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Microbial mats remove nitrogen, phosphorus from aquaculture effluents

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Heterotrophic and autotrophic microbial communities are dominated by cyanobacteria

Microbial mats were supported in floating trays during experiments at the Environmental Research Lab.

Intensive aquaculture systems produce effluents with excess nitrogen, phosphorus, and organic matter that can increase stress and promote disease in cultured organisms. In addition, these effluents contribute to the eutrophication of the water bodies into which they are discharged.

Integrated aquaculture provides a solution to this nutrient loading by introducing plant or animal system components that are able to utilize this waste as nutrients. In addition to decreasing nutrient loading, plant components provide increased dissolved oxygen.

Microbial mats

Microbial mats are heterotrophic and autotrophic microbial communities dominated by cyanobacteria (blue-green algae). They are self-organized laminated structures held together by slimy secretions produced by the microbial components. They naturally occur on soil and water surfaces, and can be cultured for specialized uses.

Microbial mats have been called ideal for bioremediation because they are immobilized ecosystems that allow easy harvest per removal. These conglomerations of species support both aerobic and anaerobic reactions, and are self-sustaining and highly resilient. In tilapia growth experiments, the high protein content in microbial mats has also been shown comparable to that of commercial fish feeds.

Prior research

Scientists at Clark Atlanta University have performed extensive research on the removal of metals and organics using microbial mats. More recently, they also studied the utilization of microbial mats in a recirculating mariculture system with black sea bass. For the duration of the experiment, they maintained an oxygen concentration of 6 to 10 milligrams per liter and ammonia concentrations below 1 milligrams per liter. The university researchers experienced very little build-up of solids and good fish growth, as well.

A separate study by scientists at the Department of Marine Biotechnology in Tijuana examined the bioremediation of shrimp culture effluent using a microbial mat bioreactor. They reported a 97 percent average efficiency for ammonia nitrogen removal and 95 percent for nitrate nitrogen. They also maintained an oxygen concentration of 8 milligrams per liter. Neither study looked at phosphorus removal.

Arizona study

A more recent study at the Environmental Research Laboratory in Tucson, Arizona, USA, analyzed the ability of microbial mats integrated into an aquaculture system to trap nitrogen and phosphorus from the culture water. The experimental design included three stages: laboratory mat construction experiments, laboratory mat nutrient removal experiments, and an experimental application within a fish culture system.

Six mat combinations were constructed in the laboratory from three substrates and two microbial consortia. Two of the successful mat combinations were then analyzed for their ability to sequester nitrogen and phosphorus from the fish effluent in the laboratory. Lastly, the mats were placed in a recirculating aquaculture system for evaluation at a larger scale.

Mat construction, growth

To construct the microbial mats, three components were required: a substrate, microbial inocula, and nutrient medium. The substrates included fescue and paspallum grasses and mesquite, a legume native to the U.S. Southwest. In prior research with constructed microbial mats, only grass had been utilized. The nutrient medium was a modified Allen Arnon nutrient medium.

One microbial inoculum was obtained from scientists at Clark Atlanta University, and the other came from the Barry Goldwater Bombing Range in Gila Bend, Arizona. The substrates were ensiled for 15 to 21 days to produce organic acids, which stimulate cyanobacterial growth. To initiate mat growth, the substrates were then placed on the surface of the nutrient medium and seeded with the mat inocula.

The mats were grown for two weeks under fluorescent lamps for each of the growth experiments. Wet and dry weights were made at the termination of each experiment. The results showed the Clark Atlanta mats grew slightly better than the Gila Bend mats. Also, both inocula had the highest growth with mesquite, then paspallum and fescue.

There are a few possibilities as to why the mesquite containers experienced the most growth. First, mat growth initiated earlier in containers with mesquite. This could be the result of the physical qualities of the mesquite or a difference in the nutrients produced in the ensiling process. Also, the two grasses tended to float on the surface of the medium, whereas the mesquite filled up more of the container. Since the containers were shallow, it is possible the three-dimensional nature of the mesquite leaves promoted more algal growth on the sides and bottoms of the containers.

Mat experiments

The sequestration laboratory experiments used the Clark Atlanta inocula with fescue and Clark Atlanta inocula with mesquite. The mats were placed in plastic containers with 1.5 liters of water from a recirculating system producing cichlids. Water samples collected every five days for 20 days analyzed nitrite-nitrogen, nitrite nitrogen, ammonia-nitrogen, total nitrogen, reative phosphorus, and total phosphorus.

The experimental results for the two treatments were very similar. The majority of the reactive phosphorus was removed from the water for both the treatments and the control alike. For total phosphorus however, there was only a 6 percent decrease for the control and 50 percent decrease for the treatments.

By day 5, all the inorganic nitrogen (nitrate, nitrite, and ammonia) was removed from the water, and 75 percent of the total nitrogen was removed for both treatments. For this reason, a second experiment was performed to evaluate nitrogen removal within the first four days. In the follow-up experiment, total nitrogen removal was 77.4 percent for the mesquite mats, 73.5 percent for the fescue mats, and just 16.6 percent for the control. The initial average nitrate concentration of 14 milligrams per liter was reduced by both treatments to 0.1 milligrams per liter within the first 24 hours.

Greenhouse experiments

Microbial mats were incorporated into a recirculating system housing tilapia at the Environmental Research Lab for the same analyses and treatments used in the laboratory experiments. Three circular tanks were used for each treatment: two repetitions and one control. Incoming water to the tanks was shut off, and the mats were placed into floating trays on the water’s surface. Plastic mesh on the bottom of each tray supported the mats and allowed them access to the water in the tanks.

A 15-day experiment was performed for each treatment with samples collected every three days. The results showed a decrease in nitrogen and phosphorus in all tanks including the controls. In fact, the total nitrogen and total phosphorus levels were significantly lower in the controls at the end of the experiments.

A combination of factors likely contributed to the limited success of the mats in the greenhouse. First, there was a much lower mat:water ratio in the greenhouse. In the laboratory experiments, approximately 24 grams of mat were used to treat each liter of water, while the greenhouse experiments saw 0.45 grams of mat per liter.

Unfortunately, the mat stock became infested with insect larvae after the laboratory experiments, which limited the amount of mat available. The control tanks in the greenhouse also had more prolific algal blooms than the treatment tanks. This was probably a result of increased sunlight to the water column as there were no mat trays floating at the surface. This algal biomass was likely responsible for the higher removal rates in the controls. Still, both the treatments and controls removed nitrogen and phosphorus from the water.

The most notable difference in favor of the treatments for these experiments was in the nitrite concentrations. For example, the fescue treatment maintained a nitrite concentration below 0.2 milligrams per liter, whereas the control increased in nitrite, peaking at 2.4 milligrams per liter on day 12.

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