microPublication BIOLOGY

11/4/2026 - *Open Access*

Passive Treatment of Acid Mine Drainage Improves Growth of Native Tree Saplings

Lauren Palmieri[1*], Alissa Packer[1§*]

[1]Biology, Susquehanna University, Selinsgrove, PA, United States

[§]To whom correspondence should be addressed: packer@susqu.edu

[*]These authors contributed equally.

Abstract

Acid mine drainage (AMD), characterized by low pH and high dissolved metal concentrations, contaminates streams and impairs ecological recovery of impacted watersheds. We evaluated whether water collected from a passive remediation system supports improved plant growth. In a greenhouse experiment, saplings of five native Pennsylvania tree species were irrigated with untreated AMD entering a passive system, partially treated AMD exiting the system, or tap water. Growth was reduced in all species exposed to untreated AMD, whereas growth of saplings receiving partially treated water did not differ from controls. These results indicate that partial treatment of AMD reduces phytotoxicity, improving growth.

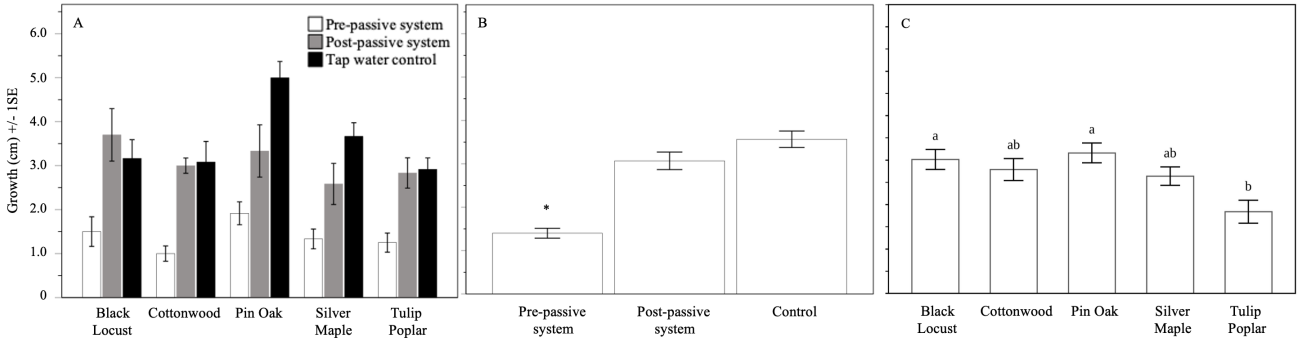


Figure 1. Growth of native saplings exposed to untreated AMD entering the passive system (pre-passive system), partially treated water exiting the final north standpipe in the passive system (post-passive system), or tap water control in a greenhouse experiment:

A. Change in sapling height across species and treatments (two-way ANCOVA). The species × treatment interaction was not significant. B. Watering treatment significantly affected growth ($F_{2,162}$ = 48.46, $p < 0.001$); saplings in the AMD water grew less than those in the partially treated water or the control (Tukey-adjusted $p < 0.001$ indicated by *); growth in the partially treated water did not differ from the control (Tukey-adjusted $p = 0.070$). Bars in A and B are raw means (±1 SE). C. ANCOVA-adjusted mean growth (± 1 SE) by species. Regardless of watering treatment, growth differed among species ($F_{4,162}$ = 5.25, $p < 0.001$); this was driven primarily by the slower growth of tulip poplar (Tukey-adjusted comparisons). Bars sharing a letter do not differ (Tukey-adjusted $p > 0.05$).

Description

Acid mine drainage (AMD), generated primarily by pyrite oxidation, produces acidic waters enriched in dissolved metals (Rose and Cravotta 1998). In streams affected by mine drainage, low pH, metal toxicity, and iron and aluminum hydroxide precipitates degrade aquatic habitat and reduce the abundance and diversity of macroinvertebrates and fish, often eliminating sensitive taxa (Cherry et al. 2001; Hogsden and Harding 2012). These impacts can extend into adjacent riparian habitats where reduced emergence of aquatic insects limits food resources for terrestrial species, such as spiders (Paetzold et al. 2011; Schmidt et al. 2013). More broadly, coal surface mining can alter terrestrial ecosystems by limiting forest establishment and recovery (Zipper et al. 2011).

Passive treatment systems are constructed features, such as aerobic wetlands, limestone drains, and vertical flow wetlands, that improve water quality using gravity flow and natural chemical and biological processes rather than continuous chemical additions (Skousen et al. 2017). The passive system examined here consists of an aerobic wetland followed by three vertical flow wetlands (McGuire et al. 2025). In the aerobic wetland, iron oxidizes and settles out as iron oxyhydroxides. In vertical flow wetlands, water passes through an organic layer (often spent mushroom compost) that

consumes oxygen and supports sulfate-reducing bacteria, and then through underlying limestone, which adds alkalinity, raises pH, and causes aluminum to precipitate (Skousen et al. 2017). Treatment success is typically judged by changes in pH, alkalinity, and dissolved metals (McGuire et al. 2025). Studies that use biological indicators of water quality often focus on benthic macroinvertebrates, measuring changes in taxon richness, abundance, or the return of metal-sensitive taxa (e.g. Kotalik et al. 2023; Dean et al. 2025), while plant responses are less frequently measured (but see Wu et al. 2022). Because reforestation is critical for long-term watershed recovery, tree growth offers a direct test of whether treated water can support vegetation.

Our study site is located in the Shamokin Creek watershed, which lies within the Western Middle Anthracite coalfield of Pennsylvania, where numerous abandoned mine discharges affect water quality (Cravotta and Kirby 2003). The region of the watershed where this passive system is located is primarily forested, with little agricultural land (Cravotta and Kirby 2003). At the study site, a portion of the AMD discharge is diverted through the treatment system before entering the creek, allowing us to collect both untreated influent and partially treated water exiting the system.

We compared growth of five native Pennsylvania tree species irrigated with untreated AMD, partially treated water from the system, or tap water under greenhouse conditions. Species were selected to represent common native hardwoods with varying tolerance to AMD-associated conditions (see Ashby and Kolar 1998; Groninger et al. 2006; Evans et al. 2013; Nkongolo et al. 2017; Zhang et al. 2022), with black locust commonly used in reclamation efforts (Ashby et al. 1985). We hypothesized that untreated AMD water would reduce sapling growth relative to partially treated and control water.

Sapling growth responded strongly to watering treatment, with no evidence of a species × treatment interaction (Fig. 1A; $F_{8,162} = 1.02$, $p = 0.422$), indicating a consistent response across species. Growth differed significantly among watering treatments (Fig. 1B; $F_{2,162} = 48.46$, $p < 0.001$), with saplings irrigated with untreated AMD (pre-passive system) exhibiting reduced growth relative to those receiving partially treated water (post-passive system) or tap water (Tukey-adjusted $p < 0.001$). Growth in the post-passive treatment did not differ from the control (Tukey-adjusted $p = 0.070$), indicating that partial treatment largely removed the growth reduction caused by untreated AMD. Across species, growth also varied (Fig. 1C; $F_{4,162} = 5.25$, $p < 0.001$), driven mainly by reduced growth in tulip poplar relative to black locust (Tukey-adjusted $p = 0.017$) and pin oak (Tukey-adjusted $p < 0.001$). Initial height was a significant covariate ($F_{1,162} = 9.61$, $p = 0.002$): taller saplings grew slightly less, and this relationship did not differ among species (species × initial height: $F_{4,156} = 0.75$, $p = 0.561$).

Water chemistry data collected by McGuire et al. (2025) at the same site in November 2023 compare influent (pre-passive treatment) to water exiting the final north standpipe (post-passive treatment), providing context for growth responses in our study. Water collected from the final standpipe had a higher pH (4.1 to 5.8) and substantially lower dissolved aluminum (Al; 2.54 to 0.11 mg/L), a metal whose solubility and phytotoxicity are enhanced under acidic conditions (Ur Rahman et al. 2024). Aluminum is known to inhibit root elongation, shoot growth, and nutrient uptake (Kochian 1995; Ur Rahman et al. 2024); its reduction in post-passive treatment water may have alleviated a key limitation on plant growth. Dissolved iron (Fe) concentrations were also reduced following treatment (14.72 to 3.20 mg/L), which may have further contributed to improved growth by decreasing metal stress and limiting iron precipitation on root surfaces. Concentrations of nickel (Ni) and cobalt (Co) were only partially reduced (83.0 to 46.9 µg/L and 55.1 to 33.1 µg/L, respectively), zinc (Zn) declined only modestly (106 to 90 µg/L), and manganese (Mn) increased slightly (2.53 to 2.84 mg/L). The improvement in growth following passive treatment was consistent with the reduced acidity and dissolved aluminum concentrations reported at this site, although water chemistry was not measured during the greenhouse experiment.

Together, these results indicate that partial treatment primarily improves plant performance by mitigating some of the chemical stressors associated with AMD, particularly low pH and elevated aluminum. Remaining metals did not detectably limit plant growth during the timeframe of this study. This finding suggests that passive treatment can reduce the adverse effects of AMD on the growth of native tree saplings, although responses may vary with water chemistry, plant species, and exposure duration.

Methods

Water used in the greenhouse experiment was collected in Coal Township, PA, from Scarlift site 15 (Corbin Water Level Drift; Latitude 40.780712, Longitude -76.515436). Site 15 discharges AMD directly into Shamokin Creek. A flow splitter diverts a portion of this untreated AMD through the passive treatment system, which was constructed by the Shamokin Creek Restoration Alliance (SCRA) and the Northumberland County Conservation District in 2006 (Shamokin Creek Restoration Alliance, n.d.). The system consists of an aerobic wetland followed by three vertical flow wetlands (VFW). Beneath each VFW is a mushroom compost layer (~ 2 ft) on top of a limestone base (~ 2-2.5 ft). Standpipes in the limestone layer of VFWs discharge water from pond to pond, and spillways connecting the ponds allow for overflow (see McGuire et al. 2025 for schematic).

For this experiment, untreated water was collected from the pipe delivering AMD from the flow splitter into the passive treatment facility (“pre-passive system” treatment). Partially treated water was collected from the north standpipe that

delivered water from the mushroom compost/limestone layer of the final VFW of the passive treatment system to the final collection point where it combined with spillover from the final pond to form the effluent that then returned to the creek ("post-passive system" treatment). Water was collected in 5 gallon buckets each week for use in the greenhouse experiment.

Native tree species tested included black locust (*Robinia pseudoacacia*), Eastern cottonwood (*Populus deltoides*), pin oak (*Quercus palustris*), silver maple (*Acer saccharinum*), and tulip poplar (*Liriodendron tulipifera*)—all fast-growing hardwoods native to Pennsylvania (Burns and Honkala 1990). Saplings (approximately 2-3 years old) were obtained from Ecotone's Native Plant Nursery (Ecotone Ecological Restoration, Inc., Forest Hill, MD, USA) in October 2022. They were grown in a mixture of two parts pine bark to one part sand (Hollins Organic Products, Inc.) in tree pots (9"tall, 3"x3" square at top of pot with slight taper towards bottom). Pots were randomized in the greenhouse. Initial sapling height was measured on 7 November 2022. For approximately two months they were allowed to grow and acclimate in the greenhouse at the Center for Environmental Education and Research (CEER) at Susquehanna University. Tap water was provided as needed during this period.

On 23 January 2023, all saplings were measured and randomly assigned to pre-passive system, post-passive system, or tap water control treatments. Plants were watered weekly with 100 mL of water according to their treatment group. Each species × treatment combination included 12 saplings, except black locust in the post-passive treatment (n = 10), because only 34 black locust saplings were available at the start of treatments. Final measurements of height were recorded on 6 March 2023, giving a 42-day treatment period.

Sapling growth, characterized as change in height between 23 January and 6 March 2023, was analyzed using a two-way ANCOVA in jamovi (version 2.6.45.0; The jamovi project 2025), with species, treatment (pre-passive system, post-passive system, control), and their interaction as fixed effects and height at the start of treatments (23 January) as a covariate. Homogeneity of regression slopes was confirmed by testing species × initial height and treatment × initial height interactions, which were not significant ($p > 0.05$) and were removed from the final model. Pairwise comparisons of estimated marginal means were Tukey-adjusted.

Acknowledgements: The authors are grateful to Steve Motyka for sharing his extensive knowledge of the restoration efforts at Scarlift site 15, and to Carl Kirby for his insights into the geochemical data presented in this work.

References

Ashby WC, Vogel WG, Rogers NF. 1985. Black locust in the reclamation equation. Gen. Tech. Rep. NE-105. Broomall, PA: U. S. Department of Agriculture, Forest Service, Northeastern Forest Experiment Station. DOI: 10.2737/NE-GTR-105

Ashby WC, Kolar C. 1998. Thirteen-year hardwood tree performance on a Midwest surface mine. In American Society for Surface Mining and Reclamation Annual National Meeting (pp. 124–133). American Society of Mining and Reclamation.

Burns RM, Honkala BH [Technical coordinators]. 1990. Silvics of North America: Volume 2. Hardwoods. United States Department of Agriculture (USDA), Forest Service, Agriculture Handbook 654.

Cherry DS, Currie RJ, Soucek DJ, Latimer HA, Trent GC. 2001. An integrative assessment of a watershed impacted by abandoned mined land discharges. Environ Pollut 111(3): 377-88. PubMed ID: 11202742

Cravotta CA III, Kirby CS. 2003. Effects of abandoned coal-mine drainage on streamflow and water quality in the Shamokin Creek Basin, Northumberland and Columbia Counties, Pennsylvania, 1999-2001 (Water-Resources Investigations Report 03-4311). U.S. Geological Survey. DOI: 10.3133/wri034311

Dean AP, Nelson J, Jones AP, Sykes A, Child F, Sweeney CJ, et al., Pittman JK. 2025. Habitat recovery from diverted acid mine drainage pollution determined by increased biodiversity of river and estuarine benthic species. Sci Total Environ 966: 178726. PubMed ID: 39923471

Evans DM, Zipper CE, Burger JA, Strahm BD, Villamagna AM. 2013. Reforestation practice for enhancement of ecosystem services on a compacted surface mine: Path toward ecosystem recovery. Ecological Engineering 51: 16-23. DOI: 10.1016/j.ecoleng.2012.12.065

Groninger JW, Fillmore SD, Rathfon RA. 2006. Stand Characteristics and Productivity Potential of Indiana Surface Mines Reclaimed Under SMCRA. Northern Journal of Applied Forestry 23: 94-99. DOI: 10.1093/njaf/23.2.94

Hogsden KL, Harding JS. 2012. Consequences of acid mine drainage for the structure and function of benthic stream communities: a review. Freshwater Science 31: 108-120. DOI: 10.1899/11-091.1

The jamovi project. 2025. jamovi (Version 2.6.45.0) [Computer Software]. Retrieved from https://www.jamovi.org

Kochian LV. 1995. Cellular Mechanisms of Aluminum Toxicity and Resistance in Plants. Annual Review of Plant Physiology and Plant Molecular Biology 46: 237-260. DOI: 10.1146/annurev.pp.46.060195.001321

Kotalik CJ, Meyer JS, Cadmus P, Ranville JF, Clements WH. 2023. Integrated Assessment of Chemical and Biological Recovery After Diversion and Treatment of Acid Mine Drainage in a Rocky Mountain Stream. Environmental Toxicology and Chemistry 42: 512-524. DOI: 10.1002/etc.5515

McGuire MM, Schultheis HM, Herman EK. 2025. Fate and transport of dissolved/colloidal metals in an aging passive treatment system for coal mine drainage: Divergent behavior of trace metals and iron. Journal of Contaminant Hydrology 273: 104609. DOI: 10.1016/j.jconhyd.2025.104609

Nkongolo KK, Narendrula-Kotha R, Kalubi K, Rainville S, Michael P. 2017. High level of nickel tolerance and metal exclusion identified in silver maple (*Acer saccharinum*). Chemistry and Ecology 33: 795-806. DOI: 10.1080/02757540.2017.1376664

Paetzold A, Smith M, Warren PH, Maltby L. 2011. Environmental impact propagated by cross-system subsidy: Chronic stream pollution controls riparian spider populations. Ecology 92: 1711-1716. DOI: 10.1890/10-2184.1

Rose AW, Cravotta CA III. 1998. Geochemistry of coal mine drainage. In K. B. C. Brady, M. W. Smith, & J. Schueck (Eds.), Coal mine drainage prediction and prevention in Pennsylvania (pp. 1.1–1.22). Pennsylvania Department of Environmental Protection.

Schmidt TS, Kraus JM, Walters DM, Wanty RB. 2013. Emergence flux declines disproportionately to larval density along a stream metals gradient. Environ Sci Technol 47(15): 8784-92. PubMed ID: 23781899

Shamokin Creek Restoration Alliance. (n.d.). Completed Projects. https://www.shamokincreek.org/

Skousen J, Zipper CE, Rose A, Ziemkiewicz PF, Nairn R, McDonald LM, Kleinmann RL. 2017. Review of Passive Systems for Acid Mine Drainage Treatment. Mine Water and the Environment 36: 133-153. DOI: 10.1007/s10230-016-0417-1

Ur Rahman S, Han JC, Ahmad M, Ashraf MN, Khaliq MA, Yousaf M, et al., Du. 2024. Aluminum phytotoxicity in acidic environments: A comprehensive review of plant tolerance and adaptation strategies. Ecotoxicology and Environmental Safety 269: 115791. DOI: 10.1016/j.ecoenv.2023.115791

Wu A, Zhang Y, Zhao X, Shi H, Xu S, Li J, Zhang G, Guo L. 2022. Experimental Research on the Remediation Ability of Four Wetland Plants on Acid Mine Drainage. Sustainability 14: 3655. DOI: 10.3390/su14063655

Zhang X, Zeng B, Li H, Huang J, Jiang L, Zhang X, et al., Huang. 2022. Soil heavy metals and phytoremediation by *Populus deltoides* alter the structure and function of bacterial community in mine ecosystems. Applied Soil Ecology 172: 104359. DOI: 10.1016/j.apsoil.2021.104359

Zipper CE, Burger JA, Skousen JG, Angel PN, Barton CD, Davis V, Franklin JA. 2011. Restoring Forests and Associated Ecosystem Services on Appalachian Coal Surface Mines. Environmental Management 47: 751-765. DOI: 10.1007/s00267-011-9670-z

Funding: Funding for this project was provided by the Department of Biology at Susquehanna University.

Conflicts of Interest: The authors declare that there are no conflicts of interest present.

Author Contributions: Lauren Palmieri: conceptualization, data curation, formal analysis, investigation, methodology, writing - original draft, writing - review editing. Alissa Packer: conceptualization, methodology, writing - review editing, formal analysis, supervision, project administration, visualization, software.

Reviewed By: Anonymous

History: Received March 20, 2026 **Revision Received** October 11, 2026 **Accepted** November 2, 2026 **Published Online** November 4, 2026 **Indexed** November 18, 2026

Copyright: © 2026 by the authors. This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International (CC BY 4.0) License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Citation: Palmieri L, Packer A. 2026. Passive Treatment of Acid Mine Drainage Improves Growth of Native Tree Saplings. microPublication Biology. 10.17912/micropub.biology.002108