GROWING KNOWLEDGE

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Nostoc: A nursery nemesis

Exploring novel approaches to long-term, sustainable management, and even use, of cyanobacteria

BY LUISA SANTAMARIA, MARIA MARLIN AND KATIE GREGOR

Figure 1: A plastic rake was used to remove the nostoc mats from half of each plot. PHOTOS COURTESY OF OREGON STATE UNIVERSITY

ONE MENTION OF NOSTOC is likely enough to fill any nursery manager with intense terror and agony. Nostoc, a green jelly-like film growing over gravel or on ground surfaces that are constantly wet, is a combination of different cyanobacteria species living together, according to initial observations at the Oregon State University North Willamette Research and Extension Center (NWREC) Pathology Lab. These are bacteria that are capable of photosynthesis.

While seemingly innocent enough, the cyanobacteria aggregate to form vast mats, composed of a gooey, gelatinous texture when wet. Nurseries in particular struggle with the omnipresent nostoc since consistent moisture and light conditions are often optimal at these establishments.

Over the years, the cyanobacterial mats accumulate and continue to grow, creating a larger and larger issue. Not only are the cyanobacteria unsightly, but they pose a major slip hazard for nursery workers.

Recommendations for management are varied, with no clear consensus for the most sustainable way to manage these microorganisms. Suggested components of nostoc management include improving soil drainage, solarization, algaecides, and herbicides. Chemical treatments are costly and ineffective against bacteria, and they are often composed of harmful chemicals that are dangerous to both humans and the environment.

We wanted to research a more environmentally friendly and sustainable method of controlling nostoc. Thus, the objectives of this study were to assess the efficacy of manual nostoc removal (cleaning) on overall control of the microorganism and test different sanitiz-

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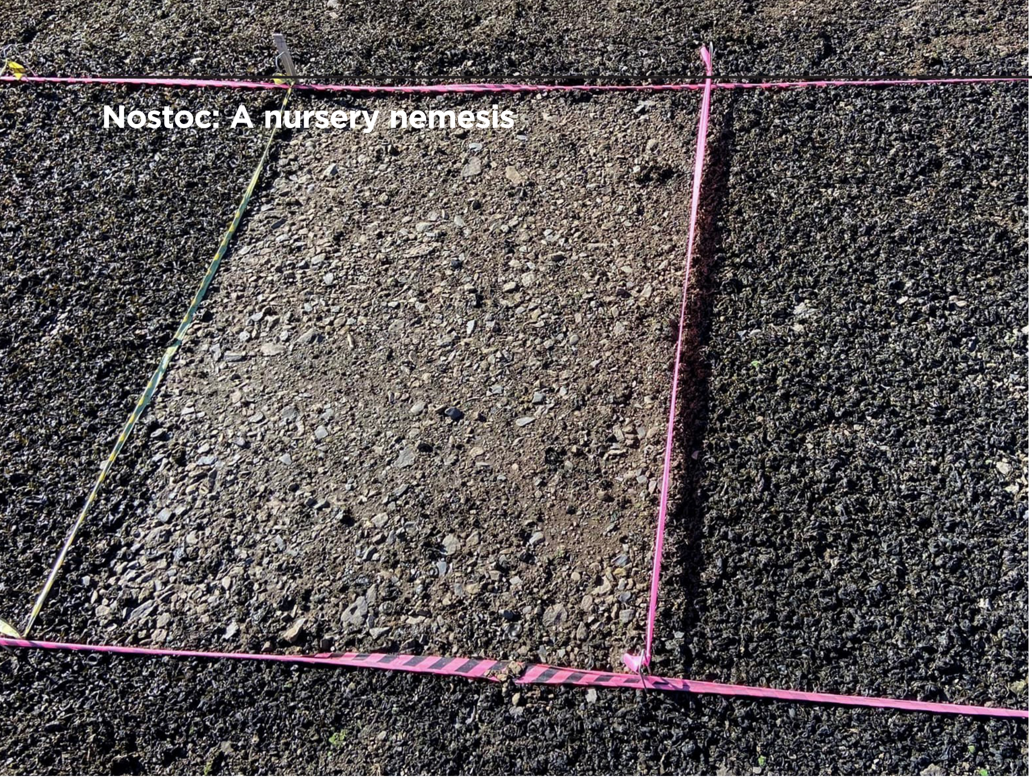

Figure 2. At the final rating (Day 42), the stark difference in nostoc coverage between the cleaned and uncleaned subplots was still very apparent.

PHOTO BY M. MARLIN

ers to determine if two maintenance applications after cleaning could further contribute to nostoc control.

In short, our question was "Does cleaning and sanitizing control nostoc presence and growth in a nursery setting?"

Methodology

We worked in collaboration with a local nursery on this project. The site selected was on level ground and had nearly 100% nostoc coverage. In addition, the irrigation was set to run continuously for 6 hours per day. On July 21, 2021 (Day 0), plots measuring 4-feet by 3-feet were established by using flagging tape and stakes.

We constructed the experiment as a randomized block design, with three blocks of five plots each. This allowed for three replicates of the five sanitizer treatments. Each plot was then divided down the middle, and half of each plot received a complete manual removal of nostoc. This removal was simple; the cyanobacterial mats were raked using a plastic rake and then removed from the area. The other half of each plot was left as is, with complete nostoc coverage.

After the respective half of each plot was cleaned, the treatments were applied to both the clean and the uncleaned sections. Treatments were applied with iPOWER™ handheld pressure sprayer units (which provided uniform droplet sizes), maintaining complete coverage over the entire plot.

Two weeks later, on August 4, we conducted a second and final cleaning and subsequent sanitizer application. First, we collected data (Day 14) on all the subplots, using a predetermined scale. A light, second cleaning was conducted in the subplots that had been cleaned previously on Day 0. We then applied the sanitizer treatments again to all plots. The final evaluation of nostoc coverage took place on September 1 (Day 42).

Results

At the final evaluation, the subplots that had been cleaned prior to sanitation had only between 0-5% nostoc coverage. In contrast, the subplots that had sanitizers applied without cleaning consistently had between 75%-100% nostoc coverage. We noticed this difference consistently at all data collection points throughout the study.

While bleach was the most effective sanitizer, a multiple linear regression analysis did not suggest a significant difference between any of the treatments. However, this model did provide a significant result

Figure 3. Average plot rating for all plots at the final evaluation. Plots, both the cleaned and uncleaned halves, were rated according to the following scale. 0* = 0% coverage, 1 = 1–5% coverage, 2 = 6–25% coverage, 3 = 26–50% coverage, 4 = 51–75% coverage, 5= 76–100% coverage. Error bars represent standard error of the mean; if there is no error bar, then the standard error was 0. *In order to better visualize the rating of 0, all zeros were converted to 0.1 for the analysis and this graphic.

Treatment	Rate
KleenGrow	0.5 fl oz/gal
Bleach	As per label: 50/50 dilution
ZeroTol®	1.5 fl oz/gal
Physan20™	0.5 fl oz/gal
Control (water)	N/A

Scale to evalueate nostoc coverage

0 = 0% coverage
1 = 1–5% coverage
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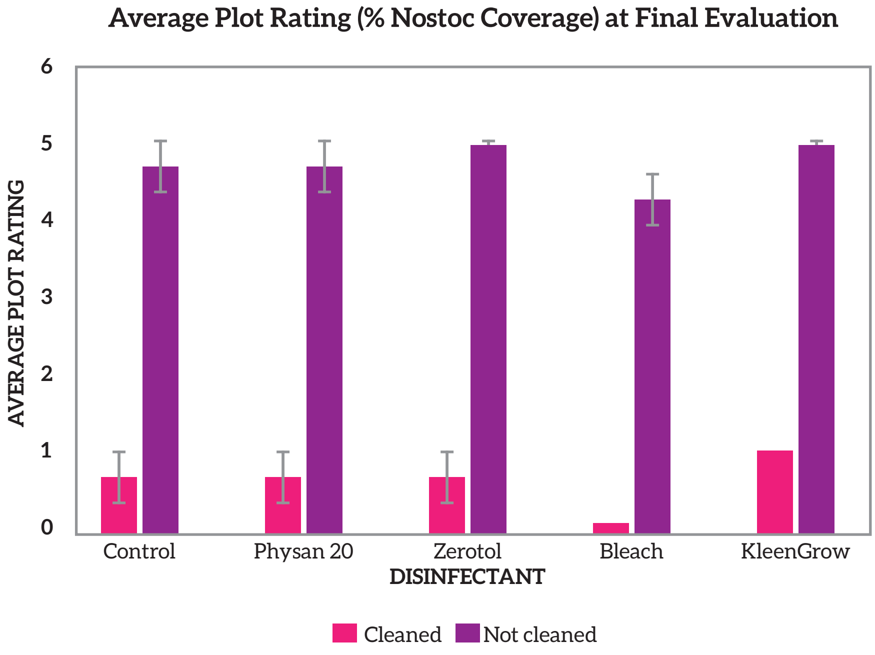
3 = 26–50% coverage
4 = 51–75% coverage
5 = 76–100% coverage



for using "cleaned vs. not cleaned" as a very effective predictor variable for nostoc coverage ($p = 2 \times 10^{-16}$).

Interestingly, the control subplots that had been cleaned and then sprayed with only water remained free of nostoc coverage; this suggests that at least in the short term, sanitizer applications might not even be needed. There was no significant difference in nostoc coverage between the clean subplots that received only water and the clean subplots that received the various sanitizers.

It's also important to emphasize the longevity of cleaning's beneficial effects. Six weeks after the first cleaning, the cleaned subplots still remained mostly free of nostoc, with very low ratings (Figure 3). Conversely, the subplots that did not receive a manual cleaning maintained nearly complete coverage, despite sanitizer application. This circles back to the very core of cleaning and sanitizing. No one can effectively sanitize a dirty surface.We conducted this project to demonstrate that investing in a robust nostoc removal program will actually save nurseries money over time. With the support of the industry, we hope to repeat this experiment and subject the removed nostoc to a compost procedure.

A further step will be to calculate the specific cost of manual labor and the corresponding equipment required vs. repeated chemical applications, but our results support the idea of simple removal for long-term control of nostoc. In addition to economic benefits, nurseries can also contribute to environmental sustainability.

Nurseries can reduce scheduled applications of herbicides or organic compounds that seem effective for temporary nostoc control. More importantly, worker safety will be enhanced as these mats won't pose a risk anymore.

Overall, a significant issue within Oregon nurseries will be addressed, and in turn, the industry can continue to produce healthy plants in an efficient manner, maintaining our nationwide reputation of producing only top-quality ornamentals.

While nostoc might pose a struggle for nurseries, cyanobacteria should not solely be considered a nuisance. There are several »

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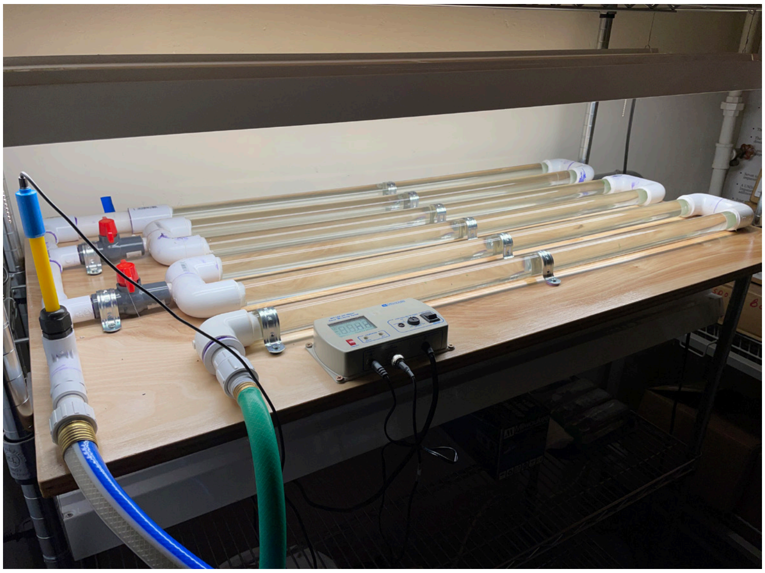

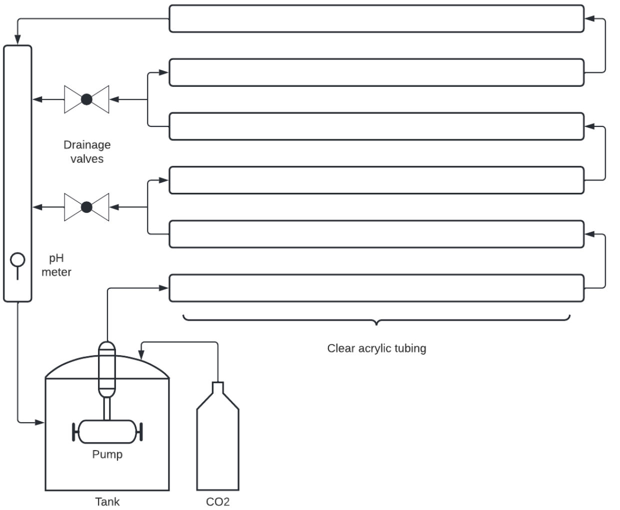

Figure 4. Schematic and example of small-scale tubular photobioreactor. Design and build by Katie Gregor. PHOTO BY K.GREGOR

species of cyanobacteria that live in water and soil and can be grown and utilized to make several products. Potentially, pigments found in cyanobacteria can be extracted for food dyes, pharmaceuticals, and cosmetics. Some metabolites can function as antimicrobials and antifungals.

Furthermore, these photosynthetic microorganisms are considered a potential source of carbohydrates and lipids to produce bioenergy. Certain species are capable of fixing nitrogen with relatively low resource requirements, resulting in a nutrient dense and environmentally friendly fertilizer.

As part of our research interest in these microorganisms, we want to learn more about their biology to better advise nurseries on management and potential uses. After isolating several native species from nursery environments, we began to produce large quantities of one species that had potential as a biofertilizer.

Our student intern in the summer of 2021 helped us accomplish this by designing a bioreactor. Bioreactors provide ideal conditions for microbial growth via open (exposed to outside elements) or closed systems. Closed photobioreactors are a great option to avoid contamination, as exposure to contaminants is minimized. While they tend to be more complex and costly than growth in open ponds, there are options for designs to reduce expenditures. Designing a mid-size closed system photobioreactor for nursery use is not beyond the scope of possibility.

We constructed a small-scale tubular system for demonstrative purposes. Very few energy requirements are needed for its operation. In the summer, this photobioreactor can be left in natural sunlight to avoid electricity costs for lighting.

A small water pump functions to push water through the system, and a CO_2 tank introduces pH control. A pH meter monitors the solution. When the pH grows too high, the pH controller triggers the release of CO_2 into the water tank to bring it back down. This tubular system was cost-effective and built to fit in small indoor spaces during months when sunlight is sparse.

There are a few key components to this design. Six snaking acrylic tubes provide an area for cyanobacteria to be exposed to light. The solution flows in one direction through these tubes and past the pH meter, which transmits the current pH in real-time to a controller. The solution then flows out into a tank containing a bubbler connected to a CO_2 tank and a water pump. Two drainage valves were added for ease of harvest. After draining, one can simply remove the water tank and harvest the cyanobacteria from within for practical use.

There are challenges that come with operating photobioreactors. Depending on the species that is being cultivated, nutrient requirements may vary as well as the pH. For this system, BG-11 growth media was added to supply nutrients. Furthermore, cyanobacteria need relatively calm water to grow.

A high flow rate can inhibit growth, while a flow rate that is exceptionally slow might encourage growth on the sides of the system. Some experimentation is needed to get a photobioreactor of this design running smoothly.

Small-scale photobioreactors such as these provide an opportunity for nurseries to grow their toolset for sustainable agriculture. No two systems will work the same, but following a basic framework allows for designs that avoid excessive costs and produce a viable product.

We hope our work inspires the industry to view cyanobacteria in a new light and to recognize their potential as a component of sustainable ornamental production.

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