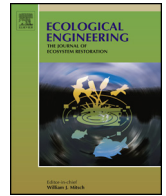




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## High diversity within the periphyton community of an algal turf scrubber on the Susquehanna River

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### ABSTRACT

Algal turf scrubber (ATS) systems have been evaluated for their ability to remove dissolved nutrients from a variety of natural waters and agricultural wastewaters. Although these systems have been well characterized with respect to productivity and nutrient removal, very little is known about the community structure and population dynamics of the underlying algal community. The objectives of this study are to describe the nutrient removal performance of a pilot-scale ATS adjacent to the Susquehanna River along with a detailed analysis of structure of the algal turf community. ATS biomass production varied seasonally, with highest values in July and August. Total production ranged from 11 to 18 g DW m<sup>-2</sup> day<sup>-1</sup> with a mean of 12.3 ± 1.6 g DW m<sup>-2</sup> day<sup>-1</sup> over the eight month growing season. Average N and P removal rates fluctuated more than 2-fold with highest values (350–450 mg N m<sup>-2</sup> d<sup>-1</sup> and 25–40 mg P m<sup>-2</sup> d<sup>-1</sup>) during the two month summer period (July/August) and lowest values (30–50 mg N m<sup>-2</sup> d<sup>-1</sup> and 3–5 mg P m<sup>-2</sup> d<sup>-1</sup>) in the early winter period (November/December). Results show high species diversity within the scrubber (182 species within the 28 m<sup>2</sup> growing surface). High diversity is likely due to a combination of factors: species diversity in the source community (the Susquehanna River), spatial heterogeneity of the ATS system, and vertical structure of the turf community. Analysis for traits for mode of attachment, habitat of origin and life form was used to illustrate the distribution of adaptations of species in the algal turf community.

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### 1. Introduction

The algal turf scrubber (ATS) is a form of ecotechnology for improving water quality of eutrophic aquatic ecosystems (Adey and Loveland, 2007; Adey et al., 2011). The system is a shallow, flowing raceway over which polluted water is pumped. Algae grow attached to screens placed in the bottom of the raceway and, in doing so, take up nutrients and other pollutants as the water flows through the system. The nutrients and pollutants are removed from the system when the algae are harvested. Because of the relatively fast growth rate of attached algae, the ATS can remove pollutants, and thereby improve water quality, at a rapid rate. Along with other attached algae-based water treatment systems (Debusk et al., 2004; Houser

et al., 2011; Markou and Georgakakis, 2011; Vymazal 1989), ATS systems have been tested in a variety of settings and locations (Adey et al., 1993, 2013; Craggs et al., 1996; D'Aiuto et al., 2015; Kangas and Mulbry, 2014; Mulbry et al., 2008, 2010; Sandefur et al., 2011) and have generally been found to be an effective alternative “best management practice” for improving water quality. An important advantage of attached algae-based systems relative to suspended algae-systems is that the algal biomass can be harvested more readily because it is easier to separate the biomass from the water used in cultivation. As evidence of their functional values, the ATS has recently been recommended to the U. S Environmental Protection Agency for use by land owners in the Chesapeake Bay watershed to meet their total maximum daily load discharge requirements (Chesapeake Bay Program, 2015).

Most ATS research to date has focused on nutrient removal capacities in different locations or using different water sources. Although the structure and dynamics of the algal communities are

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crucially important for understanding ATS performance (Cardinale, 2011), ATS algal communities responsible for nutrient removal have received little attention. Typically, dominant algal taxa in the turf community are listed, and occasionally, more detailed species lists are reported (D'Aiuto et al., 2015).

Trait analysis has emerged over the last decade in Ecology as an approach for providing insight into how communities are assembled through self-organization (Kraft et al., 2007; McGill et al., 2006; Violle et al., 2007). A trait is an identifiable or measurable property of an organism that can affect survival and reproduction (Shipley 2010). Traits can be either functional, such as growth rate, or morphological, such as cell shape. Trait analysis has been applied to phytoplankton communities (Edwards et al., 2013; Litchman and Klausmeier, 2008; Reynolds, 1988, 2006) but it has not been used as an aid for understanding the assembly of the community of species on the algal turf. The objective of this study is to describe the nutrient removal performance of an ATS along with a detailed analysis of structure of the algal turf community. Diversity of the ATS algal community is described with species lists and with a preliminary analysis of species traits. Here, morphological traits are assigned to species in order to provide a more in-depth description of the algal community that is being managed for nutrient removal function.

## 2. Materials and methods

### 2.1. Site characterization

The research was carried out at the Muddy Run Pumped Storage Hydroelectric Facility on the lower Susquehanna River in southeastern Pennsylvania. At this facility water is pumped from the river to the Muddy Run Reservoir at night when the electricity price is low. Water is then released from the reservoir and run through turbines as it flows by gravity back to the river to generate power during the day when the electricity price is high. Influent water quality (nitrate-N, phosphate-P, TKN, TP) was monitored monthly from April to October 2009. Values varied nearly 3-fold (from 0.4 to 1.4 mg l<sup>-1</sup> and 0.03–0.09 mg l<sup>-1</sup> for NO<sub>3</sub>-N and PO<sub>4</sub>-P, respectively), with lowest values in the July and highest values in the fall and spring (Supplementary material, Fig. 1). Influent and effluent water temperature values varied from 5 to 30 °C, with effluent values generally 2–3 °C higher than influent values (Supplementary material, Fig. 2).

### 2.2. Algal turf scrubber system

#### 2.2.1. Construction and operation

A pilot-scale algal turf scrubber (ATS) raceway (0.3 × 91 m, 28 m<sup>2</sup> growing area) (Hydromentia, Ocala, FL) was installed at the Muddy Run Pumped Storage facility on the Susquehanna River near Holtwood, Pennsylvania. The elevated aluminum raceway was installed at a 2% slope with 6 mm nylon mesh screen (Industrial Netting Co., Minneapolis, MN) lining the bottom of the raceway. The raceway received water from the Muddy Run Reservoir at a flowrate of approximately 60 L min<sup>-1</sup> via a 10 L surge box at the top of the raceway. The surge box released a pulse of water every 10–12 s. Effluent from the raceway drained through a nylon mesh bag at the bottom of the raceway prior to draining into the river.

#### 2.2.2. Biomass collection and analysis

Algal biomass was harvested at one to three week intervals using a portable wet/dry vacuum. Prior to each harvest, water flow to the ATS was turned off and the scrubber was allowed to drain for 30–60 min. In order to identify possible longitudinal differences in the algal community and/or in growth and nutrient removal, biomass was harvested from three different locations along the 91 m raceway. Biomass was harvested from two adjacent sections

of the scrubber near the top of the system, two sections in the middle and two sections at the bottom of the raceway. Each individual section had an area of approximately 0.9 m<sup>2</sup> (0.3 × 3 m). The entire ATS area (except for a section immediately below the surge box which was maintained as a non-harvested refuge) was harvested but only the six biomass samples from the top, middle and bottom sections were retained for dry mass and nutrient analyses. Algal biomass not retained for sample analysis was discharged into the Susquehanna River below the ATS.

After harvest, the resulting slurries were dewatered by sieving the harvested material through 3 mm nylon mesh bags (Pentair Aquatic Ecosystems, Apopka, FL). The biomass retained in the bags (termed screened solids) was air-dried at 25 °C using an electric fan, followed by drying 70 °C prior to mass determination and further analysis. Biomass in the sieved water (termed greenwater solids) was also quantified by measuring the volume of the greenwater, and determining the dry weight after drying a 1 Liter aliquot at 70 °C. Biomass production values (or net primary production values) were calculated by dividing the oven-dried mass by the number of days between harvest dates. Throughout the study 235 screened solids samples and 105 greenwater solids samples were collected and analyzed.

In order to quantify the amount of biomass that breaks off from the turf between harvests (termed slough solids) we attached a nylon mesh bag to the outlet drain pipe at the bottom of the ATS. Material collected in the bag was removed at the time of harvest and dried as described above. To calculate the slough production, the biomass collected between harvest dates was divided by the total area of the scrubber and by the number of days between harvests. Throughout the study 23 slough samples were collected and analyzed.

Dried harvested solids were ground in a Wiley Mill to pass a 3 mm sieve and stored in sealed plastic bags at 20–25 °C prior to analysis for moisture, ash, total Kjeldahl nitrogen (TKN), and total phosphorus (TP). Sample moisture content was determined after drying overnight at 65 °C. Samples were analyzed for TKN and TP content by flow injection analysis (Lachat Instruments, Milwaukee, WI) after digestion in concentrated H<sub>2</sub>SO<sub>4</sub> for 1 h at 400 °C.

#### 2.2.3. Analysis of algal community structure

Algal samples from the scrubber were collected periodically and manually examined with a compound microscope for the purpose of describing the qualitative structure of the community at the time of harvest. Two or three samples from each turf along with a sample of the green water and a sample of the sloughed material were routinely collected during each harvest and these were examined to assess the dominant alga taxa.

Samples were collected for a more in-depth assessment of the species composition of the turfs on three dates in the summer of 2008 (July 23, Aug 1, and Aug 13) and four dates in 2009 (March 7, June 12, July 17, and Oct 9) in order to construct a species list of algal taxa found on the scrubber. These samples were brought live to be analyzed at the National Museum of Natural History. Samples were analyzed live and/or preserved in 4% formaldehyde or in Lugol's solution.

Samples were analyzed between a slide and coverslip using an Olympus BX50 binocular light microscope in duplicate for each sample site. For diatom identification, a sub-sample (20 mL) was boiled in hydrogen peroxide 30% in a 1:1 sample ratio, adding potassium dichromate during the process to speed oxidization, until returning to the original volume. The cleaned material was concentrated in a centrifuge, and rinsed several times until reaching a neutral pH. This material was diluted into 3 different solutions, air dried on coverslips, and processed for either light or scanning electron microscopy (SEM).

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