Bioremediation of Acid Mine Drainage Using *Pleurotus ostreatus* Mycelium as a Biosorbent Material

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Introduction

- This project focused on utilizing a natural remedy to bioremediate Acid Mine Drainage (AMD).
- AMD is an acidic solution that contains large amounts of heavy metals such as iron and manganese.[1]
- Mycoremediation is a field of bioremediation that uses fungi to biodegrade and adsorb environmental pollutants.[4]
- Using mycelium could be a novel approach that is cheaper and more effective than current methods used to remediate AMD.

Figure 1: Acid Mine Drainage are from abandoned coal mine in PA. (The Foundation for PA Watersheds, 2022).

Background

- Acid Mine Drainage
  - Environmentally damaging mine effluent that contains sulfuric acid, heavy metals, and iron sulfides.[1]
- *Pleurotus ostreatus:* oyster mushrooms
  - The mycelium of this fugus is capable of forming coordination complexes with heavy metals.[3]
- Aquatic biofilms
  - Matrix of algae, fungi, protozoans, & bacteria
  - Good indicator of overall health of aquatic microbiome

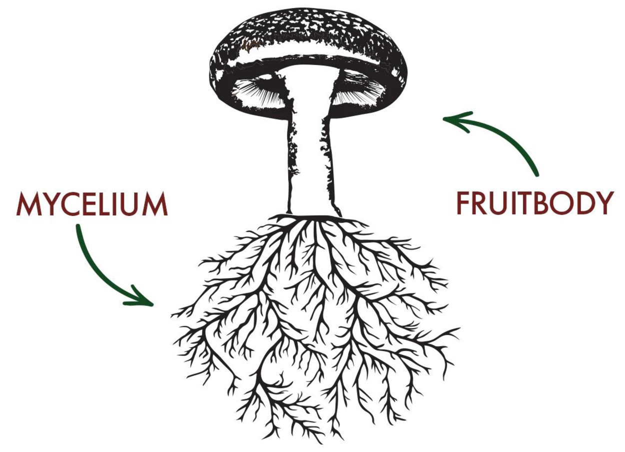

Figure 2: An illustration showing the difference between the mushroom fruiting body and mycelium (Stamets, 2021).

Hypothesis

P. ostreatus mycelium should be able to filter iron, other precipitated metals, and iron oxidizing bacteria from AMD which should lower iron concentrations and promote the growth of healthy biofilms.

Experimental Design

- AMD and healthy stream water were collected from a mine site in Coaldale, Pennsylvania.
- These water samples were then put into 2L fish tanks.
- The tanks were placed on a magnetic stirrer at 60rpm to simulate flow, a slide holder with slides was attached to the side of the tank, and subjected to light and dark cycles of 12hr/12hr.
- Biofilms were allowed to grow for 3 weeks.
- The biofilms were stained with 1% crystal violet then imaged.
- The stained slides were soaked in 30% acetic acid to dissolve the crystal violet then this solution was analyzed in a UV-Vis spectrophotometer.

Experimental Tanks
AMD (negative control)
AMD with mycelium
Creek with mycelium
Creek (positive control)

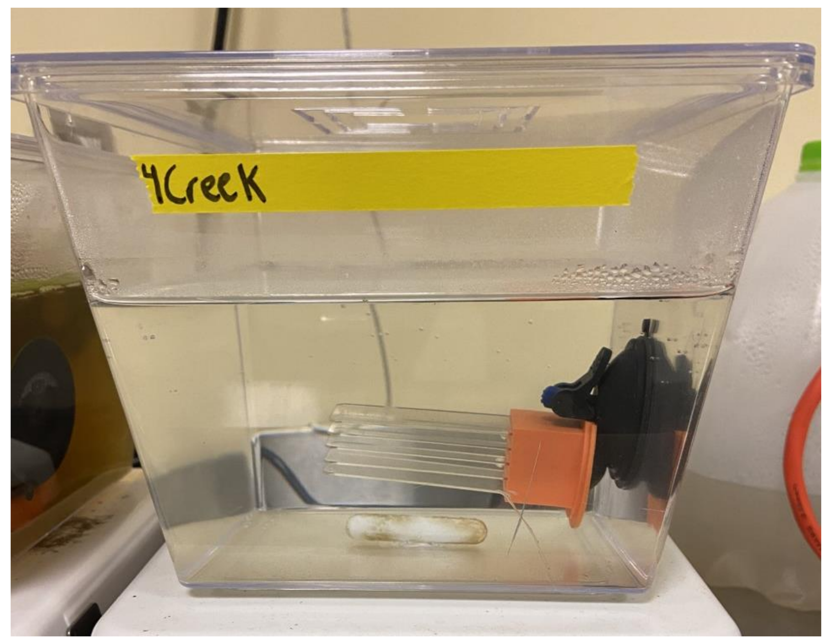

Figure 3: A tank setup that was used in the experiment (Lazar, 2022).

Results / Discussion

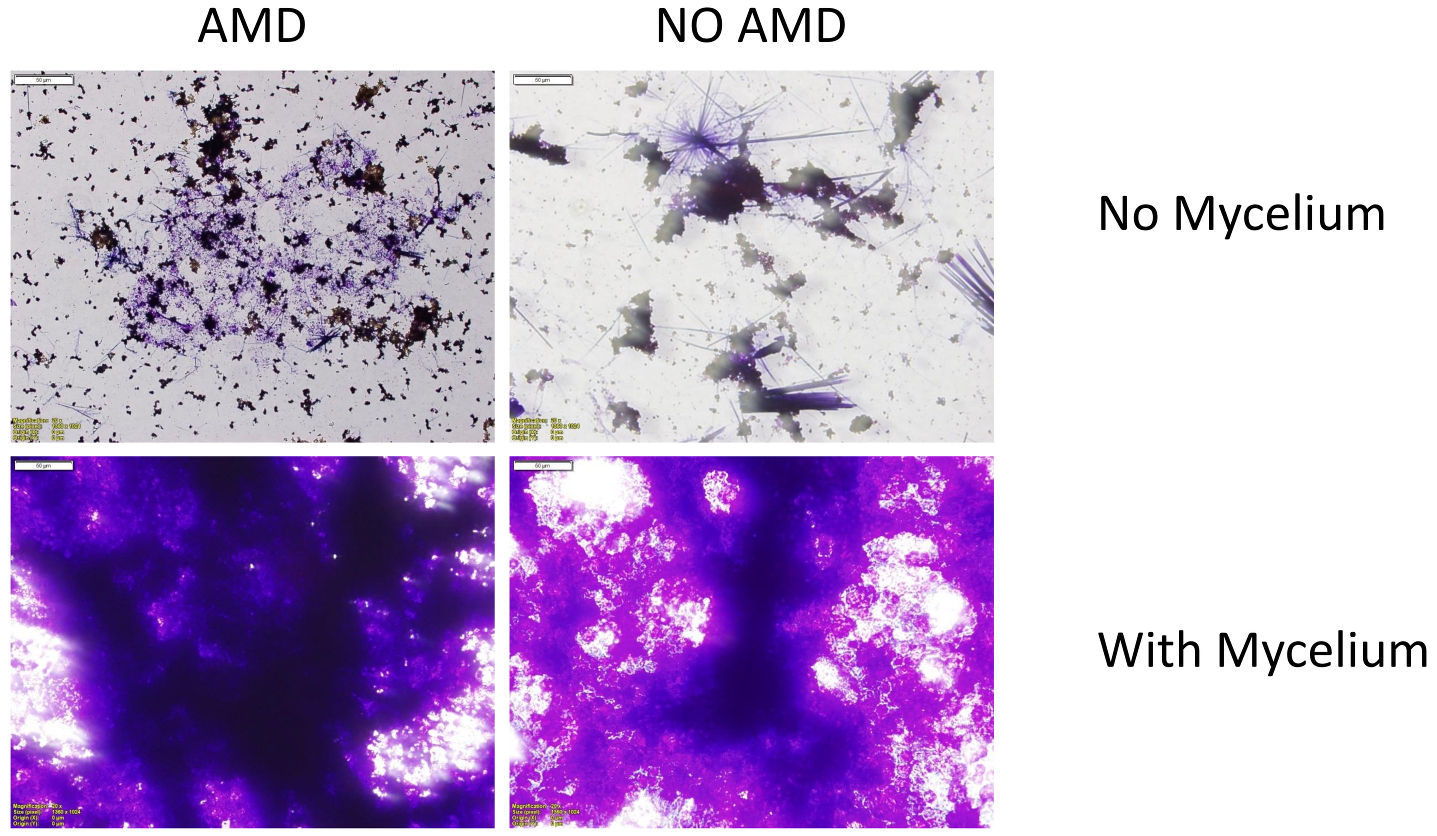

Figure 4: Imaging results of the biofilms after 3 weeks of growth. Clockwise starting with the top left photo are AMD, creek, creek with mycelium, and AMD with mycelium (Lazar, 2022).

- The images show that biofilm formation was much thicker in the two tanks that contained mycelium
- The following absorbance values were recorded from the UV-Vis
  - AMD: A= 0.092
  - AMD with mycelium: A= 2.339
  - Creek with mycelium: A= 2.380
  - Creek: A= 0.326
- Higher the absorbance value mean there was more crystal violet dissolved. Thicker biofilms carry more crystal violet.
- Tanks with mycelium had much higher absorbance values.
- Better biofilm formation may indicate that mycelium removes iron from AMD and improves microbiome health in streams.

References

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