Implementation of Mycoremediation Techniques to Reduce Aquatic Contamination of Polycyclic Aromatic Hydrocarbons and Pesticides in the Winooski River Watershed

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INTRODUCTION/BACKGROUND

Background – Ecology:

PAHs and pesticides are both present in the Winooski River Watershed and Lake Champlain Basin due to anthropogenic influence. Both classes of compounds have negative influence on the health of freshwater ecosystems and terrestrial organisms that rely on these water resources, and PAHs are potentially harmful to human health. Research has shown that both categories of compounds can be successfully removed (or reduced) using fungal bioremediation/mycoremediation, however no such study has been conducted in the Winooski.

Polycyclic Aromatic Hydrocarbons (PAHs):

- PAHs are chemicals produced when crude oil, gasoline, or coal are burned
- PAHs are thermodynamically stable, highly toxic and carcinogenic, and have low aqueous solubility
- PAHs can cause severe ecotoxicological issues due to their persistence and tendency to bioaccumulate

PAHs enter the Winooski River Watershed from various non-point sources, including runoff from roads where coal-tar based pavement sealants have been applied, and runoff from older roads with remnants of tire wear. There are specific sources in the urban areas of Chittenden County, including (but not limited to) the Farwell Street Dump recycled asphalt pile, the abandoned coal yard near Steven's Branch, outfall from wastewater treatment facilities (Burlington Inner Harbor), oil barge offloading sites (historic and present, Burlington Inner Harbor), and biorefinery wastewater from the McNeill Generating Station in the Intervale.

	Compound	Molar mass (g mol^{-1})	Cs (25 °C) (mg L^{-1})	Log Kow	Concentration in BRW (µg L^{-1})	Recommendations [a] CEQG (µg L^{-1}) * (mg kg^{-1} DW) #	Health and environmental risks *
Phenols	PCP	266.3	14	5.1	23,800 ± 2380	0.5* 7.6#	Carcinogenic Very toxic to aquatic organisms
	TCP	231.9	1×10^3	4.4	4380 ± 526	n.d.* 0.5#	
PAHs	BAA	228.3	9.4×10^{-3}	5.7	18.8 ± 2	0.018* n.d.#	Carcinogenic, teratogenic toxic to aquatic organisms
	FLU	202.3	0.26	5.1	121.4 ± 14	0.04* n.d.#	Toxic to some organs Toxic to aquatic organisms
	PHE	178.2	1	4.4	212 ± 24	0.4* n.d.#	
	PYR	202.3	0.13	4.8	86.8 ± 10	0.025* n.d.#	Toxic and bioaccumulation effect in fish, molluscs and algae

Table 1. The physiochemical properties of PAH's (and Phenols) and their respective effects on aquatic life

Pesticides

- The U.S. has few regulatory limits on allowable concentrations of pesticides in surface waters
- Pesticides are designed to be biologically active, meaning they are likely to have negative effects on biota at far lower concentrations than designated toxic levels.
- High water solubility, persistence, and toxicity to aquatic insects and Macroinvertebrates

Pesticides enter the watershed through agricultural and urban runoff, specifically from golf courses, lawns, and home gardens (urban). Aerosolized pesticides drift during application on agricultural areas and settle on surface waters or terrestrial ecosystems.

In the Lake Champlain Basin and Winooski River Watershed, high concentrations of metolachlor (herbicide) pose a concern for aquatic biota, and neonicotinoid insecticides also warrant special attention.

OBJECTIVES

Hypotheses:

- Mycoremediation in the Winooski River Watershed will be effective at lowering levels of PAHs and pesticides entering the water through urban and agricultural influence.

Research Question:

- What types of fungal bioremediation will be most effective at removing polycyclic aromatic hydrocarbons (PAHs) and pesticides from agricultural and urban runoff into the Winooski river watershed?

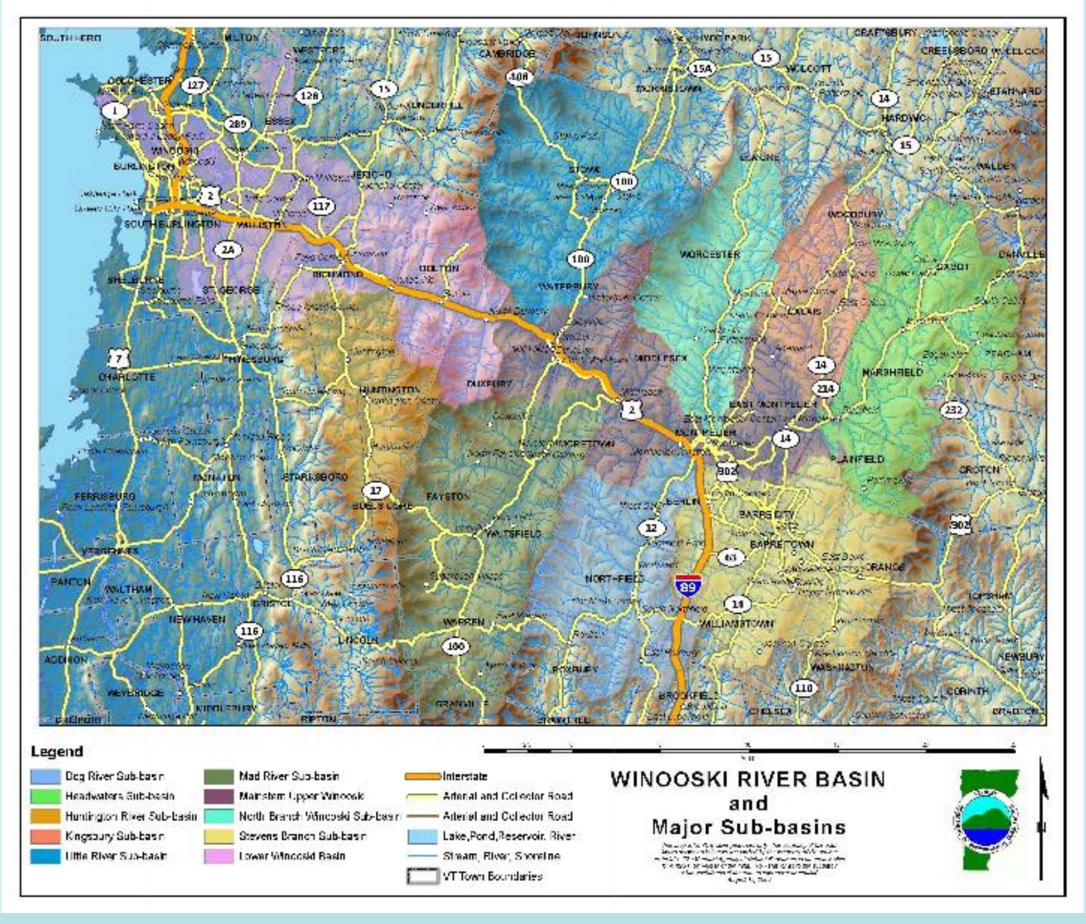

Fig. 1. Map of the Winooski River Basin and Sub-Basins

Objectives:

- Assess the effectiveness of mycofilters in preexisting structures (wastewater retention ponds, abandoned dams) as a curative and preventative approach to reduce the presence of PAH's and pesticides in the Winooski River Basin. If effective, mycofiltration could be implemented on roadsides and storm drains as well.

METHODS

Experimental Design:

We plan on conducting an experimental study to test the effectiveness of using "mycofilters" on highly impacted stream banks, stormwater retention ponds, and abandoned dams to absorb PAHs and pesticides. Mycofilters will be constructed from burlap sacs filled with a wood chip or glucose-based substrate that has been inoculated with one of the following species of fungi: Pseudogymnoascus spp., Aspergillus caesiellus, Trametes hirsuta IBB 450, Phanerochate chrysosporium ATCC 787, Pleurotus ostreatus MTCC 1804 or Cadophora spp.

T. hirsuta and Pseudogymnoascus spp. are shown to be the most effective mycoremediators in isolated trials on industrial wastewater (Ariste et al., 2017), but all six species/families demonstrate a 99% removal rate under a controlled environment where temperature, pH, and glucose levels can be controlled and manipulated. Since the environments that previous studies have used to evaluate the efficiency of mycoremediation are mainly isolated and man-made, we hypothesize that the relative effectiveness of this technique will be lower on riverbanks and in wastewater retention ponds due to our inability to manipulate the environment.

	Phylum or subphylum	Organic chemicals degraded	Major ecological characteristics
Basal fungal lineages	Microsporidia		Obligate parasites of animals
	Kickxellomycotina (2)	PAHs	Saprobes, and parasites of animals and fungi
	Zoopagomycotina		Parasites of nematodes, protozoa and fungi
	Entomophthoromycotina (2)	PAHs	Parasites of insects
	Blastocladiomycota		Saprobes, and parasites of plants and animals; aquatic and terrestrial
	Mucoromycotina (16)	Benzoquinoline, biphenyl, PAHs, pesticides, synthetic dyes and TNT	Saprobes, parasites or ectomycorrhizal symbionts
	Neocallimastigomycota		Gut symbionts of ruminant herbivores
	Chytridiomycota (2)	PAHs	Saprobes, and parasites of plants and animals; fresh water and wet soil
	Glomeromycota	PAHs and pesticides	Arbuscular mycorrhizal symbionts
Dikarya	Ascomycota (88) Pezizomycotina (57)	Alkanes, alkylbenzenes, biphenyl, chlorophenols, coal tar oil, crude oil, diesel, EDCs, fragrances, PAHs, PCDDs, pesticides, synthetic dyes, TNT and toluene	Saprobes, pathogens of plants and animals, and symbiotes of algae (lichens), plants (ectomycorrhizae, ercoid mycorrhizae and endophytes) and insects; terrestrial and aquatic
	Saccharomycotina (9)	Alkanes, alkylbenzenes, biphenyl, crude oil, EDCs, PAHs and TNT	
	Other ascomycetes (22)	Alkanes, diesel, coal tar oil, crude oil, MTBE, PAHs, pesticides, RDX, toluene and synthetic dyes	
	Basiodiomycota (53) Agaricomycotina (50)	Alkanes, BTEX compounds, chloroaliphatics, lignols and phenols, crude oil, coal tar, EDCs, PAHs, PCBs, PCDDs, PCDFs, personal care product ingredients, pesticides, pharmaceutical drugs, RDX, synthetic dyes, synthetic polymers, TNT and other nitroaromatics	Saprobes, ectomycorrhizal symbionts, pathogens of plants and animals, and parasites of other fungi; terrestrial and aquatic
	Pucciniomycotina (3)	Cresols, crude oil, dibenzothiophene, PAHs and RDX	

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Table 2. Phylum and subphylum of fungus and the compounds that they effectively remediate.

Sites where mycofiltration is implemented will be monitored over a 6-month period from May to October, when mean temperatures are adequate for supporting maximum fungal growth. Each site will be assigned a sister site, which will have the same type of stormwater management system (dam, retention pond, or natural bank) but no implementation of mycofilters to act as a control site. Since each site will inherently have different levels of pollutants and different sources (point or non-point), measurements will be taken for each site biweekly beginning 12 weeks before mycofilters are placed to establish a dataset for each site highlighting the general influx and average levels of PAHs and pesticides present in the water. Post-implementation of mycofilters, levels of PAHs and pesticides will be measured bi-weekly, on the same day for sister sites, and within 48 hours of a rain event.

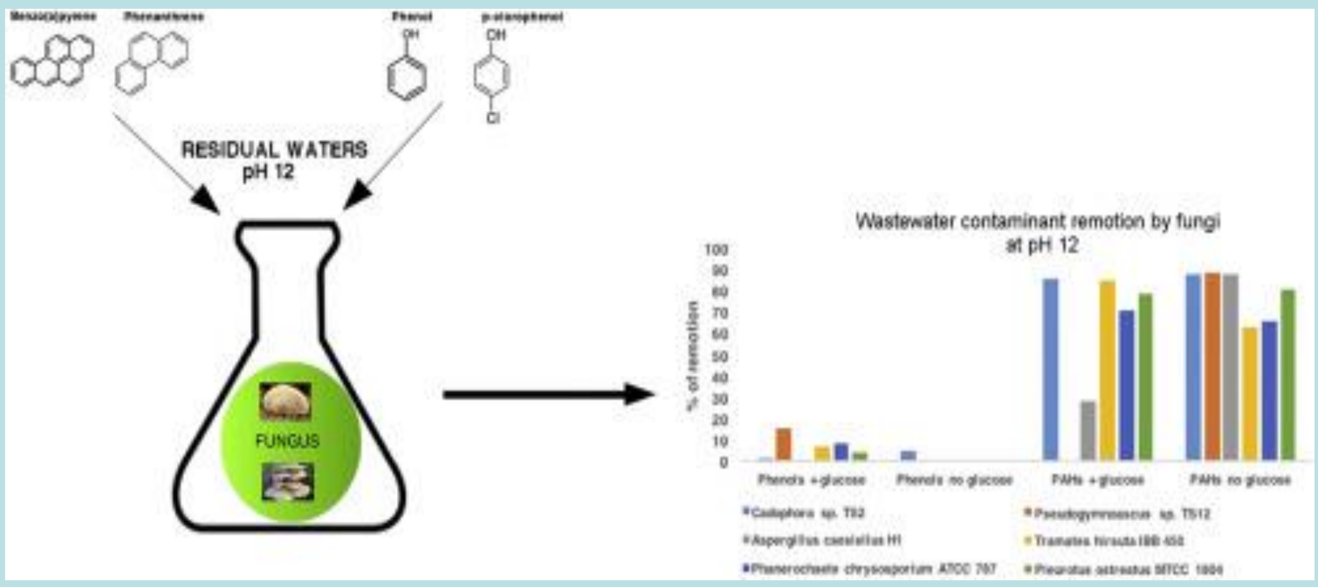

Fig. 2. Remotion of wastewater effluents in pH 12 by various fungal species with and without the presence of glucose

MANAGEMENT IMPLICATIONS

Environmental Impacts:

The proposed mycofiltration plan is expected to have very little environmental impact because no chemicals or heavy machinery are necessary for effective implementation. Using existing stormwater infrastructure eliminates the need for further environmental disturbance.

Cost:

Mycofilters would be low cost, low maintenance, and easy to implement in existing stormwater management infrastructure. Waste can be diverted from commercial mushroom growing operations and re-used to inoculate mycofilter substrates. There is potential to partner with local forestry programs and recycling centers to acquire cheap, recycled, uncontaminated fungal substrates (wood chips or pulp).

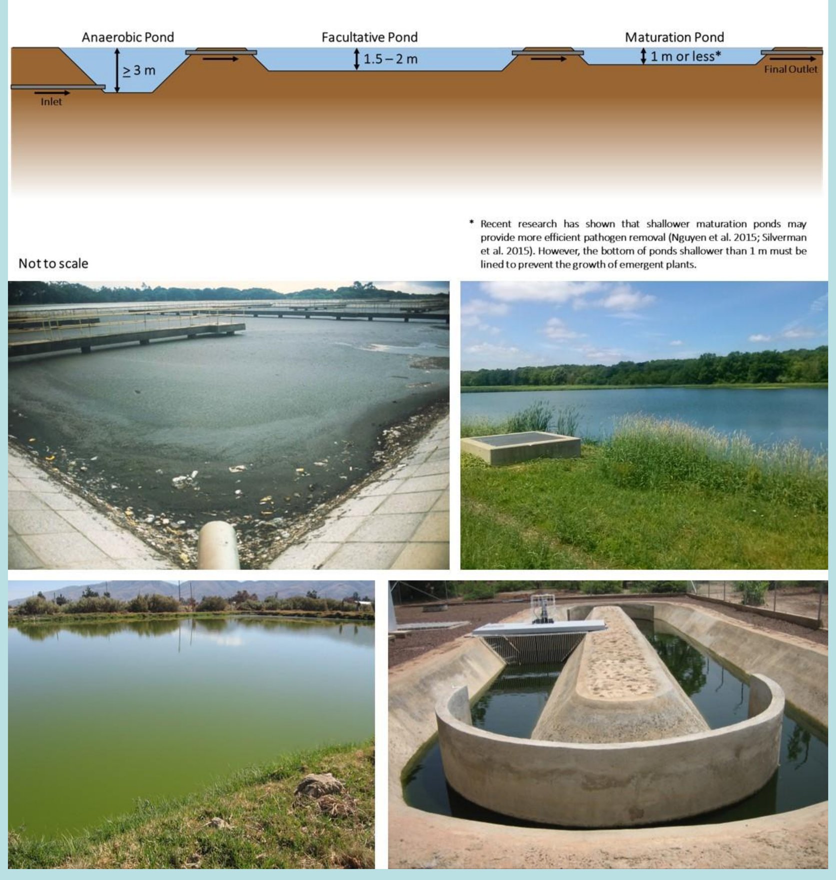

Fig. 3. Types of Wastewater Retention Ponds

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