was eliminated.

Fragilaria decreased in UV and control streams, *Planothidium* decreased in UV streams, and *Cyclotella* decreased in control streams. These streams are subject to similar levels of UV radiation, and the PAR availability decreases by only 10% between the control and the UV streams. The decreases in *Planothidium* and *Cyclotella* were insignificant, which means the only significant changes in UV and control streams were the decreases in *Fragilaria* and the increases in *Synedra, Achnanthidium*, and Melosira (in UV streams only).

The many changes in relative abundance of genera from day 0 to day 11 resulted in UV streams having the largest differences in the chi square analysis. UV-shielded streams had the smallest difference between day 0 and day 11, which indicates that UV streams had a larger change in community composition than UV-shielded streams. UV-shielded streams experienced the greatest decrease in diversity, however. The diversity index for UV-shielded streams dropped by 0.56, while the indices for control and UV streams dropped by 0.15 and 0.2, respectively. The difference between the index decreases for control and UV streams is explained by the reduction in PAR. The difference between the index decreases for UV and UV-shielding streams, therefore, is explained by the near total elimination of UV radiation.

Another interesting finding of the chi square analysis is that the difference between UV and UV-shielding streams was much higher at day 0 than at day 11. The populations became more similar with time. This result is likely due to the significantly high increases in relative abundance of *Synedra* and *Achnanthidium* in both streams, resulting in a reduction of relative abundance of other genera, like

Fragilaria in UV streams and *Martyana, Nitzschia,* and *Cyclotella* in UV-shielded streams. The sharp decrease in diversity in UV-shielded streams likely resulted in the UV and UV-shielded streams becoming more similar with time.

Comparing UV to control streams, and UV to UV-shielding streams shows the relative importance of UV radiation compared to PAR radiation. Comparing the difference between UV stream communities and control stream communities at day 0 to the same difference at day 11 shows a drop of 5 point in the chi square analysis. When comparing the differences between UV stream communities and UV-shielding stream communities, there is an decrease of 98 points. Comparing control stream communities and UV-shielding stream communities at both day 0 and day 11 shows an increase of 24 points. Clearly the largest difference occurs when UV limitation is introduced. When compared to the control stream, differences in community composition becomes more significant.

The changes in relative abundance in some of the individual genera shift the community composition and could cause repercussions throughout the food web. Protozoans tend to select prey diatoms of a given size or shape, grazing on only those genera and reducing the population of those specific genera in relation to the genera which are not grazed upon (Patrick, unpublished data, as seen in Werner 1977). Zooplankton of certain sizes graze on phytoplankton of certain sizes, and some zooplankton graze selectively on diatoms that contain certain organic compounds (Wetzel 2001). Therefore, changes in community composition of diatoms can affect food availability for zooplankton and possibly cause a change in community composition of zooplankton as well. This trend once established would likely echo throughout the food web as certain organisms only consume specific groups of lower trophic level organisms, based on availability, ease of access, and size. So the change in community composition

documented in this study could be precursors of possible changes in the food web if levels of UV radiation change.

Most of the differences of changes in relative abundance are likely due to the different levels of UV radiation and PAR available to the diatom community. The increase or decrease of UV radiation has clear impacts on community composition and biodiversity in short-term studies. Our study shows that diversity is limited more by limited UV radiation than by limited PAR. The data also shows that test populations are more significantly different from control populations when UV radiation is limited, compared to when PAR is limited. However, it should be cautioned that extrapolation of this data over long-term UV exposure should be avoided. Bothwell (1980) and Smith and Baker (1989) also cautioned against this, citing their evidence that impacts of UV radiation are evident in different ways at different times in the succession of diatom communities (Bothwell et al. 1993; Smith, Baker, and Smith 1989). While the short term impacts are clear, additional study is needed into the long term impacts of UV radiation on diatom communities in lotic systems.

References

APHA, AWWA, and WEF. 1999. *Standard Methods for the Examination of Water and Wastewater.* 20th ed. edited by L. S. Clesceri, A. E. Greenberg, and A. D. Eaton. Washington, D.C.: American Public Health Association.

Bates, D. R., and M. Nicolet. 1950. "Atmospheric Hydrogen." *Publications of the Astronomical Society of the Pacific* 62(365):106–10.

Bothwell, M. L., D. Sherbot, A. C. Roberge, and R. J. Daley. 1993. "Influence of Natural Ultraviolet Radiation on Lotic Periphytic Diatom Community Growth, Biomass Accrual, and Species Composition: Short-Term versus Long-Term Effects." *J Phycol* 29:24–35.

Buma, A. G. J., M. K. De Boer, and P. Boelen. 2001. "Depth Distributions of DNA Damage in Antarctic Marine Phyto and Bacterioplankton Exposed to Summertime UV Radiation." *Journal of Phycology* 37:200–208.

Crutzen, P. J. 1970. "The Influence of Nitrogen Oxides on the Atmospheric Ozone Content." *Quarterly Journal of the Royal Meteorological Society* 96(408):320–25.

Crutzen, P. J. 1971. "Ozone Production Rates in an Oxygen-Hydrogen-Nitrogen Oxide Atmosphere." *Journal of Geophysical Research* 76(30):7311–27.

Farman, J. C., B. G. Gardiner, and J. D. Shanklin. 1985. "Large Losses of Total Ozone in Antarctica Reveal Seasonal ClOx/NOx Interaction." *Nature* 315:207–10.

Häder, D. P., H. D. Kumar, R. C. Smith, and R. C. Worrest. 2007. "Effects of Solar UV Radiation on Aquatic Ecosystems and Interactions with Climate Change." *Photochemical & photobiological sciences : Official journal of the European Photochemistry Association and the European Society for Photobiology* 6(3):267–85.

Johnston, H. 1971. "Reduction of Stratospheric Ozone by Nitrogen Oxide Catalysts from Supersonic Transport Exhaust." *Science* 173(3996):517–22.

Litchman, E., and PJ Neale. 2005. "UV Effects on Photosynthesis, Growth and Acclimation of an Estuarine Diatom and Cryptomonad." *Marine Ecology Progress Series* 300:53–62.

McElroy, M. B., R. J. Salawitch, S. C. Wofsy, and J. A. Logan. 1986. "Reductions of Antarctic Ozone due to Synergistic Interactions of Chlorine and Bromine." *Nature* 321:759–62.

Molina, M. J., and F. S. Rowland. 1974. "Stratospheric Sink for Chlorofluoromethanes: Chlorine Atom Catalyzed Destruction of Ozone." *Nature* 249:810–12.

Pienitz, R., and W. F. Vincent. 2000. "Effect of Climate Change Relative to Ozone Depletion on UV Exposure in Subarctic Lakes." *Nature* 404:484–87.

Smith, Raymond C., Karen S. Baker, and By Raymond C. Smith. 1989. "STRATOSPHERIC OZONE , MIDDLE ULTRAVIOLET RADIATION AND PHYTOPLANKTON PRODUCTIVITY." *Oceanography.*

Sundbäck, K., C. Nilsson, S. Odmark, and A. Wulff. 1996. "Does Ambient UV-B Radiation Influence Marine Diatom-Dominated Microbial Mats? A Case Study." *Aquatic Microbial Ecology* 11:151–59.

Tung, K., M. K. W. Ko, J. M. Rodriguez, and N. D. Sze. 1986. "Are Antarctic Ozone Variations a Manifestation of Dynamics or Chemistry?" *Nature* 322(6082):811–14.

Vinebrooke, R. D., and P. R. Leavitt. 1996. "Effects of Ultraviolet Radiation on Periphyton in an Alpine Lake." *Limnology and Oceanography* 41:1035–40.

Vinebrooke, Rolf D., and Peter R. Leavitt. 1999. "DIFFERENTIAL RESPONSES OF LITTORAL COMMUNITIES TO ULTRAVIOLET RADIATION IN AN ALPINE LAKE." *Ecology* 80:223–37.

Werner, Dietrich, ed. 1977. *THE BIOLOGY OF DIATOMS*. 1st ed. University of California Press.

Wetzel, R. G. 2001. *Limnology: Lake and River Ecosystems*.

Wofsy, Steven C., Michael B. McElroy, and Yuk Ling Yung. 1975. "The Chemistry of Atmospheric Bromine." *Geophysical Research Letters* 2(6):215–18.

Yung, Y. L., J. P. Pinto, R. T. Watson, and S. P. Sander. 1980. "Atmospheric Bromine and Ozone Perturbations in the Lower Stratosphere." *Journal of the Atmospheric Sciences* (2):339–53.

Zepp, R. G., D. J. Erickson, N. D. Paul, and B. Sulzberger. 2007. "Interactive Effects of Solar UV Radiation and Climate Change on Biogeochemical Cycling." *Photochemical & photobiological sciences : Official journal of the European Photochemistry Association and the European Society for Photobiology* 6(3):286–300.

Zudaire, Laurent, and Suzanne Roy. 2001. "Photoprotection and Long-Term Acclimation to UV Radiation in the Marine Diatom Thalassiosira Weissflogii." *Journal of Photochemistry and Photobiology B: Biology* 62:26–34.