



### Effect of cow manure on instead mon the growth of water spinach in gold mine tailings

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#### ABSTRACT

#### Article History

Received: 15 March 2023

Revised: 19 June 2023

Accepted: 6 July 2023

Published: 4 August 2023

#### Keywords

Biomass  
Matter  
Nutrients  
Organic  
Plant growth  
Spinach  
Tailing  
Water  
Yield.

Gold mining activities produce abundant tailings waste. The negative impacts of tailing pollution on agriculture include soil profile loss, soil compaction, depletion of essential nutrients, organic matter, and soil pH, making it difficult for plants to grow. Cow manure (CM) is a well-known soil amendment to improve soil quality and facilitate root growth. Animal manure fertilizers contain a complete range of macro and micronutrients required by plants. In addition to its ability to enhance soil fertility, CM can also promote plant growth in former mining areas or mineral tailings. The objective of this study was to determine the effect of CM application rates on water spinach plant growth improvement. The experimental design used was a Completely Randomized Block Design (CRBD) with four treatments and six replications. The experimental treatments consisted of a control (without treatment), 10 t ha<sup>-1</sup>, 20 t ha<sup>-1</sup>, and 30 t ha<sup>-1</sup> of CM. The results of the experiment showed that CM application increased plant height, leaf count, stem diameter, and wet weight of water spinach plants. The application of 10 t ha<sup>-1</sup> CM showed the highest growth improvement in terms of plant height, leaf count, stem diameter, and wet weight of plants grown in gold mining tailings compared to other treatments. This experiment highlights the importance of CM for water spinach cultivation in tailing areas.

**Contribution/Originality:** Tailing has low levels of organic matter and nutrients, making it difficult to use tailing as a planting medium to support plant growth. Therefore, this study was conducted to determine the effect of using various types and doses of CM fertilizer in gold mine tailing planting media for tailing rehabilitation in water spinach cultivation.

## 1. INTRODUCTION

Mining is an activity carried out by humans that causes the most damage to the ecosystems. A negative impact of tailing to the environmental is the decline in the quality of soil due to the deposition of tailing, such as mine waste materials. Piles of tailing can eliminate soil profiles, increase soil density, reduce levels of essential nutrients and organic matter, and change low pH [1]. Tailings are generated in large quantities during gold mining activities. Large-scale mining companies can produce up to 2.500 t of tailing per day. Therefore, it is necessary to rehabilitate tailing land [2].

Gold mine tailing have a sandy or clayey texture that has very low water holding capacity, limited plant nutrient content, extreme acidity, and high micronutrient content [3]. In addition, gold mine tailing have low organic matter content and cation exchange capacity (CEC) [4]. The nature characteristic of these tailing does not support plant growth, therefore it is necessary to rehabilitate the quality of tailing on ex-mining land. The challenge in rehabilitating tailing is to increase carbon content and macro nutrients.

Organic matter is an important part of the soil system, has an important role in retaining water so that the availability of water in the soil becomes higher, and helps retain ions, thereby increasing ion exchange capacity or nutrient availability in the soil [5]. In addition, organic matter plays an important role in increasing soil fertility,

improving soil structure, increasing soil water holding capacity, enlarging soil pores, and increasing and supporting the development of soil microbial activity [6].

Manure derived from animal manure contains completely the nutrients, both macro and micro nutrients needed by plants [7]. The animal manure can improve soil fertility, such as the CM can increase the growth and yield of water spinach. The most important thing about CM is its nutrient content. The nutrient contents of CM that are important for plants are nitrogen (N), phosphorus (P), and potassium (K) [8]. The nutrient content contained in CM is needed by plants and soil fertility.

Water spinach (*Ipomoea reptans*) is the most popular leafy vegetable in most parts of Indonesia. Water spinach belongs to the Convolvulaceae family that grows in lowlands and highlands [9]. Water spinach requires a substrate rich in organic matter to support its growth. The organic matter needed by water spinach plants can be obtained from the crop or the animal residues composted. A good substrate must be able to hold moisture but not too moist which can trigger the growth of fungi and diseases [10]. According to Westphal [11] water spinach can grow optimally in soil that contains a lot of organic matter, has a high water content with a neutral to slightly acidic pH of 5.3–6.0, and plants that have a lot of nutritional content such as calories, protein, fat, carbohydrates, fiber, calcium, phosphorus, iron, sodium, potassium, vitamin A, vitamin B, vitamin C, carotene, hentriacontane, and sitosterol [12].

Water spinach has the ability to absorb metals from its growth media. Some metals that can be absorbed by water spinach include copper (Cu), iron (Fe), zinc (Zn), cadmium (Cd), nickel (Ni), and lead (Pb) [13]. The rapid growth rate of water spinach plants and the absorption of water is relatively high, so that it can be easily to absorb the nutrients and many of minerals.

The roots of water spinach plants can be a place for filtration and adsorption of suspended solids and microbial growth [14]. The experiment aimed was focused on the investigated the effect of using the variation of the CM composition to increasing plant height, number of leaves, stem diameter, and shoot wet weight of water spinach plants (*Ipomoea reptans*) in gold mine tailing.

## 2. MATERIALS AND METHODS

### 2.1. Experimental Implementation

The experiment was conducted in a greenhouse located at Jatiningor (Figure 1). Faculty of Agriculture, Padjadjaran University, at an altitude of  $\pm 725$  meters above sea level. The research site is located in the tropics, with climatic conditions in the research area generally influenced by tropical monsoon winds, having two seasons, namely the dry season and the rainy season.

The research was conducted from November 2022 to February 2023.

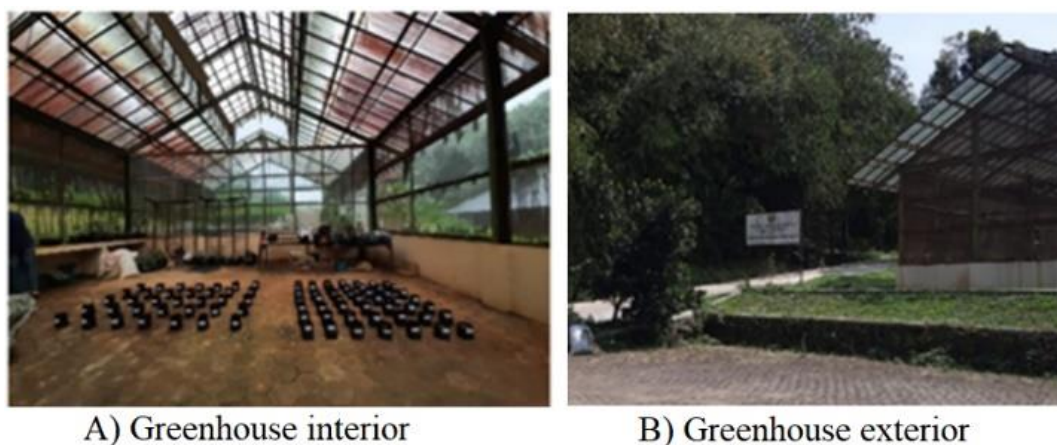


Figure 1. The pot experiment in a greenhouse.

The tailing samples were taken in Karanglayung Village, Karangjaya Sub-district, Tasikmalaya Regency. The tailing used had a slightly alkaline pH (8,08), relatively low organic matter content (1,57%), and low  $K_2O$  (12,28 mg/100g). The CM was obtained from the Beef Cattle Farm, Faculty of Animal Husbandry, Padjadjaran University with characteristics of organic carbon (C-organic) (29,25%), nitrogen (N) (1,47%), C/N ratio (19,89), and pH (6,05).

Water spinach cv Bangkok LP1 produced by East West Seed. A planting medium used was a mixture of gold mine tailing adding the CM at a variation composition of 1 kg per polybag. The tailing was previously dried then grinding was carried out for breaking the large tailing particle to fine particles size, then it was filtered using a 2 mm sieve.

Furthermore, was added by the CM, then mixed by stirring until homogeneous then put into polybags until each polybag contained a mixture of tailing and CM as much as 1 kg and incubated for 3 days at field capacity before planting (Figure 2).



A) Dewatering of the tailing      B) Grinding process of the tailing      C) Tailing sieving      D) Sieved tailing

Figure 2. Stages of tailing preparation before used as plant growth media.

### 2.2. Gold Mine Tailing Collection

The tailing location was determined based on based on the Former Gold Mine Land Distribution Study Map Tailing was collected from Karangpaningal, Karanglayung Village, Karangjaya District, Tasikmalaya Regency, West Java, Indonesia with coordinates of  $7^{\circ}23'11''\text{S } 108^{\circ}15'27''\text{E}$  (Figure 3).



A) Tasikmalaya map

B) Research location map

Figure 3. Location of gold mine tailing collection.

The mine material was extracted from a depth of 56 meters, then the gold was extracted using a grinding machine (drum) by amalgamation using mercury ( $\text{Hg}$ ) as illustrated in Figure 4. After that, the tailing was deposited in a pond or sump under the grinding machine for 1 month and the tailing were stored in a stockpile.



A) Mining material extraction tunnel

B) Mining material extraction

C) Amalgamation process in milling

D) Waste strong tank

E) Tailing pile

Figure 4. Gold mine tailing processing process gold mine tailing processing process.

### 2.3. Experimental Method

The experiment was arranged in a group randomized design consisting of four treatments of CM with six replications. The CM treatment consisted of four treatments, namely:

- A: 1000 g tailing (control).
- B: 950 g CM + 50 g tailing (5% CM).
- C: 850 g CM + 150 g tailing (10% CM).
- D: 800 g CM + 200 g tailing (15% CM).

This experiment data analyzes was using SPSS statistical by testing the normality of the observed data to determine the normality distribution. Furthermore, for normally distributed data, analysis of variance was conducted to determine the effect of treatment on the measured response at the 5% significance level. Treatments that have a real effect are further tested with Duncan's Multiple Range Test (DMRT) to determine the difference in the mean value of the response between treatments.

### 2.4. Experimental Establishment

Water spinach seeds are planted in planting holes 5 cm deep and each polybag consists of 3 planting in each hole with a distance of 5 cm. After all seeds were inserted into each planting hole, then cover the planting hole again using soil around the planting area and water ing with as much as 50 mL in each polybag. The distance between polybag is of 20x20 cm (Figure 5). Water spinach plants were planted in polybags for 35 days after planting (DAP). The N-P-K compound fertilizer (16-16-16) was applied at 15 DAP by incorporating 2,25 g per plant to the circular holes approximately 2,5 cm from the stem. The plant height, stem diameter, and number of water spinach leaves were measured at 1 to 5 weeks after planting (WAP). At the end of experiment, the plants were harvested.



A) Water spinach plant at 1 WAP



B) Water spinach plant at 3 WAP



C) Water spinach plant at 5 WAP

Figure 5. Pot experiment of water spinach on mixed tailing and CM substrate.

## 3. RESULTS

### 3.1. Plant Height (cm)

The height growth of water spinach plants aged 1–5 WAP each treatment of CM is presented in Table 1. The treatment of 10 t ha<sup>-1</sup> water spinach plants showed higher growth in the observation every week compared to the treatment of 20 t ha<sup>-1</sup> and 30 t ha<sup>-1</sup>. The treatment without the application of CM (control) revealed the plants very slow growth. The plants did not respond to the addition of organic matter, resulting in slow growth of plant height.

The results of Duncan's further test in Table 1 shows that the application of CM has no significant effect on the height of water spinach plants at the age of 1 WAP and 2 WAP, but has a significant effect on plants at 5 WAP. The treatment of 10 t ha<sup>-1</sup> is the highest plant height, it is 54,67% higher than the control at 5 WAP and the lowest height plant of water spinach plants are found in the treatment of 30 t ha<sup>-1</sup>, as is 21,74% lower than the control treatment at the age of 5 WAP. The height of water spinach plants in each gross fertilizer treatment at 3 WAP and 4 WAP was significantly different from the control.

**Table 1.** Effect of CM application on the height of water spinach planted in tailing with various concentrations of CM.

Treatment	Plant height (cm)				
	1 WAP	2 WAP	3 WAP	4 WAP	5 WAP
Control	6.21 ± 0.67	8.14 ± 0.84	8.98a ± 0.55	4.56a ± 0.29	11.45a ± 1.39
10 t ha <sup>-1</sup>	7.73 ± 3.56	10.46 ± 2.60	14.57d ± 1.15	16.21d ± 0.96	17.71c ± 0.75
20 t ha <sup>-1</sup>	5.81 ± 2.09	8.73 ± 1.73	12.07c ± 1.41	13.84c ± 2.22	15.17b ± 1.19
30 t ha <sup>-1</sup>	7.01 ± 0.97	8.63 ± 0.80	10.47b ± 0.34	11.86b ± 0.55	13.94b ± 0.96

**Note:** Numbers followed by the same letter indicate no significant difference based on the 5% significance level (Multiple Range Test, Duncan's post hoc test).

### 3.2. Number of Leaves (Sheet)

The growth pattern of the number of water spinach leaves at the age of 1 to 5 WAP with various doses of CM. The growth of the leaf count shows a relatively uniform pattern across different levels of CM dose treatment. Water spinach plants treated with a dose of 10 t ha<sup>-1</sup> of CM have a higher number of leaves. The average number of water spinach leaves at the age of 1-5 WAP due to the CM dose treatment is presented in Table 2.

The analysis of variance results in Table 2 indicate that the application of CM does not have a significant effect on the number of water spinach leaves at the age of 1-2 weeks after planting (WAP), but it does have a significant effect at 3-5 WAP. The treatment with 10 t ha<sup>-1</sup> of CM at 5 WAP is the best treatment as it shows a 36,72% increase compared to the control, while the lowest number of water spinach leaves is found in the treatments of 20 t ha<sup>-1</sup> and 30 t ha<sup>-1</sup>, which are 9,57% lower than the 10 t ha<sup>-1</sup> treatment.

**Table 2.** Effect of CM application on the number of water spinach leaves planted on tailing with various concentrations of cow manure.

Treatment	Number of leaves (Sheet)				
	1 WAP	2 WAP	3 WAP	4 WAP	5 WAP
Control	2.00 ± 0.00	3.50 ± 0.18	3.94a ± 0.14	4.22a ± 0.27	5.61a ± 0.61
10 t ha <sup>-1</sup>	2.00 ± 0.00	4.00 ± 0.52	5.33c ± 0.30	6.61c ± 0.25	7.67c ± 0.30
20 t ha <sup>-1</sup>	1.89 ± 0.27	3.94 ± 0.14	4.94b ± 0.39	6.17b ± 0.28	7.00b ± 0.56
30 t ha <sup>-1</sup>	2.00 ± 0.00	3.83 ± 0.46	5.17bc ± 0.18	6.11b ± 0.17	7.00b ± 0.30

**Note:** Caption: Numbers followed by the same letter indicate no significant difference based on the 5% significance level (Multiple Range Test, Duncan's post hoc test).

### 3.3. Stem Diameter (mm)

The stem diameter growth of water spinach plants at the age of 1-5 WAP in the treatment of CM. The growth of stem diameter of water spinach plants in the treatment of CM 10 t ha<sup>-1</sup> at the age of 5 WAP showed a higher stem diameter compared to the treatment of CM 20 t ha<sup>-1</sup> and 30 t ha<sup>-1</sup>, while the lowest stem diameter was found in water spinach plants that were not treated with CM.

The results of Duncan's further test in Table 3 show that the application of CM has no significant effect on the diameter of water spinach stems at the age of 1-3 WAP, but has a significant effect on plants at the age of 4-5 WAP. At 5 WAP, the 10 t ha<sup>-1</sup> treatment was 38,32% higher than the control, while 30 t ha<sup>-1</sup> was significantly different from the control because it was 23,35% smaller, but not significantly different from the 20 t ha<sup>-1</sup> treatment because it was 0,48% smaller at 5 WAP.

**Table 3.** Effect of CM application on the diameter of water spinach stems planted on tailing with various concentrations of CM.

Treatment	Stem diameter (mm)				
	1 WAP	2 WAP	3 WAP	4 WAP	5 WAP
Control	0.91 ± 0.26	1.17 ± 0.11	1.34 ± 0.20	1.64a ± 0.05	1.67a ± 0.03
10 t ha <sup>-1</sup>	1.06 ± 0.21	1.28 ± 0.06	1.52 ± 0.11	2.05c ± 0.17	2.31c ± 0.14
20 t ha <sup>-1</sup>	1.08 ± 0.15	1.2 ± 0.11	1.38 ± 0.14	1.86b ± 0.07	2.07b ± 0.18
30 t ha <sup>-1</sup>	1.17 ± 0.21	1.3 ± 0.09	1.48 ± 0.06	1.74ab ± 0.10	2.06b ± 0.19

**Note:** Caption: Numbers followed by the same letter indicate no significant difference based on the 5% significance level (Multiple Range Test, Duncan's post hoc test).

### 3.4. Fresh Weight of Shoots (g)

The treatment effect of CM to the wet weight of water spinach plants aged 35 days after planting (DAP). The highest of fresh weight of water spinach plants is found in the treatment of 10 t ha<sup>-1</sup> with a weight of 1,80 gram, while the smallest crown wet weight was found in the control treatment with a weight of 1,06 gram. This is because the wet weight of water spinach plants at 20 t ha<sup>-1</sup> and 30 t ha<sup>-1</sup> decreased.

The results of Duncan's post hoc test in Figure 6 indicate that the application of CM has a significant effect on the wet weight of water spinach plants. The wet weight in the treatment of 10 t ha<sup>-1</sup> is significantly different from the wet weight without CM (control), as it is 69,81% higher.

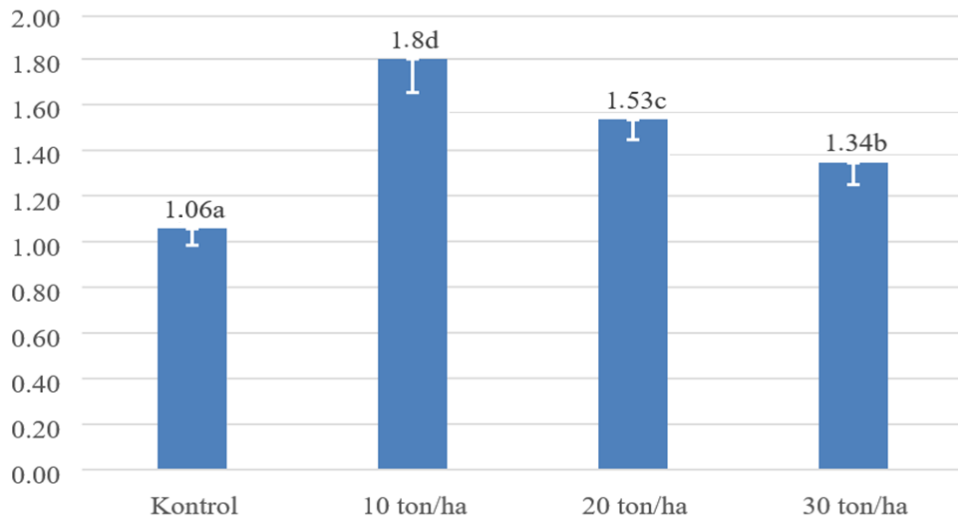


Figure 6. Fresh weight of shoots of water spinach at 1–5 WAP grown in tailing with various doses of CM.

### 3.5. Plant Pests and Diseases.

During water spinach cultivation, pests and diseases commonly encountered include the armyworm and leaf spot disease (Figure 7). The armyworm is a leaf-feeding pest, and damage symptoms occur when the larvae feed on the leaves. They start eating from the leaf edges until only the midrib remains. Mechanical control is employed by manually removing the armyworms from the leaves.

Leaf spot disease in water spinach is caused by the fungi *Cercospora bataciola* and *Fusarium* sp. Disease symptoms manifest as brown to black spots on the leaves. Among the treated plants, those subjected to a dosage of 30 t ha<sup>-1</sup> showed a higher incidence of leaf spot disease compared to plants under other treatments. Without intensive control measures, this disease can cause significant damage. Disease control is carried out mechanically by uprooting the affected water spinach plants and removing them from the cultivation area to prevent the spread of the leaf spot disease to other water spinach plants.



Figure 7. Attacks of plant disrupting organisms Attacks of plant disrupting organisms.

## 4. DISCUSSION

The results of the study showed that the CM dosage had a significant effect compared to the absence of CM. The utilization of CM can enhance soil fertility and improve the physical, chemical, and biological properties of the soil [15], which in turn leads to increased growth and development of water spinach plants. Cow manure (CM) contains essential nutrients for plant growth, such as nitrogen, phosphorus, potassium, as well as micronutrients including sulfur, magnesium, and calcium. These nutrients play a crucial role in the growth and development of water spinach plants, and they are present in sufficient quantities in CM for plant growth [16].

Organic matter is a vital component of the soil system. It plays a significant role in water retention, thereby increasing the availability of water in the soil. It also aids in retaining ions, enhancing the ion exchange capacity and nutrient availability in the soil [17]. Additionally, organic matter contributes to improving soil fertility, enhancing soil structure, increasing water retention capacity, enlarging soil pores, and supporting the development of soil microbial activity [18]. Cow manure (CM) is a potential material that can be utilized as organic fertilizer [19].

The organic carbon and nitrogen content in the soil are crucial factors for water spinach growth since organic carbon increases nutrient availability for plants [20]. This research demonstrates that the application of CM can improve the growth of water spinach plants in tailing media. A study conducted by Samkol [21] showed that a high content of organic carbon contributes to increased growth and production of water spinach. These findings are further supported by the research conducted by Yan, et al. [22], which indicates that a high organic carbon content enhances nutrient availability for water spinach plants. The optimal dose of CM for the growth of water spinach plants, as compared to other CM dosage treatments, is 10 t ha<sup>-1</sup>. This is because excessive dosages can lead to suboptimal absorption of nutrients, which hinders the promotion of water spinach plant growth [23]. Applying excessive amounts of cow manure (CM) can also increase the C/N ratio, resulting in nutrient competition in plants, particularly nitrogen. Excessive nutrients such as nitrogen, phosphorus, and potassium can damage the plant's root system and interfere with the photosynthesis process, leading to leaf yellowing and even plant death [24]. Cow manure (CM) in excessive amounts can cause toxicity in water spinach plants. The high concentration of nutrients in CM can be harmful to water spinach plants because the plants cannot efficiently absorb these nutrients, which can damage plant cells [25]. Therefore, the application of an appropriate dose of CM is an important consideration for plant growth. Soil microorganisms play a role in decomposing organic matter, releasing nutrients in a form that can be used by plants, degrading toxic residues, promoting plant growth, and enhancing the production of growth hormones, vitamins, and organic acids. They also play a crucial role in stimulating root growth [26]. Additionally, microorganisms contribute to soil fertility and play an important role in plant growth and health. High microbial populations in the soil can lead to favorable soil physical and chemical properties [27]. Microbes have significant roles in nitrogen fixation, phosphate solubilization, and phytohormone production, which enhance plant growth. Microbes such as *Rhizobium* and *Azotobacter* bacteria aid in nitrogen fixation by converting atmospheric nitrogen into compounds that can be utilized by plants, thereby increasing the availability of nitrogen for plants [28]. On the other hand, arbuscular mycorrhizal fungi (CMA) help plants absorb phosphorus from the soil by producing organic acids and phosphatase enzymes. This demonstrates that CMA can increase phosphorus availability for plants and enhance plant productivity [29].

## 5. CONCLUSION

This research shows that the application of CM to gold mine tailings contributes to an increase in plant height, leaf number, stem diameter, and shoot fresh weight in the growth of water spinach (*Ipomoea reptans*). The application of 10 t ha<sup>-1</sup> of CM resulted in the highest growth improvement of water spinach in gold mine tailings. Excessive application of CM can lead to high nutrient concentrations that can be detrimental to water spinach plants by damaging plant cells [25]. Therefore, the application of the appropriate dose of CM is an important consideration for plant growth.

**Funding:** This research is supported by the Academic Leadership Grant from Universitas Padjadjaran (Grant Number: 2203/UN6.3.1/PT.00/2022).

**Institutional Review Board Statement:** The Ethical Committee of the Universitas Padjadjaran, Indonesia has granted approval for this study (Ref. No. 2203/UN6.3.1/PT.00/2022).

**Transparency:** The authors state that the manuscript is honest, truthful, and transparent, that no key aspects of the investigation have been omitted, and that any differences from the study as planned have been clarified. This study followed all writing ethics.

**Competing Interests:** The authors declare that they have no competing interests.

**Authors' Contributions:** All authors contributed equally to the conception and design of the study. All authors have read and agreed to the published version of the manuscript.

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