

**Project Name:** Quantifying Phosphorus Uptake and Release from Periphyton and Phytoplankton Communities

**Project Purpose:** Estimate the contribution of periphyton to Phosphorus (P) cycling in the Stormwater Treatment Areas (STAs) specifically in outflow areas where P concentrations are very low (<20 micrograms per liter,  $\mu\text{g L}^{-1}$ ).

### **Introduction:**

The ability of STAs to achieve low outflow total P (TP) concentrations depends on the system's capacity to reduce P by transforming it from one form to another. One set of transformations is P uptake and release by periphyton or phytoplankton. This transformation may be very important, particularly in low-P conditions (i.e., near the STA outflows).

P uptake and release in the periphyton/phytoplankton community in the STAs have not been measured. The influence of periphyton/phytoplankton communities on TP concentrations in surface water at mid-flow and outflow regions within the STA flow-ways may differ depending on the dominant vegetation community and consequently, differences in periphyton community. For example, TP concentrations in surface water at the mid-flow and outflow regions of submerged aquatic vegetation (SAV) dominated flow-ways in STA-2 are higher under stagnant conditions than flowing conditions. In flow-ways dominated by emergent aquatic vegetation (EAV), this increase was not observed.

Phase I of this study consisted of a literature review that compiled numerous methods to evaluate growth, uptake, and release of P from the periphyton community (Laughinghaus et al. 2019, Appendix A). Each method has advantages and disadvantages. After consideration of these methods, Phase II was developed to measure bioavailability of dissolved organic phosphorus (DOP) and nitrogen (DON) using periphyton and water from outflow regions of an STA. Also, during Phase II, the applicability of utilizing more advanced genetic research techniques (i.e. metagenomics) was evaluated in a short-term field review study. Metagenomics measures DNA and can indicate the potential nutrient cycling ability of the periphyton.

### **Objectives:**

1. Estimate DOP and DON removal by periphyton and phytoplankton in outflow region of STA treatment flow-ways where TP concentrations are very low (<20 micrograms per liter [ $\mu\text{g L}^{-1}$ ]).
2. Estimate periphyton and phytoplankton growth and senescence rates in STAs where TP concentrations are very low.
3. Evaluate the influence of periphyton and phytoplankton within different dominate vegetation communities (e.g., SAV, EAV).

### **Applications of Findings:**

Estimates of DOP and DON uptake will indicate how labile this material is and determine if periphyton are key to its removal from the water column. This study will generate information critical to understand P reduction mechanisms by periphyton/phytoplankton and potential

factors affecting them. Data may support management strategies to achieve lower outflow concentrations. The study supports the Restoration Strategies Science Plan (SFWMD, 2018) to enhance the understanding of mechanisms and factors that affect P removal, particularly those that are key drivers to performance at low TP concentrations (<20 µg/L).

#### **Relevant Science Plan Key Questions & Sub-questions:**

- How can internal loading of P to the water column be reduced or controlled, especially in the lower reaches of the stormwater treatment areas (STAs)?
- How can the biogeochemical or physical mechanisms, including internal flux of P, be managed to further reduce soluble reactive P (SRP), particulate P (PP), and DOP concentrations at the outflow of the STAs?
  - What are the key physicochemical factors influencing P cycling in very low-P environments?
  - What are the sources, forms, and transformation mechanisms controlling residual P pools within the STAs, and how do they compare to the natural system?

#### **Proposed Approach**

##### **Phase I. Literature Review**

Completed – a list of various techniques to evaluate periphyton P uptake, growth, turnover and release were compiled as were the findings from the microbial-related research conducted in the STAs.

##### **Phase II. Bioavailability Study and Short-term Metagenomics Field Review Study**

Completed - Based on the literature review and evaluation of laboratory experiments to measure nutrient uptake, the two studies outlined in this Phase II were developed to evaluate the impact of the periphyton community on nutrient cycling.

The Bioavailability study was designed to measure changes in concentrations of the relatively recalcitrant DOP and DON concentrations in the surface water by the periphyton communities. Periphyton collected from outflow regions in both SAV and EAV communities was added to flasks containing surface water then placed into growth chambers and incubated under light or dark conditions. Water quality was measured every 7-days over the 21-day incubation period. The trends in the water quality changes, attributed to the periphyton communities, provided indications of the bioavailability of the nutrients and the impact the periphyton has on cycling these nutrient forms.

Also tested in Phase II was the applicability of using advanced genetic research approaches, termed bioinformatics, in understanding the periphyton community structure and the potential of the community to act upon specific forms of organic nutrients. One of these approaches, metagenomics which measures DNA in the samples, was used to review the species composition of the periphyton. Results from the short-term study showed novel insights into the physiological potential of the periphyton communities is possible through this type of analysis.

### Phase III. Further Field and Laboratory analyses

Field and laboratory studies based on the findings from Phases I and II are currently ongoing. This study utilizes metagenomics to identify the community composition of the periphyton community in the field over various flows and seasons between SAV and EAV communities at inflow and outflow regions of an STA. Laboratory studies utilized metatranscriptomics to examine what genes of the periphyton community are actively being transcribed and how they respond to changes in P availability. Genetic composition of the periphyton community and gene transcription rates are instrumental to analysis of their influence on nutrient cycling in the STA's.

### Project Deliverable and Schedule

Phase #	Description	FY 20	FY21				FY22				FY23			
		Q4	Q1	Q2	Q3	Q4	Q1	Q2	Q3	Q4	Q1	Q2	Q3	Q4
1	Literature Review (complete)													
2	Bio-availability Study (complete)	X	X	X	X									
3	Field and Laboratory Analyses						X	X	X	X	X	X	X	X

### References

- Laughinghouse H. D., D. E. Berthold, M. Barbosa and F. W. Lefler. 2019. A review on tropical and subtropical periphyton and phytoplankton processes and methods of quantification. Prepared for the South Florida Water Management District SFWMD. West Palm Beach, FL. 87 pp.
- DB Environmental, 2021. Microbial Processes Affecting Phosphorus Removal and Retention in the Everglades Stormwater Treatment Areas. Prepared for the South Florida Water Management District SFWMD. West Palm Beach, FL. 142 pp.
- Pietro, K. C., K. Inglett and A. Wright. 2023. Periphyton enzymatic activities in the water column along internal low-phosphorus nutrient gradients in the Everglades Stormwater Treatment Areas. Ecological Engineering 196: 107100.