



# Effect of *Azotobacter* Inoculation and Cow Manure Amendment on Lead (Pb) Phytoremediation in Gold Mine Tailings by Vetiver Grass (*Chrysopogon zizanioides* L.)

Salma Nur Fauziyyah<sup>1\*</sup>, Ikrima Hasya Maharani<sup>1</sup>, Raden Rhazista Noviardi<sup>2,4</sup>, Sri Handayani<sup>3</sup>, Mieke Rochimi Setiawati<sup>1</sup>, Pujawati Suryatmana<sup>1</sup>

<sup>1</sup>Department of Soil Science and Land Resources, Faculty of Agriculture, Padjadjaran University, Jatinangor, Indonesia

<sup>2</sup>Research Center for Environment and Clean Technologies, National Research and Innovation Agency (BRIN), Bandung, Indonesia

<sup>3</sup>Research Center for Mineral Technology, National Research and Innovation Agency, Bandung, Indonesia

<sup>4</sup>Research Collaboration Center for Sustainability of Environmental Carrying Capacity, Bandung, Indonesia

\*Corresponding author: [salmafauzh@gmail.com](mailto:salmafauzh@gmail.com)

Received: 23 Jun 2026; Received in revised form: 19 Jul 2026; Accepted: 26 Jul 2026; Available online: 02 Aug 2026

©2026 The Author(s). Published by Infogain Publication. This is an open-access article under the CC BY license

(<https://creativecommons.org/licenses/by/4.0/>).

**Abstract**— Gold mine tailings generally have low fertility and contain lead (Pb), a heavy metal that may inhibit plant growth. Phytoremediation using vetiver grass (*Chrysopogon zizanioides* L.) offers a simple and effective strategy for improving tailings media and reducing Pb concentrations. This study aimed to evaluate the effect of the combination of *Azotobacter* inoculation and ameliorant application on Pb reduction and to determine the best combination for enhancing the phytoremediation capacity of vetiver grass in gold mine tailings. The experiment was arranged in a Randomized Block Design with two factors and three replications. The first factor was the type of *Azotobacter* inoculation (without *Azotobacter*, *Azotobacter vinelandii*, indigenous *Azotobacter* tailing-1, indigenous *Azotobacter* tailing-2, and an *Azotobacter* consortium). The second factor was the dose of cattle manure (0 t ha<sup>-1</sup>, 10 t ha<sup>-1</sup>, and 20 t ha<sup>-1</sup>). The results showed a significant interaction between the *Azotobacter* type and cattle manure rate on the efficiency of Pb reduction in tailings. The combination of *Azotobacter vinelandii* with 10 t ha<sup>-1</sup> cattle manure and the *Azotobacter* consortium with 20 t ha<sup>-1</sup> cattle manure were the most effective treatments, achieving up to 61,55% reduction in tailing Pb concentration.

**Keywords**— indigenous *Azotobacter*, *Azotobacter vinelandii*, cattle manure, reduction efficiency



## I. INTRODUCTION

Heavy metal contamination is a major environmental issue in Indonesia, particularly in areas affected by gold mining activities. The processing leaves waste or tailings carry a high risk of health and environmental problems. Tailings from gold mines contribute to the accumulation of heavy metals, acid rock drainage, dust emissions, and surface runoff, with significant effects on agricultural lands, human health, and ecosystems [1]. Tailing leftovers from gold ore processing in Kertajaya Village are generally thrown into rivers or gardens, collected in holding ponds, or stored in sacks [2].

Lead (Pb) is one of the most persistent contaminants because of its strong affinity for soil particles, which limits its removal through natural leaching processes [3]. Gold mine tailings from Kertajaya Village, Sukabumi, West Java, contained 758.2 mg kg<sup>-1</sup> Pb, exceeding the high-risk threshold of 220 mg kg<sup>-1</sup> established by the World Health Organization [4]. In addition to heavy metal contamination, the tailings exhibited highly unfavorable characteristics for plant growth, including acidic conditions (pH 3–4), low organic matter content, low nutrient availability, and limited microbial activity [5]. Elevated Pb concentrations can inhibit root growth, chlorophyll formation, photosynthesis, and biomass

production while suppressing soil microbial activity and nutrient cycling, ultimately reducing soil productivity and ecosystem function [6,7].

Phytoremediation has been widely recognized as an environmentally friendly and cost-effective approach for remediating heavy metal-contaminated soils [8]. Vetiver grass (*Chrysopogon zizanioides* L.) is considered a promising phytoremediation species because of its high tolerance to degraded soils and ability to accumulate heavy metals [9] reported that vetiver accumulated Pb in shoots up to 10.16 mg kg<sup>-1</sup>, whereas [10] found that approximately 65–75% of absorbed metals were retained in the root system, indicating its strong phytostabilization potential. However, the effectiveness of phytoremediation is closely linked to plant growth, which are often limited by the poor fertility conditions of mine tailings.

The application of organic amendments and beneficial microorganisms has been proposed to improve tailings quality and enhance phytoremediation efficiency. Cattle manure compost can improve soil physical, chemical, and biological properties while supplying organic matter and nutrients. Humic and fulvic substances in organic amendments can also reduce Pb toxicity through organometallic complex formation [11]. Moreover, rhizosphere microorganisms influence metal availability by modifying soil pH, redox conditions, and chelation processes [12]. Among these microorganisms, *Azotobacter* are particularly important because they fix atmospheric nitrogen and produce organic acids, siderophores, and extracellular polymeric substances (EPS) that contribute to metal transformation and immobilization in the rhizosphere [13,14]

Previous studies have demonstrated the potential of combining *Azotobacter* inoculation and cattle manure compost to improve soil fertility and support heavy metal remediation. [15] reported that this combination increased soil available nitrogen to 246 mg kg<sup>-1</sup> and improved soil pH under saline conditions. Similarly, [16] observed significant interactions between cattle manure compost and *Azotobacter chroococcum* or *Azotobacter* consortia in reducing Hg concentrations and increasing Hg uptake by plants. However, the effectiveness of these interactions depends on the characteristics of the inoculated strains and the environmental conditions of contaminated tailings. Indigenous *Azotobacter* isolated from mercury-contaminated soils tolerated HgCl<sub>2</sub> concentrations up to 15 mg kg<sup>-1</sup>, although bacterial growth declined at 20 mg kg<sup>-1</sup> [17]. In addition, inappropriate compost application rates may increase metal mobility under acidic conditions and reduce remediation efficiency [18]. Therefore, determining the optimal combination of *Azotobacter* inoculation and

cattle manure compost application is essential to improve vetiver growth and maximize Pb phytoremediation in gold mine tailings.

## II. MATERIALS AND METHODS

### 2.1 Materials

Gold mine tailings were collected from an artisanal gold mining area in Kertajaya Village, Sukabumi Regency, West Java, Indonesia. The tailings have a pH of 3.52, organic carbon content of 0.55%, total nitrogen of 0.017%, total phosphorus of 31.88 mg/100 g, potassium of 6.61 mg/100 g, low cation exchange capacity (CEC), and very low base saturation (BS), with a silty clay loam texture. The cattle manure compost was obtained from the Waste Processing Unit of the Faculty of Animal Husbandry, Universitas Padjadjaran. The cattle manure had the following characteristics: 25.47% organic carbon, 59% moisture content, pH 8.3, and 1.41% total nitrogen.

*Azotobacter vinelandii*, indigenous *Azotobacter* tailing-1, indigenous *Azotobacter* tailing-2, and their consortium were propagated in Vermani liquid medium, resulting in a final inoculum density of approximately 10<sup>8</sup> CFU mL<sup>-1</sup>.

### 2.2 Pot Experiment

The experiment was conducted from December 2025 to April 2026. Vetiver (*Chrysopogon zizanioides* L.) cultivation was carried out at the Ciparanje Experimental Farm, Universitas Padjadjaran, located at approximately 752 m above sea level.

The experiment was arranged in a factorial randomized complete block design consisting of two factors with three replications, resulting in 45 experimental units. The first factor was *Azotobacter* inoculation, comprising five levels: A<sub>0</sub> (without *Azotobacter*), A<sub>1</sub> (*Azotobacter vinelandii*), A<sub>2</sub> (indigenous *Azotobacter* tailing-1), A<sub>3</sub> (indigenous *Azotobacter* tailing-2), and A<sub>4</sub> (consortium of the three isolates). The second factor was cattle manure application rate, consisting of B<sub>0</sub> (0 t ha<sup>-1</sup>), B<sub>1</sub> (10 t ha<sup>-1</sup>), and B<sub>2</sub> (20 t ha<sup>-1</sup>).

The growth medium consisted of a 1:1 mixture of gold mine tailings and topsoil, amended with cattle manure according to the treatment rates. Each treatment was inoculated with 20 mL of a bacterial suspension at a density of 10<sup>9</sup> CFU mL<sup>-1</sup> before being transferred into 10-kg polybags. Three vetiver tillers were planted per polybag and maintained for 13 weeks, during which plant growth, *Azotobacter* population density, Pb concentration, and Pb reduction efficiency were evaluated. NPK fertilizer (16–16–16) was applied at 4 and 8 WAP at a rate equivalent to 300 kg ha<sup>-1</sup> (2.25 g polybag<sup>-1</sup>).

## 2.3 Data Collection and Analytical Procedures

*Azotobacter* population density was determined at 0 and 13 weeks after planting (WAP) using the Total Plate Count (TPC) method on Ashby's medium. At harvest (13 WAP), lead (Pb) concentrations in the tailings and planting media were determined by wet digestion followed by Atomic Absorption Spectrophotometry (AAS) [19].

Data on *Azotobacter* population density, plant height, and Pb concentration were analyzed using IBM SPSS Statistics 27.0. Analysis of variance (ANOVA) was performed to determine the effects of the treatments, and when significant differences were detected, means were compared using Duncan's Multiple Range Test (DMRT) at the 5% significance level.

## III. RESULTS AND DISCUSSION

### 3.1 *Azotobacter* Population Density

Table I. Main Effects of Treatments on *Azotobacter* Population Density

| Treatment                               | <i>Azotobacter</i> Population Density                     |
|-----------------------------------------|-----------------------------------------------------------|
|                                         | 0 WAP <sup>1</sup> (10 <sup>5</sup> CFU g <sup>-1</sup> ) |
| <i>Azotobacter</i> Type                 |                                                           |
| Without <i>Azotobacter</i>              | 5.98 a                                                    |
| <i>Azotobacter vinelandii</i>           | 6.34 b                                                    |
| Indigenous <i>Azotobacter</i> tailing-1 | 6.37 b                                                    |
| Indigenous <i>Azotobacter</i> tailing-2 | 6.66 d                                                    |
| Consortium                              | 6.48 c                                                    |
| Cattle Manure Rate                      |                                                           |
| 0 t ha <sup>-1</sup>                    | 6.30 a                                                    |
| 10 t ha <sup>-1</sup>                   | 6.34 a                                                    |
| 20 t ha <sup>-1</sup>                   | 6.36 b                                                    |

Means followed by the same letter within a column are not significantly different according to Duncan's Multiple Range Test (DMRT) at the 5% significance level. <sup>1</sup>WAP, week after planting.

Analysis of variance indicated that *Azotobacter* population density at 0 WAP was significantly affected by the main effects of *Azotobacter* inoculation and cattle manure compost application, whereas their interaction was not significant (Table I). The absence of a significant interaction indicates that each *Azotobacter* type was able to grow and persist across all cattle manure application

rates, exhibiting distinct growth characteristics without a specific dependence on a particular level of organic matter input.

The consortium treatment resulted in a significantly higher population than the uninoculated control and *Azotobacter vinelandii*, indicating a synergistic effect among microbial members. However, its population remained lower than that of the indigenous *Azotobacter* tailing-2 isolate, suggesting that indigenous microorganisms possess greater ecological fitness and adaptation to the tailing environment than introduced strains [20]. Cattle manure compost application at 20 t ha<sup>-1</sup> significantly increased *Azotobacter* population density compared with 10 t ha<sup>-1</sup> and the control treatment. This finding indicates that the higher compost rate supplied sufficient organic carbon to support bacterial growth and metabolism. Organic carbon serves as an energy source for respiration, generating ATP and reducing equivalents required for nitrogenase activity during biological nitrogen fixation [21].

Table II. Interaction Effects of Treatments on *Azotobacter* Population Density

| <i>Azotobacter</i> Type                 | Cattle Manure Rate                                          |                       |                       |
|-----------------------------------------|-------------------------------------------------------------|-----------------------|-----------------------|
|                                         | 0 t ha <sup>-1</sup>                                        | 10 t ha <sup>-1</sup> | 20 t ha <sup>-1</sup> |
|                                         | 13 WAP <sup>1</sup> (10 <sup>13</sup> CFU g <sup>-1</sup> ) |                       |                       |
| Without <i>Azotobacter</i>              | 15.05 a                                                     | 15.10 a               | 15.19 a               |
| <i>Azotobacter</i>                      | A                                                           | A                     | A                     |
| <i>Azotobacter vinelandii</i>           | 15.06 a                                                     | 15.18 b               | 15.31 bc              |
|                                         | A                                                           | AB                    | C                     |
| Indigenous <i>Azotobacter</i> tailing-1 | 15.12 a                                                     | 15.18 b               | 15.21 ab              |
|                                         | A                                                           | B                     | B                     |
| Indigenous <i>Azotobacter</i> tailing-2 | 15.19 a                                                     | 15.23 b               | 15.37 c               |
|                                         | A                                                           | A                     | B                     |
| Consortium                              | 15.19 a                                                     | 15.30 c               | 15.41 c               |
|                                         | A                                                           | AB                    | C                     |

Means followed by the same letter are not significantly different according to Duncan's Multiple Range Test (DMRT) at the 5% significance level. Capital letters indicate comparisons among cattle manure compost rates within the same *Azotobacter* species (row comparison), whereas lowercase letters indicate comparisons among *Azotobacter* species within the same cattle manure compost rate (column comparison).

*Azotobacter* populations increased from 0 (Table I) to 13 WAP (Table II) with population densities increasing by

more than 100% across all inoculated treatments. The increase was likely driven by the continuous supply of root exudates, including sugars, organic acids, and amino acids released by vetiver roots, which serve as carbon substrates for microbial growth [22]. In addition, the increase in soil pH during the experiment created a more favorable environment for *Azotobacter* proliferation, reducing acidity stress and enhancing nutrient availability [23].

A significant interaction between *Azotobacter* inoculation and cattle manure compost application was observed at 13 WAP (Table II). The highest *Azotobacter* population density was obtained in the consortium treatment combined with 20 t ha<sup>-1</sup> cattle manure compost, which was significantly higher than the uninoculated control and indigenous *Azotobacter* tailing-1 at the same compost rate, but not significantly different from *Azotobacter vinelandii* and indigenous *Azotobacter* tailing-2. This result indicates that the consortium and 20 t ha<sup>-1</sup> compost provided the most favorable conditions for sustaining *Azotobacter* populations in the tailings medium. The high population density was likely associated with the adaptation of

indigenous isolates to heavy metal-contaminated tailings and the ability of *Azotobacter vinelandii* to produce extracellular polymeric substances (EPS), particularly alginate, which enhance bacterial protection and colonization under environmental stress [24]. Furthermore, organic matter application improved nutrient availability, water retention, and rhizosphere conditions, thereby supporting microbial proliferation [21]. Although population densities in the uninoculated control remained significantly lower, they increased throughout the experiment, indicating the presence of native *Azotobacter* populations and the contribution of vetiver root exudates as carbon sources for microbial growth [25].

### 3.2 Vetiver Plant Height

Based on Duncan's Multiple Range Test (DMRT) at the 5% significance level (Table III) there was no significant interaction between *Azotobacter* type and cattle manure application rate on vetiver plant height. The absence of interaction may be attributed to the high Pb concentration and poor tailing quality, which limited plant physiological processes and nutrient uptake [26,27].

Table III. Main Effect of Treatments on Vetiver Plant Height

| Treatment                               | Plant Height (cm)         |         |       |       |       |          |
|-----------------------------------------|---------------------------|---------|-------|-------|-------|----------|
|                                         | Week After Planting (WAP) |         |       |       |       |          |
|                                         | 2                         | 4       | 6     | 8     | 10    | 13       |
| <i>Azotobacter</i> Type                 |                           |         |       |       |       |          |
| Without <i>Azotobacter</i>              | 27.20 a                   | 39.62 a | 65.18 | 76.06 | 80.20 | 84.98 a  |
| <i>Azotobacter vinelandii</i>           | 35.53 b                   | 55.72 b | 70.40 | 78.50 | 83.24 | 94.28 b  |
| Indigenous <i>Azotobacter</i> tailing-1 | 36.68 b                   | 51.38 b | 67.25 | 77.53 | 83.18 | 93.55 ab |
| Indigenous <i>Azotobacter</i> tailing-2 | 39.00 b                   | 56.11 b | 70.21 | 74.13 | 80.60 | 90.98 ab |
| Consortium                              | 43.84 c                   | 65.03 c | 75.87 | 85.13 | 94.02 | 106.86 c |
| Cattle Manure Rate                      |                           |         |       |       |       |          |
| 0 t ha <sup>-1</sup>                    | 35.50                     | 49.02 a | 65.72 | 73.54 | 80.71 | 88.00 a  |
| 10 t ha <sup>-1</sup>                   | 35.34                     | 54.50 b | 71.16 | 80.52 | 86.56 | 93.40 a  |
| 20 t ha <sup>-1</sup>                   | 38.51                     | 57.19 b | 72.47 | 80.75 | 85.47 | 101.01 b |

Means followed by the same letter within a column are not significantly different according to Duncan's Multiple Range Test (DMRT) at the 5% significance level. <sup>1</sup>WAP. week after planting.

*Azotobacter* inoculation significantly enhanced plant growth compared with the uninoculated control. The consortium treatment produced the greatest plant height at 2 and 4 WAP and was significantly higher than several single-inoculation treatments and the control. At 13 WAP, the consortium again resulted in the highest plant height and differed significantly from all other treatments. This

response was likely associated with enhanced biological nitrogen fixation and the production of plant growth-promoting substances, including indole acetic acid (IAA), cytokinins, and gibberellins, which stimulate cell division and elongation [28]. In addition, higher chlorophyll content in inoculated plants may have improved photosynthetic efficiency [29].

The application of cattle manure compost at 20 t ha<sup>-1</sup> consistently produced the highest plant height and significantly increased growth at 4 and 13 WAP compared with lower rates. The limited response during the early growth stage suggests that vetiver roots were still developing and that the benefits of organic amendments had not yet been fully realized [30]. As decomposition and mineralization progressed, the higher compost rate improved nutrient availability and tailing properties, including organic matter content, cation exchange capacity, water retention, and the availability of N, P, and K thereby supporting greater nutrient uptake and plant growth during later developmental stages [31,32].

### 3.3 Lead Concentration and Reduction Efficiency

DMRT analysis at the 5% significance level revealed a significant interaction between *Azotobacter* inoculation and cattle manure compost application on the reduction of Pb concentration in the tailing medium (Tables IV and V).

Table IV. Interaction Effects of Treatments on Lead (Pb) Concentration in the Tailing Medium

| Azotobacter Type              | Cattle Manure Rate   |                       |                       |
|-------------------------------|----------------------|-----------------------|-----------------------|
|                               | 0 t ha <sup>-1</sup> | 10 t ha <sup>-1</sup> | 20 t ha <sup>-1</sup> |
|                               | mg kg <sup>-1</sup>  |                       |                       |
| Without                       | 509.71 bc            | 495.57 c              | 455.51 b              |
| <i>Azotobacter</i>            | A                    | A                     | A                     |
| <i>Azotobacter vinelandii</i> | 418.13 a             | 244.33 a              | 388.56 b              |
|                               | C                    | A                     | B                     |
| Indigenous                    | 541.39 c             | 446.83 bc             | 419.51 b              |
| <i>Azotobacter</i> tailing-1  | B                    | A                     | A                     |
| Indigenous                    | 493.68 abc           | 440.11 bc             | 393.13 b              |
| <i>Azotobacter</i> tailing-2  | A                    | A                     | A                     |
|                               | 452.06 ab            | 374.06 b              | 300.30 a              |
| Consortium                    | B                    | AB                    | A                     |

Means followed by the same letter are not significantly different according to Duncan's Multiple Range Test (DMRT) at the 5% significance level. Capital letters indicate comparisons among cattle manure compost rates within the same *Azotobacter* species (row comparison), whereas lowercase letters indicate comparisons among *Azotobacter* species within the same cattle manure compost rate (column comparison).

The initial Pb concentration in the tailings was 635.45 mg kg<sup>-1</sup> and all treatments reduced Pb levels to varying degrees. Even without compost application, several

*Azotobacter* treatments, particularly *Azotobacter vinelandii*, reduced Pb concentrations, although with lower efficiency than treatments receiving organic amendments. Under nutrient-poor conditions, Pb remains more mobile and available in the soil solution, while *Azotobacter* can produce extracellular polymeric substances (EPS), siderophores, and organic acids that influence Pb mobility and availability in the rhizosphere [33,34]. The highest Pb reduction efficiency (61.55%) was achieved by *Azotobacter vinelandii* combined with 10 t ha<sup>-1</sup> cattle manure compost, which was significantly higher than the 20 t ha<sup>-1</sup> treatment for the same inoculant. This result suggests that 10 t ha<sup>-1</sup> provided an optimal level of organic matter to support microbial activity and Pb phytoremediation [35].

Table V. Interaction Effects of Treatments on Lead (Pb) Reduction Efficiency

| Azotobacter Type              | Cattle Manure Rate   |                       |                       |
|-------------------------------|----------------------|-----------------------|-----------------------|
|                               | 0 t ha <sup>-1</sup> | 10 t ha <sup>-1</sup> | 20 t ha <sup>-1</sup> |
|                               | %                    |                       |                       |
| Without                       | 19.79 ab             | 22.01 a               | 28.32 a               |
| <i>Azotobacter</i>            | A                    | A                     | A                     |
| <i>Azotobacter vinelandii</i> | 34.20 c              | 61.55 c               | 38.85 a               |
|                               | A                    | C                     | B                     |
| Indigenous                    | 14.08 a              | 29.68 ab              | 33.98 a               |
| <i>Azotobacter</i> tailing-1  | A                    | B                     | B                     |
| Indigenous                    | 22.31 abc            | 30.74 ab              | 38.13 a               |
| <i>Azotobacter</i> tailing-2  | A                    | A                     | A                     |
|                               | 28.86 ab             | 41.13 b               | 52.74 b               |
| Consortium                    | A                    | AB                    | B                     |

Means followed by the same letter are not significantly different according to Duncan's Multiple Range Test (DMRT) at the 5% significance level. Capital letters indicate comparisons among cattle manure compost rates within the same *Azotobacter* species (row comparison), whereas lowercase letters indicate comparisons among *Azotobacter* species within the same cattle manure compost rate (column comparison).

In contrast, the *Azotobacter* consortium showed increasing Pb reduction efficiency with increasing compost rates, reaching 53.74% at 20 t ha<sup>-1</sup>, although this value was not significantly different from that at 10 t ha<sup>-1</sup>. Higher organic matter inputs likely enhanced microbial activity and increased the abundance of functional groups capable

of binding Pb, thereby influencing its mobility and availability in the tailings medium [36,37]. Despite these reductions, final Pb concentrations in all treatments remained above 220 mg kg<sup>-1</sup> and were therefore still classified as high risk according to World Health Organization criteria [4]. Nevertheless, the combinations of *Azotobacter vinelandii* with 10 t ha<sup>-1</sup> cattle manure compost and the *Azotobacter* consortium with 20 t ha<sup>-1</sup> compost demonstrated considerable potential for enhancing Pb phytoremediation in gold mine tailings, although their effectiveness remained lower than the 88% Pb reduction reported following inoculation with *Bacillus wiedmannii* and *Bacillus altitudinis* [38].

#### IV. CONCLUSION

The interaction between *Azotobacter* inoculation and cattle manure compost application significantly affected *Azotobacter* population density at 13 WAP, final Pb concentration, and Pb removal efficiency in gold mine tailings. In contrast, plant height was independently influenced by both *Azotobacter* inoculation and cattle manure compost application. The combination of *Azotobacter vinelandii* and 10 t ha<sup>-1</sup> cattle manure compost resulted in the highest Pb removal efficiency, reaching 61.55%. Meanwhile, the consortium combined with 20 t ha<sup>-1</sup> cattle manure compost produced the highest *Azotobacter* population density at 13 WAP. In addition, consortium inoculation and the application of 20 t ha<sup>-1</sup> cattle manure compost each significantly promoted vetiver growth. These findings demonstrate that integrating *Azotobacter* inoculation with cattle manure compost application is a promising strategy for enhancing Pb phytoremediation of gold mine tailings using vetiver.

The combination of *Azotobacter vinelandii* with 10 t ha<sup>-1</sup> cattle manure compost and the *Azotobacter* consortium with 20 t ha<sup>-1</sup> compost may be considered promising biological amendment strategies for enhancing Pb phytoremediation in moderately contaminated gold mine tailings. Further studies should evaluate successive phytoremediation cycles using renewed vetiver plants to increase Pb removal efficiency and achieve Pb concentrations below the permissible limits for soil.

#### REFERENCES

- [1] Noviardi, R., Joy, B., Sudirja, R., Setiawati, M. R., Djuwansah, M. R., Karuniawan, A., & Suryatmana, P. (2024). The effect of cow manure amendment and the ratio of gold mine tailings on sweet potato gold
- phytomining. *Journal of Degraded and Mining Lands Management*, 12(1), 6665–6673.
- [2] Nurjayati, R., Pratiwi, I., Hidayat, E., Damayanti, R., Handayani, S., Ridwan, Y. S., ... & Noviardi, R. (2026). Mercury Accumulation in Sweet Potato Plants: Effect of Gold Mine Tailing Contamination and Compost Amendment. *BioResources*, 21(1), 1990.
- [3] Ur Rahman, S., Qin, A., Zain, M., Mushtaq, Z., Mehmood, F., Riaz, L., Naveed, S., Ansari, M. J., Saeed, M., Ahmad, I., & Shehzad, M. (2024). Pb uptake, accumulation, and translocation in plants: Plant physiological, biochemical, and molecular response: A review. *Heliyon*, 10(6), e27724.
- [4] World Health Organization. (1996). Permissible limit of heavy metals in soils and plants. In *Environmental Health Criteria* 165. International Programme on Chemical Safety, World Health Organization.
- [5] Cao, J., Xie, C., & Hou, Z. (2022). Ecological evaluation of heavy metal pollution in the soil of Pb-Zn mines. *Ecotoxicology*, 31(2), 259–270.
- [6] Sharma, P., & Dubey, R. S. (2005). Lead toxicity in plants. *Brazilian Journal of Plant Physiology*, 17(1), 35–52.
- [7] Zhang, X., Chen, B., An, J., Lin, C., & Gong, P. (2025). Overlooked deforestation from global mining activities in the 21st century. *Nature Communications*, 17(1), 804. <https://doi.org/10.1038/s41467-025-67501-1>
- [8] Amri, K., & Adifa, F. (2025). Peran fitoremediasi dalam pemulihan tanah tercemar logam berat: Tinjauan mekanisme dan aplikasi. *Jurnal Greenation Ilmu Teknik*, 3(2).
- [9] Paz-Alberto, A. M., Sigua, G. C., Bauí, B. G., & Prudente, J. A. (2007). Phytoextraction of lead-contaminated soil using vetivergrass (*Vetiveria zizanioides* L.), cogongrass (*Imperata cylindrica* L.), and carabagrass (*Paspalum conjugatum* L.). *Environmental Science and Pollution Research International*, 14(7), 498–504. <https://doi.org/10.1065/espr2007.05.415>
- [10] Kumar, P. (2025). Biomass growth and remediation efficiency of *Chrysopogon zizanioides* (L.) Nash for artificially contaminated alluvial soil. *International Journal for Research in Applied Science and Engineering Technology*, 13(2), 1282–1294. <https://doi.org/10.22214/ijraset.2025.67078>
- [11] Firda, F., Mulyani, O., & Yuniarti, A. (2017). Pembentukan, karakterisasi serta manfaat asam humat terhadap adsorpsi logam berat. *Soilrens*, 14(2). <https://doi.org/10.24198/soilrens.v14i2.11032>

- [12] Jing, Y., He, Z., & Yang, X. (2007). Role of soil rhizobacteria in phytoremediation of heavy metal contaminated soils. *Journal of Zhejiang University SCIENCE B*, 8(3), 192–207.
- [13] Hindersah, R., Nurfitriana, N., & Fitriatin, B. N. (2018). Azotobacter chroococcum dan pembersihan tanah untuk menurunkan serapan kadmium oleh tanaman padi (*Oryza sativa* L.). *Agrologia*, 6(1). <https://doi.org/10.30598/a.v6i1.176>
- [14] Das. H. K. (2019). *Azotobacters* as biofertilizer (pp. 1–43).
- [15] Aisyah, A. N., Priyadarshini, R., & Siswanto, S. (2025). Effect of Azotobacter sp. and cow manure on nitrogen availability in saline soil, root length, and vitamin C content of tomato. *Jurnal Teknik Pertanian Lampung*, 14(6), 2111–2119. <https://doi.org/10.23960/jtepl.v14i6.2111-2119>
- [16] Hindersah, R., Nurhabibah, G., & Harryanto, R. (2021). Inokulasi *Azotobacter* dan aplikasi kompos untuk bioremediasi tailing terkontaminasi merkuri. *Jurnal Teknologi Mineral dan Batubara*, 17(1), 39–46.
- [17] Hindersah, R., Mulyani, O., & Osok, R. (2017). Proliferation and exopolysaccharide production of Azotobacter in the presence of mercury. *Biodiversity Journal*, 8, 21–26.
- [18] Hanifah, N. N., Yuliani, Y., & Fitrhidajati, H. (2018). Bioremediasi tanah tercemar minyak bumi dengan penambahan kompos berbahan baku limbah cair tahu dan kulit pisang. *Jurnal Lentera Bio: Berkala Ilmiah Biologi*, 7(1).
- [19] Badan Standardisasi Nasional. (2009). SNI 6989.8:2009—Water and wastewater: Test method for lead (Pb) using flame atomic absorption spectrophotometry (FAAS) [in Indonesian]. Badan Standardisasi Nasional.
- [20] Bashan, Y., de-Bashan, L. E., Prabhu, S. R., & Hernandez, J.-P. (2014). Advances in plant growth-promoting bacterial inoculant technology: Formulations and practical perspectives (1998–2013). *Plant and Soil*, 378(1–2), 1–33.
- [21] Wu, C., Herold, R. A., Knoshaug, E. P., Wang, B., Xiong, W., & Laurens, L. M. L. (2019). Fluxomic analysis reveals central carbon metabolism adaptation for diazotroph *Azotobacter vinelandii* ammonium excretion. *Scientific Reports*, 9(1), 13209. <https://doi.org/10.1038/s41598-019-49717-6>
- [22] Yuniarti, A., Suriadikusumah, A., & Gultom, J. U. (2018). Pengaruh pupuk anorganik dan pupuk organik cair terhadap pH, N-total, C-organik, dan hasil pakcoy pada Inceptisols. *Seminar Nasional 2017*, 213–219.
- [23] Upadhyay, S. K., Srivastava, A. K., Rajput, V. D., Chauhan, P. K., Bhojiya, A. A., Jain, D., Chaubey, G., Dwivedi, P., Sharma, B., & Minkina, T. (2022). Root exudates: Mechanistic insight of plant growth promoting rhizobacteria for sustainable crop production. *Frontiers in Microbiology*, 13.
- [24] Núñez, C., López-Pliego, L., Ahumada-Manuel, C. L., & Castañeda, M. (2022). Genetic regulation of alginate production in *Azotobacter vinelandii*, a bacterium of biotechnological interest: A mini-review. *Frontiers in Microbiology*, 13. <https://doi.org/10.3389/fmicb.2022.845473>
- [25] Schimel, J., Weintraub, M. N., & Moorhead, D. (2022). Estimating microbial carbon use efficiency in soil: Isotope-based and enzyme-based methods measure fundamentally different aspects of microbial resource use. *Soil Biology and Biochemistry*, 169, 108677.
- [26] Shanker, A., Cervantes, C., Lozavera, H., & Avudainayagam, S. (2005). Chromium toxicity in plants. *Environment International*, 31(5), 739–753.
- [27] Rizvi, A., & Khan, Mohd. S. (2018). Heavy metal induced oxidative damage and root morphology alterations of maize (*Zea mays* L.) plants and stress mitigation by metal tolerant nitrogen fixing *Azotobacter chroococcum*. *Ecotoxicology and Environmental Safety*, 157, 9–20.
- [28] Apriliya, I., & Mulyawan, R. (2022). Utilization of *Azotobacter* as bio-fertilizer to support sustainable agriculture. *International Journal of Chemical and Biochemical Sciences*, 21, 197–206.
- [29] Tambaru, E. (2024). Analisis morfologi, faktor lingkungan, dan klorofil daun *Cassia fistula* L. dan *Bauhinia acuminata* L. di Hutan Kota Universitas Hasanuddin. *Jurnal Ilmu Alam dan Lingkungan*, 15(1).
- [30] Amaral, C. S., Silva, E. de B., Pereira, I. M., Amaral, W., Silva, M. A. P. da, & Nardis, B. O. (2019). Vetiver growth with different fertilizations in quartzite mining tailings. *Floresta e Ambiente*, 26(1). <https://doi.org/10.1590/2179-8087.026316>
- [31] Islam, M. R., Bilkis, S., Hoque, T. S., Uddin, S., Jahiruddin, M., Rahman, M. M., Rahman, M. M., Alhomrani, M., Gaber, A., & Hossain, M. A. (2021). Mineralization of farm manures and slurries for successive release of carbon and nitrogen in incubated soils varying in moisture status under controlled laboratory conditions. *Agriculture*, 11(9), 846.
- [32] Nurjanah, E., Sumardi, S., & Prasetyo, P. (2020). Pemberian pupuk kandang sebagai pembersihan tanah untuk pertumbuhan dan hasil melon (*Cucumis melo*

- L.) di Ultisol. *Jurnal Ilmu-Ilmu Pertanian Indonesia*, 22(1), 23–30. <https://doi.org/10.31186/jipi.22.1.23-30>
- [33] Kwiatkowska-Malina, J. (2018). Functions of organic matter in polluted soils: The effect of organic amendments on phytoavailability of heavy metals. *Applied Soil Ecology*, 123, 542–545. <https://doi.org/10.1016/j.apsoil.2017.06.021>
- [34] Nugroho, B. L. A., & Lestari, N. D. (2021). Pengaruh abu terbang batubara terhadap sifat kimia tanah dan serapan timbal (Pb) oleh tanaman akar wangi (*Vetiveria zizanioides* L.). *Jurnal Tanah dan Sumberdaya Lahan*, 8(2), 471–480.
- [35] Wahab, A. (2020). Kemampuan konsorsium bakteri sedimen rawa dalam menurunkan kandungan sulfat dan kandungan logam berat timbal (Pb) pada air asam tambang batubara, 40–42.
- [36] Louis, B. P., Maron, P.-A., Viaud, V., Leterme, P., & Menasseri-Aubry, S. (2016). Soil C and N models that integrate microbial diversity. *Environmental Chemistry Letters*, 14(3), 331–344.
- [37] Wiyono, L., Nazudin, N., & Manuhutu, J. (2023). Pengaruh penambahan pupuk organik terhadap penyerapan logam timbal (Pb) oleh tanaman lidah mertua (*Sansevieria trifasciata*). *Molucca Journal of Chemistry Education* (MJoCE), 12(2). <https://doi.org/10.30598/MJoCEvol12iss2pp119-127>
- [38] Pertiwi, B. L. S., Ustiatik, R., & Nuraini, Y. (2024). Lead (Pb)-Resistant Bacteria Improve *Brassica chinensis* Biomass and Reduce Pb Concentration in Pb-Contaminated Soil. *Journal of Tropical Biodiversity and Biotechnology*, 9(1), 86174. <https://doi.org/10.22146/jtbb.86174>