

Research

Enhancing phytoremediation of bauxite mine subsoil by *Jatropha curcas* L. using sewage sludge and poultry sludge

Mingyuan Lim¹ · Abd. Wahid Samsuri² · Mohd. Yunus Abd Shukor³ · Lai-Yee Phang¹

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Abstract

Phytoremediation is a sustainable technology for cleaning up heavy metal contamination at mining sites. However, degraded soils at these sites create a harsh environment for plants to survive and properly yield biomass. In this study, sewage sludge and poultry sludge were applied as soil amendments in bauxite mine subsoil to determine their impact on the growth and heavy metal uptake of *Jatropha curcas* L. Both sewage sludge and poultry sludge were applied at 25% and 50%. *J. curcas* was grown in the amended soils for 120 days under greenhouse conditions. Changes in soil physico-chemical properties, plant growth and heavy metal uptake of *J. curcas* were determined after that. An increase in EC, CEC, total C, total N, total available P and total extractable K was detected in the amended soils. These improvements enhanced the growth of *J. curcas*, particularly in the development of above-ground plant biomass. Increased plant biomass subsequently led to higher bioaccumulation and translocation efficiency of Al, Fe, Pb and Zn. As a result, higher heavy metal removal of up to 98.03% was detected in the amended treatments. The findings indicated that the application of sewage sludge and poultry sludge improves soil conditions for plant development.

Keywords Phytotechnology · *Jatropha curcas* · Sustainability · Waste management · Soil amendment · Heavy metal uptake

1 Introduction

Phytoremediation is a remediation technology that utilizes plant species to extract, sequester and/or detoxify contaminants from the environment [1]. It is regarded as more sustainable and less invasive as compared to conventional physical and chemical treatments. Apart from providing in situ environmental remediation, its holistic approach also has the potential of achieving additional environmental benefits such as improvement of soil functionality, control of soil erosion, wildlife rehabilitation and production of economically viable crops at the same time [2, 3]. As such, repurpose and reuse of the remediated site is possible, which opens up opportunities for long-term financial returns [4]. In Malaysia, unregulated mining activity and poor post-mining management have left barren bauxite mining sites unvegetated, leading to severe red mud floods. These floods have contributed to water pollution, contamination of water sources and the loss of natural fishing areas [5]. This situation highlights the urgent need for effective remediation technologies and strategies

✉ Lai-Yee Phang, phanglaiyee@upm.edu.my; Mingyuan Lim, Ming_OnATrip@live.com; Abd. Wahid Samsuri, samsuriaw@upm.edu.my; Mohd. Yunus Abd Shukor, mohdyunus@upm.edu.my | ¹Department of Bioprocess Technology, Faculty of Biotechnology and Biomolecular Sciences, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia. ²Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia. ³Department of Biochemistry, Faculty of Biotechnology and Biomolecular Sciences, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia.



to address these environmental problems. Research on phytoremediation techniques to solve the issues associated with bauxite-mined soil in Malaysia is still limited. However, the application of phytoremediation for rehabilitating bauxite-mined sites has been very successful worldwide.

Hyperaccumulator plants are plants that exhibit unusually high accumulation of heavy metals and metalloid trace elements in their tissues without suffering from visible toxicity symptoms [6]. For example, accumulation of more than 1% of manganese (Mn); more than 0.3% of zinc (Zn); more than 0.1% of arsenic (As), chromium (Cr), nickel (Ni) and/or lead (Pb); more than 0.03% of cobalt (Co) and/or copper (Cu); more than 0.01% of cadmium (Cd), selenium (Se) and/or thallium (Tl) and more than 0.001% of mercury (Hg) in the plant shoot dry mass is considered as hyperaccumulation [7, 8]. Hyperaccumulator plants have long been the target plants for phytoremediation in heavy metal-contaminated soils. However, this presents two major shortcomings, which are the slow growth rate and low biomass production found in hyperaccumulator species [9–11]. Moreover, poor soil functionality of contaminated sites creates a harsh condition for non-hyperaccumulator and non-metal tolerant plant species to survive.

The application of municipal and industrial wastewater sludge as soil amendment is an effective way to overcome the aforementioned challenges faced in phytoremediation. Additionally, it offers a sustainable waste management approach. Management of sewage sludge is one of the most concerning environmental issues in Malaysia, due to the large amount of sewage sludge produced following expanded population, rapid urbanization and economic development, which makes sludge disposal challenging [12, 13]. Sewage sludge is a suitable soil amendment because it has the ability to improve soil properties such as porosity, bulk density, aggregate stability and water retention capacity [13]. Meanwhile, the discharge of poultry sludge, or abattoir wastewater sludge has also increased due to the increased poultry meat production in order to meet the rising demand for human consumption [14]. Proper management and disposal of poultry sludge is required as it could be an environmental and human health hazard due to its high organic impurities [14, 15]. The use of poultry sludge as a soil amendment is appealing as it contains high content of total nitrogen (TN), total phosphorus (TP) and total organic carbon (TOC) which are essential for plant growth [14].

Previous study has demonstrated the potential of *J. curcas* to be grown in bauxite mine subsoil as a biofuel and phytoremediation crop [16]. Nevertheless, its growth was limited by the poor nutrient content in bauxite mine subsoil. Limited growth will undoubtedly restrict the potential and hinder the establishment of *J. curcas* as an economically practical biofuel crop in marginal soils [10, 17]. The use of industrial sludge in phytoremediation could solve two issues at the same time, namely the limited plant growth on contaminated soils and proper management of industrial sludges. Therefore, this study aimed to investigate the impact of sewage sludge and poultry sludge on the growth and heavy metal uptake of *J. curcas* in bauxite mine subsoil. The efficacy of sewage sludge and poultry sludge as soil amendment was evaluated by determining the changes in soil physico-chemical properties following their application.

2 Materials and methods

2.1 Study area

The whole study was conducted in Universiti Putra Malaysia (UPM). The pot experiment was carried out in a 100 m² greenhouse located in Taman Pertanian Universiti, Universiti Putra Malaysia, Serdang (2°58'54.1"N 101°42'49.2"E) from January 2019 to April 2019. The mean monthly temperature and average annual rainfall in Serdang were 26.9 °C and 2369 mm respectively throughout the pot experiment. Subsequent soil and plant analyses were carried out in laboratories situated in the Faculty of Biotechnology and Biomolecular Sciences and the Faculty of Agriculture, UPM.

2.2 Soil sampling

Subsoil was sampled from the bauxite mining site at FELDA Bukit Goh, Kuantan (3°55'09.8"N 103°14'58.0"E). The soil is classified as Geric Ferralsols which produced bauxite from deep weathering. The lower horizon subsoil was originally lying as deep as 6 m underground but was exposed due to mining activities. At the time of sampling, bauxite mining was halted as a result of the implementation of a governmental moratorium. About 300 kg of soil was packed and transported to UPM in gunny bags.

2.3 Test plant

J. curcas seedlings were germinated from seeds yielded from previous *J. curcas* trees that were purchased from the supplier BIOTECH EQUIPMENT & ENTERPRISE in Johor, Malaysia. After germination, the seedlings were allowed to grow for a month under optimal conditions with a daily irrigation of 200 ml water. Subsequently, the healthy seedlings were transplanted to the treatment media in black polybags for the pot experiment.

2.4 Soil characterization

The physico-chemical properties of the treatment soils were determined using the methods described in Soil Analysis: Handbook of Reference Methods [18] with necessary modifications, unless stated otherwise.

Soil sample was mixed with 1 N potassium chloride (KCl) in a ratio of 1 g: 2.5 mL for pH determination. Similarly, soil electrical conductivity (EC) was determined by mixing the soil sample with deionized water in a ratio of 1 g: 2.5 mL. Cation exchange capacity (CEC) was evaluated using the shaking method by using 1 N ammonium acetate (NH₄OAc) as the leaching agent. Atomic absorption spectrometer (AAS) was used as the detector. Total carbon (C) and nitrogen (N) were determined using a carbon/nitrogen/sulfur (CNS) determinator based on the Dumas method from Jimenez and Ladha [19] with modifications. Meanwhile, total available phosphorus (P) and extractable potassium (K) were determined according to the Bray 2 method and leaching method with NH₄OAc, respectively.

Mehlich 1 solution was used as the extractant to determine the bioavailability of targeted heavy metals in the soils. The heavy metals aluminium (Al), iron (Fe), lead (Pb) and zinc (Zn) were targeted in this study to investigate their immediate effects on the plant. The term 'bioavailability' refers to the portion of heavy metals in the soil that are readily available for plant uptake, such as free soluble metal ions and metal ions adsorbed on ion exchange sites. The determination was carried out according to the method described in Soil Analysis: Handbook of Reference Methods [17]. Mehlich 1 was prepared by mixing 0.05 N of concentrated hydrochloric acid (HCl) and 0.025 N of concentrated sulfuric acid (H₂SO₄) prior to making up volume with deionized water. Soil sample was then immersed in Mehlich 1 and shaken for 5 min before filtrating the suspension. Upon filtration, the filtrate was injected into inductively coupled plasma-optical emission spectrometer (ICP-OES) for heavy metal detection. Standards for ICP with a stock concentration of 1000 mg/L from Sigma-Aldrich were used as the certified reference material for heavy metal detection. The standards were diluted to four different concentrations in order to construct the standard curve before the sample analysis.

2.5 Pot experimental design

Two kinds of soil amendments were used in this study, namely sewage sludge and poultry sludge. Sewage sludge and poultry sludge were collected from Indah Water Konsortium and Worldsign Industries Sdn. Bhd. respectively. After drying, each sludge was then mixed with bauxite mine subsoil in two ratios, which were 25% and 50% (w/w). The treatments were as follows: C (100% bauxite mine subsoil as control), SS25 (25% sewage sludge + 75% bauxite mine subsoil), SS50 (50% sewage sludge + 50% bauxite mine subsoil), PS25 (25% poultry sludge + 75% bauxite mine subsoil) and PS50 (50% poultry sludge + 50% bauxite mine subsoil). After mixing, the treatment soils were left to stabilize for two weeks. Acclimatized *J. curcas* seedlings were transplanted to approximately 20 kg of the treatment soil. The plants were then aligned in Randomized Complete Block Design (RCBD) with triplicates for each treatment. An irrigation scheme of 200 mL water in every two days was provided to the plants. The pot experiment lasted for 120 days in a greenhouse condition where the average humidity was at 80% and natural sunlight was the sole light source [16].

2.6 Soil analysis

Treatment soils were sampled at the end of pot experiment for analysis. Rhizospheric soil was collected, air dried, ground and sieved through a 2 mm sieve as a preparation step. The tests described in section "soil characterization" were carried out to evaluate the impact of each treatment on the physico-chemical properties of soil.

2.7 Plant analysis

2.7.1 Growth measurement

The growth of *J. curcas* was evaluated through the measurement of growth parameters which included the number of leaves, plant height and basal diameter [20]. Only healthy leaves were counted. A measuring tape and digital calipers were used in measuring plant height and basal diameter of the plants, respectively. The growth measurement was taken in an interval of 15 days throughout the pot experiment.

2.7.2 Biomass and heavy metal analysis

The plants were harvested and separated into root, stem and leaf at the end of the pot experiment. They were washed with tap water and rinsed with distilled water before being air dried. After measuring the fresh weight, the plant samples were dried in an oven at 55 °C in order to remove moisture. The dry weight was then measured when constant weight was achieved.

Dry ashing method was adopted to determine the heavy metal concentration in the plant samples. The plant samples were ground, and heated in a muffle furnace at 30 °C and subsequently 500 °C. After the heating stage, the plant samples were digested with 20% nitric acid (HNO₃) in a water bath at 80 °C for an hour. They were then brought up to volume with deionized water. Lastly, the solution was filtered through Whatman no. 1 filter paper and analyzed by ICP-OES for heavy metal determination.

2.7.3 Heavy metal accumulation and translocation efficiency

Three indices, namely biological accumulation coefficient (BAC) [21, 22], biological transfer coefficient (BTC) [21, 22] and bioconcentration factor (BCF) [21, 23] were computed to evaluate the heavy metal accumulation and translocation efficiency of *J. curcas* in this study. Each index is calculated based on the following equations:

$$\text{BAC} = \frac{\text{heavy metal concentration in plant shoot}}{\text{heavy metal concentration in soil}}$$

where: BAC refers to biological accumulation coefficient,

$$\text{BTC} = \frac{\text{heavy metal concentration in plant shoot}}{\text{heavy metal concentration in plant root}}$$

where: BTC refers to biological transfer coefficient,

$$\text{BCF} = \frac{\text{heavy metal concentration in plant root}}{\text{heavy metal in soil}}$$

where: BCF refers to bioconcentration factor.

2.8 Statistical analysis

One-way ANOVA was performed to determine the statistical significance of the results by comparing the means in soil physico-chemical properties, plant growth performance and plant heavy metal uptake. All statistical analysis in this study was performed by using SPSS version 16.0 (SPSS Inc., Chicago, IL, USA) at 95% confidence level.

Table 1 Physico-chemical properties and heavy metal bioavailability of the treatment soils at day 0 of pot experiment

Treatment	C	SS25	SS50	PS25	PS50
Day 0					
pH	5.73 ± 0.20	4.36 ± 0.30	3.65 ± 0.15	6.17 ± 0.26	5.95 ± 0.19
EC (µS/cm)	249.33 ± 18.15	2113.33 ± 179.26	3000.00 ± 265.14	1563.00 ± 52.62	1954.00 ± 182.43
CEC (cmol/kg)	3.03 ± 0.21	9.02 ± 0.55	14.05 ± 0.45	20.45 ± 0.87	29.54 ± 0.31
Total C (%)	0.16 ± 0.07	2.60 ± 0.26	6.04 ± 0.99	6.42 ± 0.88	7.67 ± 1.89
Total N (%)	Traces	0.68 ± 0.11	0.77 ± 0.02	0.67 ± 0.08	0.73 ± 0.22
Total Available P (µg/g)	Traces	Traces	478.94 ± 196.80	1548.63 ± 181.16	1746.03 ± 146.02
Total Extractable K (µg/g)	35.23 ± 3.60	155.37 ± 16.10	1041.60 ± 124.84	718.53 ± 32.24	1032.20 ± 102.51
Al (mg/kg)	1532.13 ± 75.34	1835.20 ± 29.92	5020.00 ± 339.718	8561.33 ± 626.90	8184.00 ± 249.06
Fe (mg/kg)	154.72 ± 29.16	351.17 ± 64.05	1611.47 ± 116.89	135.08 ± 18.75	147.11 ± 10.92
Pb (mg/kg)	20.91 ± 8.28	19.64 ± 16.68	21.76 ± 5.71	11.55 ± 6.11	18.67 ± 4.78
Zn (mg/kg)	2.75 ± 0.37	1106.67 ± 281.82	4665.33 ± 747.80	205.48 ± 25.15	212.76 ± 18.70

C 100% bauxite mine subsoil as control; SS25 25% sewage sludge + 75% bauxite mine subsoil; SS50 50% sewage sludge + 50% bauxite mine subsoil; PS25 25% poultry sludge + 75% bauxite mine subsoil; PS50 50% poultry sludge + 50% bauxite mine subsoil. Data presented here represent mean ± standard deviation with three replicates

3 Results and discussion

Table 1 shows the physico-chemical properties of the treatment soils at day 0 of the pot experiment. As indicated, the different kinds of sludge impacted the soil pH differently. A lower soil pH was recorded in soils amended with sewage sludge while a high pH was recorded in soils amended with poultry sludge. Soil pH in bauxite mine soil was reported to be negatively correlated to organic matter content [16]. Therefore, a pH-lowering effect was expected from sewage sludge amendment as sewage sludge has high organic matter content [13]. High organic matter content was also observed in poultry sludge as indicated by the high total C and N shown in Table 1. Yet, poultry sludge amendment had an increment effect on soil pH. This could be due to the utilization of hydrogen (H⁺) ions in the process of urea hydrolysis by ammonia-producing bacteria which are found in poultry waste [24–26].

A significant improvement was observed in the EC of amended soils. Soil EC increased to as high as 3000.00 µS/cm in amended soil. Addition of both sewage sludge and poultry sludge enhanced soil EC to the range of moderate to high salinity based on the Australian salinity class [27]. The level of organic matter and macronutrient also improved significantly following the addition of sewage sludge and poultry sludge. The highest total carbon (C) and total available phosphorus (P) were recorded in PS50, notching 7.67% and 1746.03 µg/g, respectively. Meanwhile, the highest total nitrogen (N) and total extractable potassium (K) were recorded in SS50, notching 0.77% and 1041.60 µg/g, respectively. As reported previously, positive impacts of organic wastes as soil amendment include enhancement of soil aggregate stability, soil moisture content, water retention capacity, the concentration of organic matter and macronutrients [13, 28, 29].

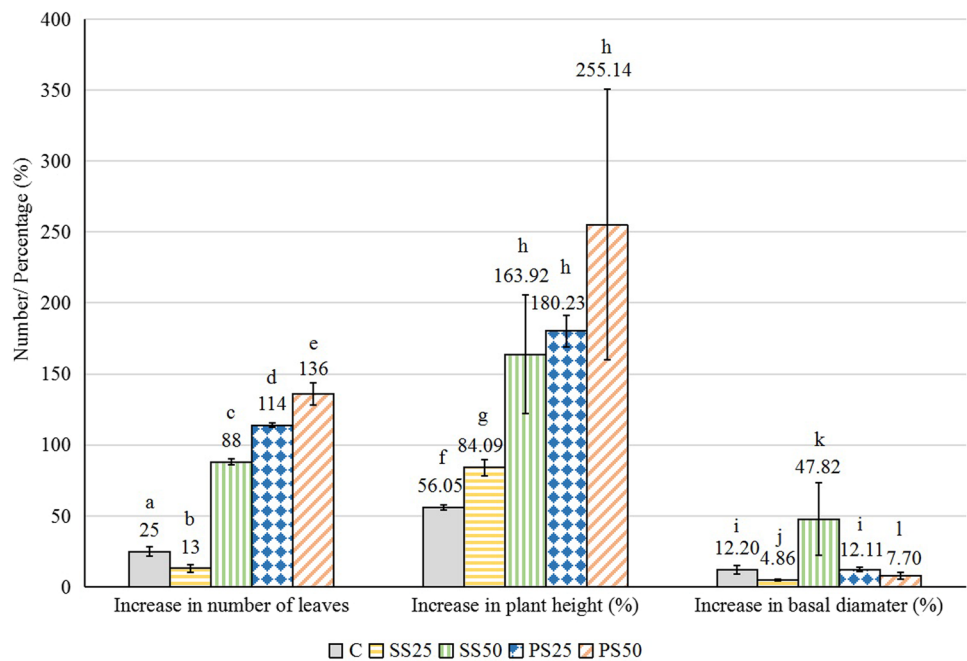
Bioavailability of heavy metals can provide a direct insight into their potential impact on plant performance as they are the portion of heavy metals that are readily available for plant uptake [30, 31]. The bioavailability of heavy metals in the treatment soils at day 0 is listed in Table 1. When sewage sludge or poultry sludge was added, the soils became more concentrated with bioavailable heavy metals. This is evident when comparing the bauxite soil without any amendments (designated as C) to the bauxite soil with the addition of sewage sludge (SS25 and SS50) and poultry sludge (PS25 and PS50). Poultry sludge had a larger impact on the bioavailability of Al, with PS25 recording the highest value, 8561.33 mg/kg. On the contrary, sewage sludge had a greater impact on the bioavailability of Fe and Zn, where the highest concentration was recorded in SS50, with values of 1611.47 and 4665.33 mg/kg, respectively. Poultry waste has been reported to have the potential to enrich the heavy metal concentration in soils as it contains fairly rich amounts of heavy metals such as chromium (Cr), copper (Cu), Fe, nickel (Ni), Pb and Zn [32–34]. However, the presence of Cu, Cr, Pb and Zn at the range of 0.004 mg/L–22 mg/L, 0.0007–0.14 mg/L, 0.09–2.73 mg/L, 0.001–0.52 mg/L in the reported poultry waste was relatively low [33]. The concentration of heavy metals in poultry wastes, referred to as abattoir samples, varied based on the waste handling processes and the specific waste

Table 2 Physico-chemical properties of the treatment soils at day 120 of pot experiment

Treatment	C	SS25	SS50	PS25	PS50
Day 120					
pH	5.32±0.26	2.96±0.33	2.88±0.36	6.24±0.15	6.49±0.12
EC (µS/cm)	226.00±4.95	2100.00±70.00	2663.33±80.83	1893.33±188.53	1579.67±96.44
CEC (cmol/kg)	2.68±0.13	8.87±0.77	12.33±0.78	18.45±0.58	28.37±0.18
Total C (%)	0.88±0.03	5.60±0.61	6.17±2.75	3.86±0.54	6.89±1.20
Total N (%)	0.00±0.00	0.54±0.13	0.63±0.09	0.54±0.03	0.77±0.07
Total Available P (µg/g)	Traces	Traces	657.88±334.55	1314.93±194.53	1321.69±149.48
Total Extractable K (µg/g)	55.50±4.07	243.87±47.80	679.22±78.60	730.77±156.55	1092.80±31.40

C 100% bauxite mine subsoil as control; SS25 25% sewage sludge + 75% bauxite mine subsoil; SS50 50% sewage sludge + 50% bauxite mine subsoil; PS25 25% poultry sludge + 75% bauxite mine subsoil; PS50 50% poultry sludge + 50% bauxite mine subsoil. Data presented here represent mean ± standard deviation with three replicates

Fig. 1 The overall growth performance of *J. curcas*. C: 100% bauxite mine subsoil as control; SS25: 25% sewage sludge + 75% bauxite mine subsoil; SS50: 50% sewage sludge + 50% bauxite mine subsoil; PS25: 25% poultry sludge + 75% bauxite mine subsoil; PS50: 50% poultry sludge + 50% bauxite mine subsoil. Different letters indicate that the means of each growth parameter between treatments were significantly different at $p < 0.05$. Error bars indicate mean ± standard deviation (SD) with replicates



collection locations. This is evidenced in another study [34] whereby the abattoir samples contained 728 mg/kg Fe, 0.74 mg/kg Pb, 20.61 mg/kg Zn, 17.49 mg/kg Cu, 0.21 mg/kg Cr and 10.39 mg/kg Ni. Application of sewage sludge also could increase the concentration and bioavailability of heavy metals like Cu, manganese (Mn), Ni, Pb and Zn in direct proportion to the rate of application [35, 36]. The concentrations of Pb, Cd and Cu in sewage sludge obtained from Malaysia's national sewerage company, varied from 36 to 308 mg/kg, 0.51 to 6.49 mg/kg and 63 to 732 mg/kg, respectively and the concentration of Mn and Ni ranged from 32 to 420 mg/kg and 10 to 151 mg/kg, respectively [37]. However, the concentrations of Cu, Mn, Pb and Zn in sewage sludge were 4.93, 9.73, 10.35 and 68.38 mg/kg, respectively [38], which are relatively lower than those reported previously. Hence, the application of sewage sludge and poultry sludge in this study is considered extrinsic, leading to elevated concentrations of heavy metals in the bauxite soil, particularly because the studied heavy metals are present in significant amounts in sludge. Conversely, if the heavy metal content in the sludge is low or negligible, the impact on soil concentrations will be minimal.

The impact of soil amendment on the bioavailability of heavy metals in soil was influenced by the changes in soil physico-chemical properties. Soil pH and organic matter content were among the main influences in this study. As soil pH decreases, the bioavailability of heavy metals like Al increases, thus increasing the susceptibility to phytotoxicity [39–41]. Elevated level of organic matter content also provided additional adsorptive medium for the heavy metals in soil [42].

At the end of the pot experiment, the physico-chemical properties of amended soils remained favorable for plant growth as displayed in Table 2. The highest EC was recorded in SS50, with a value of 2663.33 $\mu\text{S}/\text{cm}$. Meanwhile, PS50 had the highest pH, CEC, total C, total N, total available P and total extractable K by the end of the pot experiment. This implies that PS50 has the potential to produce plants with the best growth. Optimal plant growth leading to increased biomass is a crucial factor in determining the success of phytoremediation efforts [43]. The growth of *J. curcas* in bauxite mine subsoil with and without amendment is presented in Fig. 1.

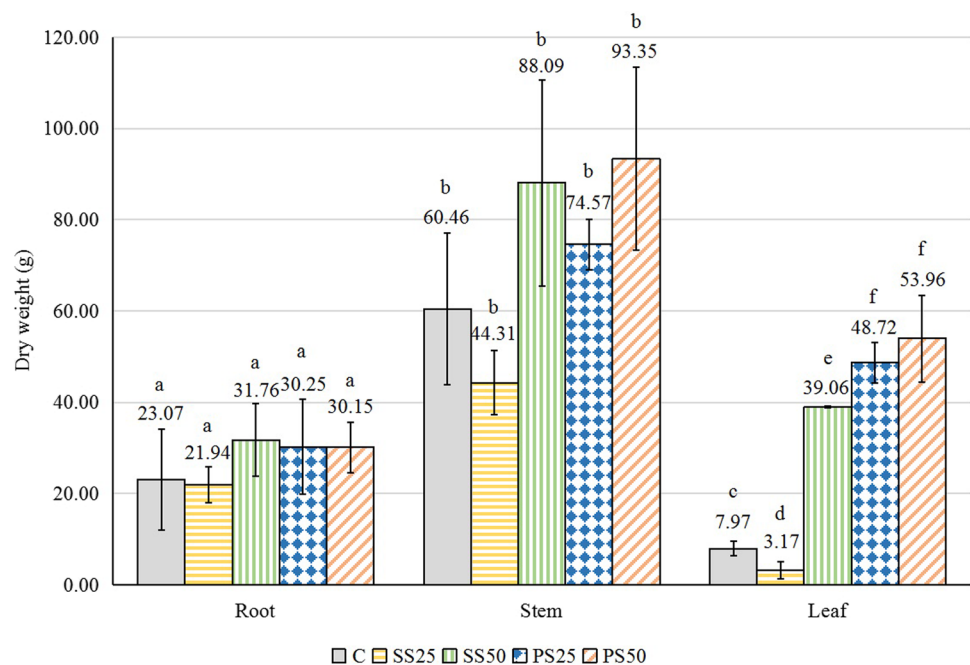
The application of soil amendment had a strikingly positive effect on the plant growth. The highest increase in number of leaves was recorded in *J. curcas* grown in PS50, where an increase of 136 was observed. The highest increase in plant height was recorded in PS50 as well, where a total of 255.14% increment was observed. Meanwhile, there was no significant impact observed on the increase in basal diameter, except for *J. curcas* grown in SS50, where a 47.82% increment was recorded.

As shown in Fig. 2, there was a slight, insignificant increase in the root biomass of *J. curcas* grown in the amended soils compared to that in the control soil. The root biomass produced by *J. curcas* in this study ranged from 21.94 g in SS25 to 31.76 g in SS50. Similarly, although the application of soil amendment produced *J. curcas* with higher stem biomass compared to the ones without soil amendment, the difference was not statistically significant. PS50 produced the highest stem biomass recorded, which was 93.35 g. The stem biomass production could be limited by the lack of significant increment in basal diameter despite having a considerable increase in plant height (Fig. 1). The leaf biomass produced by *J. curcas* grown in bauxite mine subsoil with soil amendment was significantly larger than that of the control. The highest leaf biomass production was measured in PS50, which was 53.96 g. The enhanced production of the stem and leaf biomass could lead to increased photosynthetic activity and subsequently better plant performance.

Figures 1 and 2 also imply that *J. curcas* had improved growth regardless of the higher heavy metal bioavailability following the application of soil amendment. There were no visible heavy metal toxicity symptoms observed on the plants, even though high concentrations of Al and Fe could induce nutrient deficiency, especially in acidic conditions [44]. However, the adverse condition was seemingly alleviated by the high nutrient content of the soil amendments which improved plant growth [35]. Improved plant growth and biomass production following the application of sewage sludge and poultry waste have also been documented in previous studies [28, 35, 45–47].

Most Al and Fe accumulation occurred in the root of *J. curcas* as depicted in Fig. 3a and b. The highest Al and Fe accumulation were both detected in the root of *J. curcas* grown in the control. Al accumulation in the plant root across the treatments ranged from 4244.22 mg/kg in SS50 to 5996.86 mg/kg in PS25. Meanwhile, Fe accumulation in the plant root across the treatments ranged from 3669.56 mg/kg in PS50 to 5397.78 mg/kg in SS25. The high accumulation of metals in plant roots may result from direct soil contact and the presence of the rhizosphere, which enhances the absorption and metabolism of heavy metals within plant tissues [38, 48]. Additionally, the movement of metals

Fig. 2 The biomass production by *J. curcas* grown in bauxite mine subsoil with and without amendment after 120 days of pot experiment. C: 100% bauxite mine subsoil as control; SS25: 25% sewage sludge + 75% bauxite mine subsoil; SS50: 50% sewage sludge + 50% bauxite mine subsoil; PS25: 25% poultry sludge + 75% bauxite mine subsoil; PS50: 50% poultry sludge + 50% bauxite mine subsoil. Different letters indicate that the means of biomass between treatments were significantly different at $p < 0.05$. Error bars indicate mean \pm SD with replicates



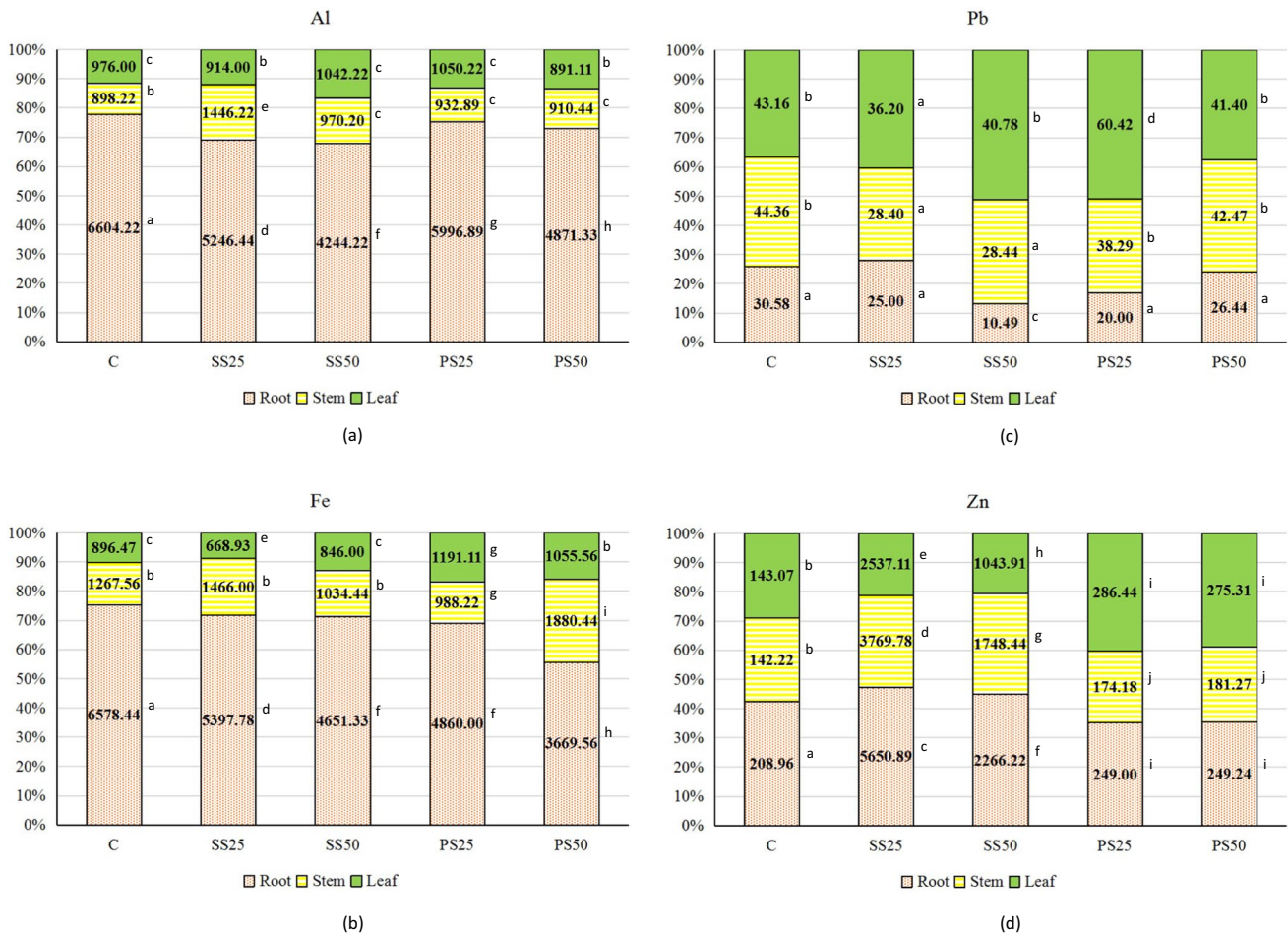


Fig. 3 Heavy metal uptake (mg/kg): **a** Al; **b** Fe; **c** Pb and **d** Zn by different parts of *J. curcas* grown in C: 100% bauxite mine subsoil as control; SS25: 25% sewage sludge + 75% bauxite mine subsoil; SS50: 50% sewage sludge + 50% bauxite mine subsoil; PS25: 25% poultry sludge + 75% bauxite mine subsoil; PS50: 50% poultry sludge + 50% bauxite mine subsoil. Different letters indicate that the means of heavy metal uptake between treatments and plant parts were significantly different at $p < 0.05$

may be restricted by the high cation-exchange capacity of the plant cell wall [49]. However, a higher accumulation of Pb could be observed in the above-ground tissues (stem and leaf) as shown in Fig. 3c instead of the root of the plants. Despite that, typical Pb toxicity symptoms like yellow and dark brown dots were not observed on the leaves of the plants [50]. Pb toxicity could be avoided by Pb compartmentalization in the above-ground plant tissues through mechanisms like vacuolar sequestration, homeostasis and stress protection [4].

A higher percentage of Zn accumulation was observed in the plant leaves of the poultry sludge treatments, which was a different trend compared to that of the control and sewage sludge treatments (Fig. 3d). Additionally, the translocation efficiency for Zn could be seen as having increased following the application of poultry sludge as indicated by the higher BTC when compared to control (Table 3). This implies that Zn was effectively translocated to the leaf tissues for storage in the poultry sludge treatments. A similar observation was reported by Sahito et al. [51] where the translocation of Zn was higher in test plants grown in soil amended with poultry waste. The significantly higher leafing intensity observed in the poultry sludge treatments could be contributing to the higher Zn accumulation in plant leaves (Figs. 1 and 2). Zn accumulation in plant shoots is regarded as a strategy employed by plants to overcome high Zn concentrations that could induce phytotoxicity [52, 53].

The application of soil amendment improved the translocation efficiency of Al in *J. curcas*, as evident by the increased BTC against the control. The highest BTC for Al was recorded in SS50, which was 0.71. An increase in BAC, BTC and BCF for Fe were also observed in the soil amendment treatments, especially in the poultry sludge treatments. Higher bioaccumulation and translocation efficiency of Fe might be related to the higher plant growth and

Table 3 Biological accumulation indices of *J. curcas* grown in bauxite mine subsoil with and without soil amendment

Sample	Al	Fe	Pb	Zn
BAC				
C	1.23±0.27	14.59±4.61	4.57±0.44	68.23±41.28
SS25	1.10±0.24	5.51±0.91	4.90±2.02	2.62±0.30
SS50	0.47±0.03	1.33±0.25	5.88±0.43	1.37±0.02
PS25	0.24±0.07	23.91±4.12	6.73±4.61	2.28±0.07
PS50	0.22±0.03	21.90±1.22	4.83±0.92	2.16±0.34
BTC				
C	0.37±0.11	0.36±0.09	2.86±0.15	1.34±0.25
SS25	0.54±0.04	0.41±0.01	12.09±6.99	1.23±0.10
SS50	0.71±0.09	0.53±0.08	4.00±1.53	1.16±0.14
PS25	0.47±0.12	0.46±0.04	5.44±1.50	1.90±0.19
PS50	0.56±0.05	0.97±0.23	4.27±0.18	1.91±0.27
BCF				
C	4.34±1.22	45.58±14.53	1.61±0.24	56.13±23.46
SS25	2.31±0.56	13.71±0.07	2.46±0.88	2.10±0.38
SS50	1.02±0.26	3.29±0.88	3.42±0.43	1.24±0.08
PS25	0.70±0.12	51.50±12.54	2.85±0.34	1.22±0.25
PS50	0.60±0.02	24.95±3.41	1.52±0.52	1.19±0.18

BAC Biological accumulation coefficient ([heavy metal shoot]/[heavy metal soil]); BTC Biological transfer coefficient ([heavy metal shoot]/[heavy metal root]); BCF Bioconcentration factor ([heavy metal root]/[heavy metal soil]); C 100% bauxite mine subsoil as control; SS25 25% sewage sludge + 75% bauxite mine subsoil; SS50 50% sewage sludge + 50% bauxite mine subsoil; PS25 25% poultry sludge + 75% bauxite mine subsoil; PS50 50% poultry sludge + 50% bauxite mine subsoil. Data presented here represent mean ± standard deviation with three replicates

biomass production, particularly in the plant shoot as observed in the poultry sludge treatments since Fe is vital for plant processes such as photosynthesis, respiration and metabolic activities [51].

Although Pb has no known biological function, the plant shoots exhibited a higher Pb accumulation than the plant roots. The application of soil amendment further enhanced BAC, BTC and BCF for Pb. Application of poultry sludge and sewage sludge have demonstrated similar enhancement effects on the translocation and bioaccumulation efficiency of heavy metals in plants previously [35, 51]. The uptake of heavy metals by plants is influenced by the bioavailability of heavy metals in soil, which in turn, can be altered by the application of soil amendment through mechanisms like the formation of organic and inorganic complexes, increased surface charge, metal reduction and also metal precipitation [54]. Soil nutritional level is also considered as another factor in enhancing heavy metal uptake by plants by increasing the bioavailability of heavy metals [51].

Despite the improved bioaccumulation and translocation efficiency displayed by the amended treatments, the heavy metal uptake rate (mg/kg) was not significantly different than that of the control (Fig. 3). However, the heavy metal removal from the soil showed a considerable improvement as depicted in Fig. 4. The highest Al and Fe removal achieved were measured in PS50, with values of 94.61% and 98.03% respectively (Fig. 4a and b). Meanwhile, the highest Pb and Zn removal was recorded in SS50 (66.28%) and PS25 (63.63%) respectively (Fig. 4c and d). The difference was stark when compared to the heavy metal removal of the control, which were 19.22% (Al), 34.11% (Fe), 21.94% (Pb) and 46.61% (Zn).

The enhancement could be attributed to the higher growth and biomass production stimulated by the application of sewage sludge and poultry sludge. Plant biomass proves to be a crucial factor in heavy metal removal efficiency. Aishah et al. [35] stated that the uptake ability of Zn and Cu was directly proportional to the plant biomass which was influenced by the application rate of sewage sludge. It was reported that the enhanced uptake of Zn and Cu by *J. curcas* and *Hibiscus cannabinus* was a result of increased biomass production stimulated by the application of sewage sludge. Ebbs et al. [55] reported that *Brassica juncea* exhibited a higher Zn removal efficiency and credited it to its higher biomass against *Thlaspi caerulescens*, despite the fact that *T. caerulescens* is a known Zn hyperaccumulator. In this study, the application of sewage sludge and poultry sludge improved the growth and increased the biomass production of *J. curcas* significantly (Figs. 1 and 2), thereby increasing the capacity for total heavy metal accumulation, leading to a higher amount of heavy metal being removed from the soil (Fig. 4).

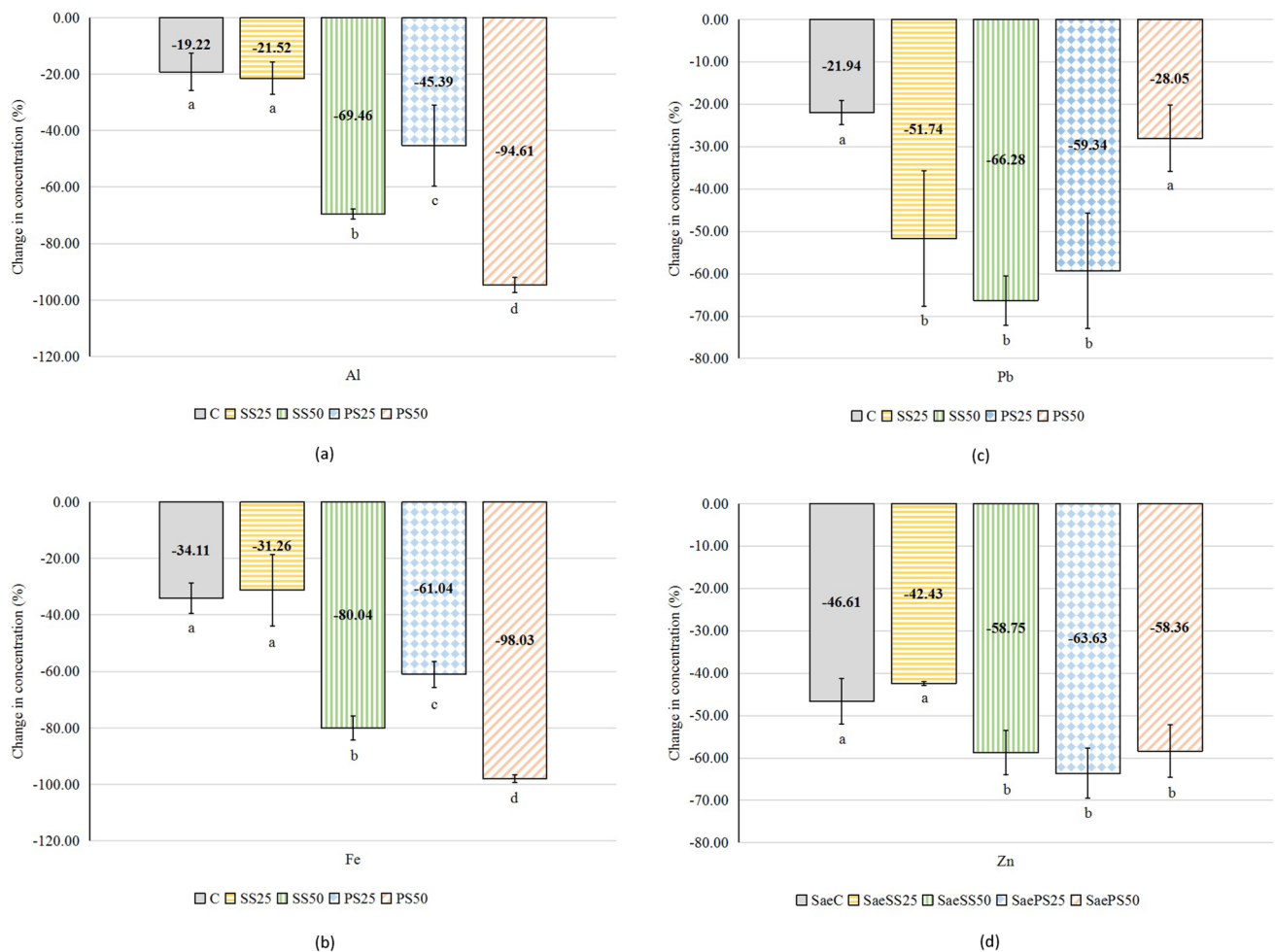


Fig. 4 The changes in the concentration of bioavailable heavy metal in the treatments after soil amendment experiment; **a** Al, **b** Fe, **c** Pb and **d** Zn. C: 100% bauxite mine subsoil as control; SS25: 25% sewage sludge + 75% bauxite mine subsoil; SS50: 50% sewage sludge + 50% bauxite mine subsoil; PS25: 25% poultry sludge + 75% bauxite mine subsoil; PS50: 50% poultry sludge + 50% bauxite mine subsoil. Different letters indicate that the means between treatments were significantly different at $p < 0.05$. Error bars indicate mean \pm SD with replicates

4 Conclusions

As demonstrated in the findings of this study, application of sewage sludge and poultry sludge ameliorated the condition in bauxite mine subsoil for plant growth. Essential soil properties like EC, CEC, total C, total N, total available P and total extractable K were enhanced significantly following the application of both sewage sludge and poultry sludge. This considerably improved the growth of *J. curcas* compared to the control. Higher growth and biomass production by *J. curcas* consequently augmented heavy metal removal from the soil to a great extent. Application of sewage sludge and poultry sludge also improved the bioaccumulation efficiency of Fe and Pb, as well as the translocation efficiency of Al, Fe, Pb and Zn in *J. curcas*. The findings established that sewage sludge and poultry sludge improve soil conditions for plant development which enhance the efficacy of phytoremediation.

Author contributions All authors contributed to the conception of the paper. The literature search was performed by all authors. The first draft of the manuscript was written by Mingyuan Lim and Lai-Yee Phang. All figures were prepared by Mingyuan Lim. Tables 1 and 2 were prepared by Abd. Wahid Samsuri and Mohd Yunus Abd Shukor. Table 3 was prepared by Lai-Yee Phang. Data analysis was performed by Mingyuan Lim and Lai-Yee Phang. Lai-Yee Phang, Abd. Wahid Samsuri and Mohd Yunus Abd Shukor commented on the previous version of the manuscript and critically revised the manuscript. All authors read and approved the final manuscript.

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Data availability All data underlying the results are available as part of the article and no additional source data are required.

Declarations

Competing interests The authors declare no competing interests.

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