## Biostabilization of gold mine tailings using cyanobacteria

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The dispersion of fine soil particles from the erosive force of wind has become a growing threat across the globe, especially in contaminated areas such as mine tailings impoundments. Transport of metals and metalloids in dust from mining operations can have numerous detrimental effects on human health and ecosystem services (Csavina et al., 2012). Given the adverse effects of wind erosion, establishing effective surface protection methods is critical to reduce dust emissions from mine tailings. Mechanical, chemical, and agricultural methods of wind erosion prevention are commonly adopted; however, the efficiency and success of these strategies have been limited due to their temporary effectiveness, instability, time-consuming application, high cost, and adverse environmental impacts on-site/off-site throughout their life cycle (Karol, 2003; Ye et al., 2002).

Biotechnological techniques based on using cyanobacteria for stabilizing erosion-susceptible soils have been developed in recent years as environmentally responsible and sustainable long-term solutions to wind erosion mitigation (Fattahi et al., 2020; Rossi et al., 2022). The physical structure of soils inoculated by cyanobacteria is influenced through the secretion of EPSs and the filamentous morphology of some genera that entangle loose soil particles and create a cohesive net (Chamizo et al., 2018). Cyanobacterial physiological activities increase soil organic matter content and improve soil water holding capacity (Adessi et al., 2018). These features favor the formation of resilient matrices that resist the erosive impacts of wind. Despite the potentials of cyanobacteria to stabilize soil surface against wind erosion, limited information exists in their application for artificial ecosystems such as tailings deposits, as most studies have been conducted on natural substrates such as desert soils. The purpose of this study is to determine the feasibility of using cyanobacteria for the creation of stable and durable biocrusts on mine tailings.

In this study, the performance of two nitrogen-fixing cyanobacteria (*Anabaena* sp. and *Nostoc muscorum*), individually and as a consortium, were investigated in promoting the biophysicochemical properties of gold mine tailings. Tailings materials were collected from a gold mine site in Northern Ontario. In the single inoculation experiments, the inoculation was conducted by dispersing either *N. muscorum* or *Anabaena* sp. over the entire surface of the tailings samples with a biomass concentration of 4 g dry weight m<sup>-2</sup>. In the co-inoculation experiments, the two cyanobacterial suspensions were well mixed in equal amounts prior to the inoculation. In the non-inoculated sample, distilled water without the inoculum was added at an equivalent volume of the cyanobacterial suspension. Samples were maintained in a growth chamber for 6 weeks. Five experimental conditions were considered with three replicates for each condition: tailings without treatment (BT), tailings treated with distilled water (DW), tailings samples either treated with single inoculation of *N. muscorum* (NI), single inoculation of *Anabaena* sp. (AI), or co-inoculation of the two cyanobacteria species (AI+NI). Chlorophyll-a, EPS fractions, degree of wind erosion, and compressive strength of the formed biocrusts under different treatment conditions were analyzed to assess the fertility properties and dust mitigation efficiencies of the treated tailings.

The inoculated samples (NI, AI, and NI+AI) showed significant reductions in the mass loss and improvements in the compressive strength compared to the non-inoculated samples (BT and DW), as presented in Table 1 ( $P \le 0.01$ ). The stabilization capacity of the inoculated samples from highest to lowest was NI+AI, NI, and AI. Regarding chlorophyll-a and total EPS, the highest values were found under the co-inoculation of the cyanobacteria strains (NI+AI), followed by the inoculation of *Anabaena* sp. (AI), and the inoculation of *N. muscorum* (NI) resulted in the least amounts of chlorophyll-a and total EPS.

Table 1. Compressive strength and degree of wind erosion (%) of the non-inoculated and inoculated tailings samples exposed to 25 m s<sup>-1</sup> wind velocity after a 6-week incubation time. The data represent means and standard deviations of three biological replicates.

Treatment	Degree of wind erosion (%)	Compressive strength (kg cm <sup>-2</sup> )
BT	$100\pm0.00$	0.00
DW	$81.99\pm3.54$	$0.10\pm0.00$
NI	$2.44\pm0.19$	$1.40\pm0.07$
AI	$8.51\pm0.78$	$0.85\pm0.00$
NI+AI	$0.85\pm0.03$	$1.90 \pm 0.00$

The co-inoculation approach resulted in the development of a resistant biocrust against wind erosion with high compressive strength. *Anabaena* sp. promoted higher synthesis of chlorophyll-a and secretion of total EPS compared to *N. muscorum* presumably due to its high resistance to the metal toxicity of the tailings. *N. muscorum* favored the formation of a more stabilized structure against wind erosion compared to *Anabaena* sp. possibly due to its high resistance to the metal toxicity of the tailings. *N. muscorum* favored the formation of a more stabilized structure against wind erosion compared to *Anabaena* sp. possibly due to its higher secretion of the more condensed TB-EPS and filamentous growth. With inoculating the cyanobacteria in a mixture, the beneficial effects obtained with the use of single strains on biocrust formation could be combined and the synergistic effects of the species would lead to a comparatively stronger structure.

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