

Project Title:
The effect of terrestrial leaf litter on sediment organic matter processing and nutrient retention

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\$4900.00

Keywords:
man-made pond; nutrient cycling; metabolism; terrestrial leaf litter; microbes

Abstract:

Man-made ponds represent an important aquatic habitat in Virginia and the Southeastern United States, but little is known about how these systems process terrestrial leaf litter. Terrestrial leaf litter is recognized to be an important source of organic nutrients to aquatic systems and can alter their productivity and the way they retain inorganic nutrients. The proposed project evaluates the effect of terrestrial leaf litter on sediment metabolism and nutrient retention in man-made ponds. We propose a laboratory experiment where we will incubate sediments from man-made ponds under ambient and elevated nutrient conditions with and without terrestrial leaf litter. We will measure the effect of these treatments on the retention of nutrients (N, P, and organic C) by the sediments and on the metabolic rate and growth of sediment microorganisms. These data will allow us to develop a better understanding of the role of man-made ponds in landscape nutrient and organic matter processing, which will inform decisions about nutrient management in regions with abundant man-made ponds like Virginia.

Statement of Objectives and Significance

The abundance of man-made ponds and lakes is increasing globally (Downing et al. 2006) and in regions like Virginia that lack natural ponds and lakes, man-made impoundments represent an important aquatic habitat. Despite the prevalence of man-made ponds in Virginia, there has been little research on how organic and inorganic nutrients cycle in these systems. Aquatic systems are an important component of the processing of terrestrially derived organic matter, and globally, natural aquatic systems process and decompose 70% of their total organic matter inputs (Tranvik et al. 2009). Terrestrial leaf litter supports microbial and animal production and supplies aquatic systems with organic and inorganic nutrients (Gessner et al. 1999). Furthermore ponds play an important role in watershed management due to their ability to retain nutrient run-off from the land (Johnston 1991, Hansson et al. 2005). This retention may be affected by terrestrial leaf litter, since, as it decomposes in aquatic systems, terrestrial leaf litter sequesters inorganic nutrients (Trant et al. 2013). Given that a significant portion of the Chesapeake Bay watershed drains Virginia, understanding how man-made ponds alter nutrient transport is vital to predicting appropriate nutrient management strategies.

Based on data collected by the Fortino lab, man-made ponds near Farmville, VA, have an average of 82 mg of terrestrial leaf litter m^{-2} in the sediments, indicating significant inputs of terrestrial organic matter into these systems. Our proposed research project focuses on understanding the way in which man-made ponds are contributing to the processing of watershed organic matter and nutrients via 2 research questions and 3 objectives:

Research Question 1 - How does the input of terrestrial leaf litter affect sediment metabolism and nutrient retention in the sediments of man-made ponds?

Research Question 2 - How does the input of terrestrial leaf litter alter the sensitivity of pond metabolism to nutrient enrichment?

Objective 1 - Quantify the effect of terrestrial leaf litter on the movement of nutrients across the sediment water interface. We hypothesize that the presence of terrestrial leaf litter will increase the N and P fluxes into the sediments, due to the sequestration of N and P by the fungi on the decomposing leaves and increase the movement of dissolved organic nutrients out of the sediments due to the release of dissolved organic nutrients from the leaves (i.e., leaching).

Objective 2 - Quantify how terrestrial leaf litter alters microbial (bacteria and fungi) biomass and respiration. We hypothesize that terrestrial leaf litter will increase microbial abundance and respiration, due to the release of dissolved organic matter from the decaying leaves, and growth on the leaf surface.

Objective 3 - Quantify how terrestrial leaf litter alters the response of sediment respiration to nutrient enrichment. We hypothesize that terrestrial leaf litter will increase sediment respiration when the sediments are enriched with N and P, since litter associated fungi have been shown to be more nutrient limited than sediment microbial communities.

Technical Approach and Outcomes

To address our research questions and objectives, we have devised a single laboratory experiment that allows us to simultaneously manipulate the availability of terrestrial leaf litter and inorganic nutrients to the sediments of a man-made pond, while also measuring changes in nutrient movement and microbial abundance. Our experimental set-up will consist of 20, 300 ml septum topped glass jars filled with an approximately 4 cm layer of sediment collected from a local, man-made pond. The sediment samples will be run through a 250 μm mesh sieve to remove any terrestrial leaf litter particles and macroinvertebrates that may be present. The jars' remaining volume will be filled with pond water from samples collected at the time of initial sediment sample collection.

To create the terrestrial leaf litter treatments, 0.5 mg of senescent tulip poplar leaf disks will be added to the sediment surface of the treatment jars. To ensure that the mass of organic matter remains consistent between the treatments, the control jars (no leaf litter) will receive sufficient sieved pond sediment equal to the mass of organic matter added by the leaves.

The nutrient addition treatment will be created by adding NO_3 and PO_4 to the desired treatment jars until the water concentration of the jars is twice the concentration measured in the original water samples from the pond. The control treatments, those without added nutrients, will remain unaltered. The terrestrial leaf litter and nutrient treatments will be replicated 5 times and crossed in a complete factorial design (Table 1).

Table 1. Experimental design of the proposed complete factorial experiment. Rows indicate the levels of the terrestrial leaf litter treatment, columns indicate the levels of the nutrient addition treatment, and the cells indicate the number of replicate jars in each treatment combination.

	Ambient Nutrients	N + P Addition
No Terrestrial Litter	5	5
Terrestrial Litter Addition	5	5

To evaluate the effect of terrestrial leaf litter on the nutrient and microbial dynamics of the system, we will measure changes in five response variables:

- 1) inorganic nitrogen (NO_3 and NH_4) and soluble reactive phosphorus flux,
- 2) microbial respiration (measured as sediment oxygen demand),
- 3) dissolved organic matter flux (measured as near UV and visible light absorbance),
- 4) bacterial abundance, and
- 5) fungal biomass (estimated from ergosterol extraction).

Sediment samples will be collected from a local, man-made pond with an Ekman dredge. Once back at the lab, the sediment samples will be run through a 250 μm sieve to remove any terrestrial leaf litter particles and macroinvertebrates that may be present. Next, 300 ml septum topped glass jars will be filled with a 4 cm layer of the sieved sediment. The jars' remaining

volume will be filled using the pond water samples collected with a Van-Dorn sampler simultaneously with the sediment sample.

The previously described treatments will be applied to the jars. Following the treatments, the sample jars will be incubated, uncovered at a constant temperature in darkness for 6 weeks. For the first week the jars will be sampled daily, and then weekly for the remaining 5 weeks. During each sampling event, 25 ml of water will be removed from the jar and filtered through a 0.7 μm glass fiber filter. One ml of the water will be used to measure absorbance from 250 - 900 nm on a Nanopore Spectrophotometer, while the remaining 24 ml will be frozen for nutrient analysis at Virginia Commonwealth University. A 5 ml sample will be filtered onto a black membrane filter (0.2 μm) and stained with a fluorescent nuclear stain (DAPI) for quantification of bacterial abundance. Bacteria will be counted on the stained filters using an epifluorescent microscope. In order to quantify sediment oxygen demand, we will refill the jars with pond water and then seal them without an airspace using a septum top lid. After 1, 3, and 6 hours of dark incubation, a 15 ml sample will be extracted from the jar using a glass syringe. Pond water will be simultaneously introduced to the jars with a second syringe while the samples are taken so that air will not be introduced into the jars during sample extraction. The oxygen content of each sample will be determined using the Winkler titration method (Carpenter 1965) and the sediment oxygen demand will be calculated as the change in oxygen concentration of the water over time. At the conclusion of the 6 weeks, fungal biomass on the terrestrial leaf litter discs will be measured by extracting ergosterol (a fungi specific molecule) in methanol. The extracted ergosterol concentration will be measured by an outside laboratory. The data we collect will be useful for developing a quantitative understanding of the way that man-made ponds alter the processing of inorganic nutrients and organic matter. This understanding will allow for better landscape nutrient management in regions with abundant man-made ponds like Virginia.

In our proposed timeline, we plan to begin sediment and pond water sample collection and creation of treatments in early June 2014. By late June/ early July, we will collect the sediment oxygen demand and absorbance samples' data. In the fall, bacterial abundance data will be collected and nutrient and ergosterol samples will be sent for analysis. During the winter of 2014/2015, the data will be analyzed and the manuscript describing our findings will be prepared. In the spring of 2015, the manuscript will be finalized and we will prepare our results for presentation at a national professional meeting (The Society for Freshwater Science).

Project Budget

Item	Amount Requested from VWRRC
<i>Laboratory Costs</i>	
Nutrient Analysis (200 samples @ \$18/sample)	\$ 3,600
Ergosterol Samples (20 samples @ \$50/sample)	\$ 1,000
<i>Supplies</i>	
Bacterial Abundance Quantification	
DAPI Nuclear Stain for Bacterial Counts	\$ 200
Membrane Filters	\$ 100
<i>Total</i>	\$ 4900

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Education

- High School Degree – Dec. 2012
Spyglass Academy, Stafford, VA
- 49 credits earned (36 were D.E. credits) – Jan. 2012 - Jun. 2013
Germanna Community College, Fredericksburg, VA
- 63 credits earned towards Biology Major, Chemistry Minor – Aug. 2013- Present
Longwood University, Farmville, VA

Experience

- Experience with Leaf Decomposition Rates in Man-made Ponds
 - Student Assistant in Fortino Laboratory
 - PRISM Program
- The National Society of Leadership and Success
 - Sigma Alpha Pi
 - New Member