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Research Article

Removal of chromium from chromium-contaminated soil and physiological response of shallot (*Allium ascalonicum* L.) on treatments of biochar and mycorrhizae

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Abstract

Food safety and soil degradation were the reasons to treat contaminated soil. Shallots are high-value commodities, so cultivation is carried out intensively. Continuous use of agrochemicals can cause heavy metal contamination. This study aimed to investigate chromium removal, physiological characters, and yield of shallot (*Allium ascalonicum* L.) on biochar and mycorrhizae application on chromium-contaminated soil. A pot experiment was conducted at the screen house ex-farm of the Faculty of Agriculture, Jenderal Soedirman University. The treatments tested consisted of two factors. The first factor was biochar dosage (B) consisting of 4 levels, i.e., B0 = without biochar, B1 = 1.2 g biochar kg^{-1} of soil, B2 = 2.4 g biochar kg^{-1} of soil, and B3 = 4.8 g biochar kg^{-1} of soil. The second factor was mycorrhizae inoculation consisting of 3 levels, i.e., M0 = without mycorrhizae, M1 = 0.1 g mycorrhizae kg^{-1} of soil, M2= 0.2 g mycorrhizae kg^{-1} of soil. The twelve treatments were arranged in a randomized block design with three replications. The results showed that the application of 1.2 g, 2.4 g, and 4.8 g biochar kg^{-1} of soil had been able to increase plant height and the percentage of root infection. The application of mycorrhizae 0.1 g and 0.2 g mycorrhizae kg^{-1} of soil was able to increase plant height, percentage of root infection, and plant tissue P uptake. Both applications of biochar and mycorrhizae increased plant height and the percentage of root infection by mycorrhizae.

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Introduction

Shallot (*Allium ascalonicum* L.) is an important horticultural commodity that has high economic value. The nutritional content in every 100 g of shallots contains 88 g water, 1.5 g protein, 6.3 g fat, 9 g carbohydrates, 0.7 g fiber, 0.6 g ash, 40 mg P, 0.8 mg Fe, 36 mg Cu, 5 IU vitamin A, 0.03 mg vitamin B1, 2 mg vitamin C with an energy value of 160 KJ 100 g^{-1} (Fitriani and Darda, 2018). Besides being used as a cooking spice, shallots can also be used as medicine, including cholesterol reducer and blood sugar reducer (Mohammadi-Motlagh et al., 2011). There are many different benefits, including antibacterial and antifungal properties (Amin and Kapadnis, 2005; Yin and Tsao, 1999), beneficial haematological influences (Owoyele et al., 2004), and antioxidant properties (Leelarungrayub et al., 2006). Shallot cultivation is prone to failure due to environmental factors and pests, so the use of fertilizers and pesticides is massive. Increased public awareness of food quality and safety is a major concern in the production and marketing of vegetables. Domestic vegetable products generally have inconsistent quality and have a fairly high level of contamination (Miskiyah and Munarso, 2009). Shallots are the main kitchen spice, so the level of consumption in the community is quite high.

The practice of shallot cultivation uses agrochemicals continuously, so it is possible that the absorption of heavy metal residues in shallot plants in the long term will be harmful to human health (Shekhawat et al., 2015). The issue of food safety is one of the topics that must be considered. Food contamination can be caused by the accumulation of heavy metals in soil, water, or plants (Stoica et al., 2020). Heavy metal contamination in soil is an important concern issue because of its great impact on human health and ecological systems (Alghanmi et al., 2015). Chromium (Cr) is a type of heavy metal that can pollute the environment and its residues accumulated in plant tissues. According to SNI, the threshold for Cr in the soil is 5 ppm (Thakur et al., 2007; Puspita et al., 2011). A soil is considered polluted by Cr if the concentration of Cr in the soil is above 10 ppm (Bruce, 2002). The increasing Cr accumulation in the soil cause the nutrient cycle in the soil to be disrupted and ultimately decrease soil fertility. Sources of heavy metals in agricultural soil come from the use of synthetic fertilizers and pesticides, industrial waste, motor vehicle emissions, and household waste that enters irrigation canals (Unver et al., 2015). The consumption of chromium that can be tolerated by the human body is a maximum of 0.07 ppm per day (Yang et al., 2020). The accumulation of Cr in the human body can cause damage to the respiratory organs and cause cancer in humans (Tasneem et al., 2021)

Efforts to remediate heavy metal polluted soil can be made by utilizing soil amendment materials such as biochar and mycorrhizal biofertilizers. Biochar is a soil amendment of carbon produced from the pyrolysis process of organic materials such as rice straw, corn cobs, or wood. The pyrolysis process or heating is carried out in a kettle with a high temperature of 300-500 ºC (Shenbagavalli and Mahimairaja, 2012). Biochar can stabilize heavy metals in polluted soil by reducing the absorption of heavy metals by plants and can improve the physical, chemical, and biological properties of the soil (Ippolito et al*.,* 2012; Komarek et al., 2013). Lignocellulosic waste is agricultural waste containing cellulose, hemicellulose, and lignin that are hard to be converted into other compounds biologically. Cellulose is a carbon source that can be used by microorganisms as a substrate in the fermentation process (Shofiyanto, 2008). According to Lehmann and Joseph (2009), biochar can be produced from organic materials that are difficult to decompose, which are burned imperfectly (pyrolysis) or without oxygen at high temperatures, 300-500 ºC, so that agricultural waste can be useful for remediation of contaminated soil. Mycorrhizae have been reported to be potential for remediating heavy metal-contaminated soils (Riri et al., 2011). Mycorrhizal fungi can play a role in the bioremediation of heavy metal pollution in soil; they reduce heavy metal toxicity by metabolizing these metals (Bano and Asfaq, 2013). Mycorrhizae symbiosis with plant roots also increases plant resistance to extreme drought and humidity and supports to accumulation of elements that are toxic to plants, such as As, Cr, and Pb. Mycorrhizae of the Glomus genus associated with plants are effective in absorbing heavy metals, such as Cd, Zn, and Pb (Riaz et al., 2021). Shallots are the main kitchen spices, so the demand is quite high. Therefore, efforts to reduce heavy metal residue on this commodity are interesting issues to be studied.

This study aimed to investigate chromium removal, physiological characters, and yield of shallot (*Allium ascalonicum* L.) upon biochar and mycorrhizae application on chromium-contaminated soil.

Materials and Methods

Preparation of Cr-contaminated soil medium

The soil medium used in the pot experiment in the screen house was Cr contaminated Inceptisols taken from shallot cultivation area in Songgom Village, Songgom District of Brebes Regency with location maps of 7.019176926846719 and 108.982348621595. The soil has a pH of 5.27 and a Cr content of 18.78 ppm. According to the SNI of Indonesia, this Cr content is above the maximum residual limit of Cr of 5 ppm in soil. The shallot cultivation area is near the main road to Cirebon, and there are technical irrigation canals. Composite soil samples were taken with a soil drill at a depth of 20 cm. The composite soil sampling method aims to obtain soil in a relatively homogeneous area (Saraswati et al., 2007).

Preparation of biochar

The biochar used in this experiment was corncob biochar which is one of the lignocellulosic wastes that are widely available in Indonesia. Corncob biomass was put into a furnace and then tightly closed with a combustion temperature between 100-300 ºC for 3-5 hours. After 3-5 hours, the combustion fire was turned off, and the furnace was allowed to cool for about 12 hours. The biochar was subjected to a refinement process by sieving (Ratnasari et al., 2020)

Preparation of mycorrhizae and shallot planting

A pot experiment conducted in a screen house was carried out from April to September 2019 at the experimental farm of the Faculty of Agriculture, Jenderal Soedirman University. The altitude of the experimental farm is 110 m above sea level, the average daily temperature is 27.5 ºC, and the average daily humidity is 67%. Glomus vesicular-arbuscular mycorrhizae mixed with zeolite material was used as a biofertilizer; 1 g of zeolite contained 10 spores of Glomus. Bima Brebes variety of shallot was planted on a polybag with the size of 35 x 40 cm consisting of 10 kg of soil. N, P, and K basal fertilizers were applied ten days after planting. During the experiment, 500 mL of water was supplied daily.

Experimental design

The treatments tested consisted of two factors. The first factor was biochar dosage (B) consisting of four levels, i.e., B0 = without biochar, B1 = 1.2 g biochar kg^{-1} of soil, B2 = 2.4 g biochar kg^{-1} of soil, and B3 = 4.8 g biochar kg^{-1} of soil. The second factor was mycorrhizae inoculation consisting of three levels, i.e., M0 = without mycorrhizae, M1 = 0.1 g mycorrhizae kg^{-1} of soil, and M2 = 0.2 g mycorrhizae kg^{-1} of soil. The twelve treatments were arranged in a randomized block design with three replications.

Variables observed

Variables observed were plant height, total root length, root dry weight, plant growth rate, plant net assimilation rate, leaf chlorophyll, percentage of root infection, P uptake by plant tissue, Cr in shallot plant tissue, Cr removal efficiency, number of tubers, and tuber weight. Chromium content in the plant was determined by AAS (Atomic Absorption Spectrometer). Removal efficiency (RE) of Cr from the soil by the plant was calculated using the formula previously used by Hardiani et al. (2009) as follows:

$$\text{RE}\ (\%) = \frac{\text{IMC} - \text{FMC}}{\text{IMC}}\ \text{x}\ 100\%$$

where:

RE = removal efficiency
IMC = initial metal concentration in soil
FMC = final metal concentration in soil

Data analysis

The data obtained from the observations were subjected to analysis of variance (ANOVA) to determine the effect of treatment, followed by the Duncan Multiple Range Test (DMRT) with a *p*-value = 0.05

Results and Discussion

The growth and ability of plants to adapt to heavy metal stress conditions are determined by the supply of nutrient and soil enhancers. Adequate nutrient absorption will make plant metabolism good. Physiological responses and yields of shallots with biochar and mycorrhizal treatments in soil conditions contaminated with Cr are shown in Table 1.

Effect of biochar on physiological characteristics and yield of shallots

Biochar has the ability to soil pollutant removal (Oni et al., 2019) and improve plant growth, and generally enhance the root uptake of several elements (Ferreiro et al., 2015). The effects of biochar on physiological characters and yields of shallots grown on chromium-contaminated soil are presented in Table 2. Data of shoot length, plant growth rate, net assimilation rate, leaf chlorophyll content, and plant tissue P uptake indicate that shallot plants can adapt to media contaminated with chromium.

Table 1. Matrix of analysis results of ANOVA application of biochar and mycorrhizae on physiological characters and yield of shallots on chromium-contaminated soil.

Observed Variables	**Effect**		
	B	**M**	**B x M**
Plant height	s	s	s
Total Root Length	ns	ns	ns
Root Dry Weight	ns	ns	ns
Crop Growth Rate	ns	ns	ns
Net Assimilation Ratio	ns	ns	ns
Leaf Chlorophyll Content	ns	ns	ns
Percentage of Root Infection	s	s	s
P uptake by Plant Tissue	ns	s	ns
Chromium content in plant tissue	s	s	ns
Chromium Removal from Soil	s	s	ns
Number of tubers per clump	ns	ns	s
Bulb weight per clump	ns	ns	s

Remarks: B = biochar, M = mycorrhizae, B x M = interaction between biochar and mycorrhizae inoculation, ns = not significantly different, s = significantly different on the ANOVA with a *p* = 0.05.

Results of the analysis of variance showed that the application of biochar had no significant effect on plant growth and physiological aspects such as roots. According to Cobbett (2000), plant cells respond to heavy metal stress using various defence mechanisms, including exclusion, immobilization, chelation, and compartmentalization of metal ions. According to Liu and Kotteke (2003), shallot plants can prevent excessive metal ions from entering the cytosol and can localize these metal ions in certain areas. Ion compartmentation in the vacuole is a mechanism that plays an important role in the detoxification process and tolerance to metal ions. Based on the data analyzed, In this study, the growth and physiological characteristics of shallot plants did not experience significant damage and were able to reach the next generative phase, namely tuber ripening. Biochar application significantly affected the accumulation of chromium in plants and the removal of chromium from the soil. Planting media that was not applied with biochar showed the highest accumulation of chromium in plant tissue at 1.21 ppm, which was significantly different from the application of 1.2 g biochar kg^{-1} of soil, which was 1.07 ppm. The addition of 2.4 g biochar kg^{-1} of soil and 4.8 biochar kg^{-1} of soil was not significantly different from the application of 1.2 g biochar kg^{-1} of soil; namely, there were accumulations of chromium of 0.96 ppm and 0.94 ppm, respectively. In this study, the application of biochar was able to reduce the accumulation of chromium in plant tissue, but the addition of a higher dose of biochar did not show a better reduction in Cr accumulation. According to Irhamni et al. (2018), heavy metals are carried from the soil through the roots, and they are then transported through the stele through the endodermis, then translocated through the xylem to the plant shoots.

Table 2. Effect of biochar on the physiological character and yield of shallots grown on chromium-contaminated soil.

Biochar Treatments	**SL (cm)**	**SW (g)**	**CGR (g day^{-1})**	**NAR (g cm^{-2}day^{-1})**	**CC (mg L^{-1})**	**P uptake (ppm)**	**Cr PT (ppm)**	**Cr R (%)**
0 g biochar kg^{-1} of soil	307.29	0.07	0.071	0.040	12.72	139.45	1.21 a	26.97 a
1.2 g biochar kg^{-1} of soil	296.76	0.06	0.106	0.02	12.55	179.27	1.07 b	35.82 a
2.4 g biochar kg^{-1} of soil	293.34	0.07	0.145	0.029	12.66	155.47	0.96 bc	50.77 b
4.8 g biochar kg^{-1} of soil	334.04	0.06	0.090	0.058	12.48	162.82	0.94 bc	53.34 b
CV (%)	24.15	15.38	25.4	21.32	14.42	22.07	20.62	16.35

Remarks: Numbers followed by different letters in the same column indicate a significantly different in the DMRT (5%). SL = Total Shoot Length, SW = Root Dry Weight, CGR = Crop Growth Rate, NAR = Net Assimilation Rate, CC = Leaf Chlorophyll Content, PU = Plant tissue uptake, Cr PT = Chromium accumulation in plant tissue, Cr R = soil Chromium removal.

Results of this experiment showed that the application of 2.4 g biochar kg^{-1} of soil and 4.8 biochar kg^{-1} of soil had the ability to remove chromium in the soil 50.77% and 53.34% better than the control. Utilization of biochar in contaminated soil is to remove the activity of heavy metal ions (Laoli et al*.,* 2020), stabilize the metal, and significantly reduce the absorption of heavy metals (Puga et al., 2015) so that the amount of chromium absorbed by the roots and translocated throughout the shallot plant tissue can be reduced.

Effect of mycorrhizae on physiological characteristics and yield of shallots

The capability of plants to obtain water and nutrients, such as phosphorus (P), may increase by the existence of mycorrhizae and subsequently promote the growth of plants even under heavy metal stress conditions (Saleh et al., 2021). The effects of mycorrhizae on the physiological characteristics of shallots grown on chromium-contaminated soil are presented in Table 3.

Table 3. Effect of mycorrhizae application on the physiological character and yield of shallots on chromium-contaminated soil.

Mycorrhizae Treatments	**SL (cm)**	**SW (g)**	**CGR (g day^{-1})**	**NAR (g cm^{-2} day^{-1})**	**CC (mg L^{-1})**	**P uptake (ppm)**	**Cr PT (ppm)**	**Cr R (%)**
0 g mycorrhizae kg^{-1} of soil	295.40	0.07	0.096	0.048	13.12	132.48 a	1.17 a	23.76 a
0.1 g mycorrhizae kg^{-1} of soil	292.52	0.06	0.085	0.044	11.84	167.68 b	0.79 b	47.62 b
0.2 g mycorrhizae kg^{-1} of soil	335.65	0.07	0.129	0.018	12.80	177.25 b	0.71 b	56.25 c
CV (%)	24.15	15.38	25.4	21.32	14.42	22.07	20.62	16.35

Remarks: Numbers followed by different letters in the same column indicate a significantly different in the DMRT (5%). SL = Total Shoot Length, SW = Root Dry Weight, CGR = Cropt Growth Rate, NAR = Net Assimilation Rate, CC = Leaf Chlorophyll Content, PU = Plant tissue uptake, Cr PT = Chromium accumulation in plant tissue, Cr R = soil chromium removal.

Application of 0.1 g mycorrhizae kg^{-1} of soil and 0.2 g mycorrhizae kg^{-1} of soil had a significant effect on increasing the P uptake of plant tissue, namely 167.68 ppm and 177.25 ppm, respectively, compared to the mycorrhizal treatment, which was only able to absorb P of 132.48 ppm. On the other hand, the application of 0.1 g mycorrhizae kg^{-1} of soil and 0.2 g mycorrhizae kg^{-1} of soil reduced the accumulation of chromium in plant tissues by 0.09 ppm and 0.11 ppm, respectively. On shallot plants that were not applied with mycorrhizae, the accumulation of chromium had the highest value of 1.17 ppm. P uptake of plant tissues is related to the ability of mycorrhizae to infect plant roots, spore multiplication occurs, and hyphae develop to reach nutrient sources. Mycorrhizal symbiosis with plant roots contributes significantly to plant nutrition, especially phosphorus absorption (Jefferies et al., 2003). Mycorrhizae also had the ability to remove chromium contaminants in soil; application of 0.2 g mycorrhizae kg^{-1} of soil showed the best treatment, which can reduce 56.25% chromium contaminant. AMF hyphae network is very efficient in nutrient absorption (Plenchette et al., 2005).

Mycorrhizae form extra radical mycelium, which is extensive to increase the area of root absorption and phosphorus is immobile so that the element is not easily absorbed except with the help of hyphae which release enzymes that break down insoluble P complexes (Vence et al., 2003). The reduced accumulation of chromium in plant tissues is due to the ability of mycorrhizae to mobilize heavy metals in the rhizosphere by deposition in the soil, adsorption to the root surface, or absorption and accumulation in the roots. Research conducted by Audet and Charest (2006) showed that heavy metal treatment by plants could accumulate in roots and shoots. Besides that, hyphae from mycorrhizae are also able to protect the tip of the root as a place for the absorption of substances that are toxic to plants (Suharno and Sancayaningsih, 2013).

Effect of the interaction between mycorrhizae and biochar on plant height, root infection, and yield of shallots

Mycorrhizal and biochar treatments showed an interaction that significantly affected plant height, percentage of root infection, number of tubers, and tuber weight. Biochar and mycorrhizal applications increased plant height and the percentage of root infection. Table 4 shows that the height of the plants treated with 0.1 g mycorrhizae kg^{-1} of soil with 1.2 g biochar kg^{-1} of soil, 2.4 g biochar kg^{-1} of soil, 4.8 g biochar kg^{-1} of soil, and 0.2 g mycorrhizae 10 kg soil^{-1} with 1.2 g biochar 10 kg soil^{-1}, 2.4 g biochar kg^{-1} of soil, 4.8 g biochar kg^{-1} of soil did not show a significant difference with a plant height range of 33.57-38.07 cm. However, the treatment was significantly different from the treatment without mycorrhizal with 4.8 g biochar kg^{-1} of soil and 0.1 g mycorrhizae kg^{-1} of soil without biochar, which resulted in plant heights of 32.62 cm and 32.66 cm.

Treatments without mycorrhizal and with 1.2 g biochar kg^{-1} of soil, 2.4 g biochar kg^{-1} of soil, and 0.2 g mycorrhizal kg^{-1} of soil with treatment without biochar showed no significant difference with the plant height range of 29.78-31.78 cm. However, the treatment was significantly different from the treatment without mycorrhizae and without biochar which was the lowest plant height, which was 26.81 cm. Rokhminarsi et al. (2019) explained that giving mycorrhizae had a significant role in increasing plant P uptake, degree of root infection, P content in the soil, dry weight of plants, and tended to suppress soil-available Cd levels and plant Cd uptake. The use of mycorrhizae at the right dose can increase soil fertility and maintain environmental balance. According to Charisma et al. (2012), mycorrhizal activity can produce organic acids and phosphatase enzymes that can change P element in the labile zone to be absorbed by plants. Sasli and Agus (2012) stated that the uptake of P nutrients increased in the mycorrhizal plant group compared to the non-mycorrhizal plant group. This is because mycorrhizae can increase the ability of roots to reach nutrient sources and dissolve phosphates. According to Permanasari et al. (2016), the ability of plants to absorb P is supported by mycorrhizae which are able to increase nutrient uptake in the soil through root infection. Mycorrhizae, which are capable of fixing with roots, are able to help roots absorb nutrients from the soil. The higher the infection rate, the more roots that contain mycorrhizae.

Mycorrhizae and biochar treatments showed a significant interaction effect on the tuber weight (Table 4). Treatments without mycorrhizae, when added with biochar, showed significantly different yields, and the yields increased, ranging from 18.56-29.76 g. The treatment of 0.1 g mycorrhizae kg^{-1} of soil with biochar had a significant effect. The total weight increased, ranging from 22.62-29.61 cm. However, the treatment of mycorrhizal 0.1 g kg^{-1} of soil on biochar 1.2 g, 2.4 g, and 4.8 g kg^{-1} of soil did not have a significant effect, and the treatment without biochar got the lowest results. Mycorrhizal treatment of 0.2 g kg^{-1} of soil with the addition of biochar gave an effect that was not significantly different, with weights ranging from 25.03-30.69 g. The application of 4.8 g biochar kg^{-1} of soil without mycorrhizae gave the highest yield of shallots (28.40 g), although this value was not significantly different from other treatments. The lowest yield of shallots (18.56 g) was observed at the control treatment (without the application of biochar and mycorrhizae).

The application of biochar and mycorrhizae increased plant height. Nurida (2014) reported that the addition of biochar increased N and P, water holding capacity of the soil so that biochar could keep nitrogen from being easily leached out from the soil. According to Kusuma (2018), the application of biochar and organic fertilizers had a significant effect on the plant height of *Brahiaria decumbens* grass because of the increase in nitrogen uptake by the plant. Interaction of biochar and mycorrhizae also can increase the percentage of root infection. This happens because biochar can provide P nutrients for plants, and mycorrhizal fungi can help the process of dissolving P so that it is more mobile to be absorbed by plants. Putri et al. (2017) reported that the application of biochar could increase the available P and total N in soils. Biochar enhanced rice crop growth and yield at sites with low availability of phosphorus (Asai et al., 2009). According to Satriawan and Handayanto (2015), plants need P for root development, accelerating flowering and maturation, as well as root and seed formation.

Table 4. Effect of interaction of application of biochar and mycorrhizae on plant height, percentage of root infection of shallots, and yield components.

Treatments		Variables			
Mycorrhizae doses	Biochar Dosage	Plant height (cm)	Root Infection Percentage (%)	Number of Tuber	Weight of Tuber (g)
0 g mycorrhizae kg^{-1} of soil	0 g biochar kg^{-1} of soil	26.81 d	18.56 d	5.00 c	18.56 d
	1.2 g biochar kg^{-1} of soil	29.78 c	24.31 b	6.41 b	24.31 b
	2.4 g biochar kg^{-1} of soil	31.78 c	28.40 a	7.28 b	28.40 a
	4.8 g biochar kg^{-1} of soil	32.62 b	29.76 a	9.50 a	29.76 a
0.1 g mycorrhizae kg^{-1} of soil	0 g biochar kg^{-1} of soil	32.66 b	22.22 c	7.33 b	22.22 c
	1.2 g biochar kg^{-1} of soil	34.17 a	27.03 a	7.72 a	27.03 a
	2.4 g biochar kg^{-1} of soil	33.97 a	28.06 a	8.72 a	28.06 a
	4.8 g biochar kg^{-1} of soil	38.07 a	29.61 a	8.94 a	29.61 a
0.2 g mycorrhizae kg^{-1} of soil	0 g biochar kg^{-1} of soil	31.20 c	25.03 a	7.00 b	25.03 a
	1.2 g biochar kg^{-1} of soil	33.57 a	26.76 a	8.25 a	26.76 a
	2.4 g biochar kg^{-1} of soil	34.65 a	27.69 a	7.42 b	27.69 a
	4.8 g biochar kg^{-1} of soil	37.89 a	30.69 a	10.78 a	30.69 a
CV (%)		25,56	22,22	31,74	25,56

Remarks: Numbers followed by different letters in the same column indicate a significant difference based on the DMRT (*p* = 0,05). CV = coefficient of variation.

P absorbed by plant roots depends on the amount and availability of P in the soil. The great contribution of the AM to plant growth and crop production in the tropics has been reviewed by Naher, Othman, and Panhwar (2013). However, among horticulture crops, allium species are more sensitive to AM application due to their less growth root system, thus causing low absorption capability of soil nutrients (Golubkina et al., 2020). The increase in growth and absorption of P was probably because of the work of mycorrhizal hyphae in reaching and dissolving P in the soil. This process made the plants metabolize well, and the photosynthate produced was distributed in tubers to increase the weight and number of tubers.

Conclusion

The condition of chromium heavy metal stress of 18.78 ppm in soil media did not affect the physiological characteristics and growth of shallot plants. Still, it is necessary to pay attention to the safety of agricultural commodities consumed and efforts to reduce heavy metal residues in foodstuffs. The application of 2.4 g biochar kg^{-1} of soil could reduce the accumulation of chromium in shallot plant tissues by 20.66% and remove soil chromium content by 50.77%. Mycorrhizae application could increase P uptake by plant tissue by 26.57%, decrease chromium uptake by 32.47%, and remove 56.25% of chromium from the soil. The interaction between 1.2 g biochar kg^{-1} of soil and 0.1 g mycorrhizae kg^{-1} of soil could increase plant height and percentage of root infection, number of tubers, and tuber weight of shallot

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References

Alghanmi, S.I., Al Sulami, A.F., El-Zayat, T.A., Alhogbi, B.G. and Salam, M.A. 2015. Acid leaching of heavy metals from contaminated soil collected from Jeddah, Saudi Arabia: kinetic and thermodynamics studies.

International Soil and Water Conservation Research 3(3):196-208, doi:10.1016/j.iswcr.2015.08.002.

Amin, M. and Kapadnis, B.P. 2005. Heat stable antimicrobial activity of Allium ascalonicum against bacteria and fungi. *Indian Journal of Experimental Biology* 43:751-759.

Asai, H., Samson, B.K., Stephan, H.M., Songyikhangsuthor, K., Homma, K., Kiyono, Y., Inoue, Y., Shiraiwa, T. and Horie, T. 2009. Biochar amendment techniques for upland rice production in Northern Laos: 1. Soil physical properties, leaf SPAD and grain yield. *Field Crops Research* 111(1-2):81-84, doi:10.1016/ j.fcr.2008.10.008.

Audet, P. and Charest, C. 2006. Effects of AM colonization on "wild tobacco" plants grown in zinc-contaminated soil. *Mycorrhiza* 16:277-283, doi:10.1007/s00572-006-0045-x.

Bano, S.A. and Asfaq, D. 2013. Role of mycorrhiza to reduce heavy metal stress *Natural Science* 5(12):16-20, doi:10.4236/ns.2013.512A003.

Bruce, R. 2002. *Chemical Transformations of Chromium in Soils: Relevance to Mobility, Bio-availability and Remediation.* The Chromium File. International Chromium Development Association, College Park USA.

Charisma, A.M., Rahayu Y.S. and Isnawati. 2012. The effect of the combination of Trichoderma compost and vesicular-arbuscular mycorrhizae (MVA) on the growth of soybean (*Glycine max* (L.) Merill) on calcareous soil growing media. *Jurnal Lentera Bio* 1(3):111-116 (*in Indonesian*).

Cobbett, C.S. 2000. Phytochelatins and their roles in heavy metal detoxification, *Plant Physiology* 123(3):825-832, doi:10.1104/pp.123.3.825.

Ferreiro, J., Gascó, G., Méndez, A. and Reichman, S.M. 2018 Soil pollution and remediation. *International Journal of Environmental Research and Public Health* 15:1657; doi:10.3390/ijerph15081657.

Fitriani, V. and Efendi, D. 2018. Effect of paclobutrazol and benzyl adenine on growth and multiplication of shallots (*Allium cepa* L.) of Bima Brebes variety by in vitro comm. *Horticulturae Journal* 2(2):22-27, doi:10.29244/chj.2.2.22-27 (*in Indonesian*).

Golubkina, N., Krivenkov, L., Sekara, A., Vasileva, V., Tallarita, A. and Caruso, G. 2020. Prospects of arbuscular mycorrhizal fungi utilization in production of Allium plants. *Plants* 9(2):279, doi:10.3390/plants9020279.

Hardiani, H., Kardiansyah, T. and Sugesty, S. 2009. Bioremediation of lead metal (Pb) in contaminated soil sludge waste paper of the drinking process industry. *Jurnal Selulosa* 1(1):31-41 (*in Indonesian*).

Ippolito J.A., Laird, D. and Busscher, W. 2012. Environmental benefits of biochar. *Journal of Environmental Quality* 41:967-972, doi:10.2134/jeq2012.0151.

Irhamni, Pandia, S., Purba, E. and Hasan, W. 2018. Study of accumulators of several aquatic plants in absorbing heavy metals by phytoremediation. *Jurnal Serambi Engeneering* 3(2):344-351 (*in Indonesian*).

Jeffries, P., Gianinazzi, S., Perotto, S., Turnau, K. and Barea, J.-M. 2003. The contribution of arbuscular mycorrhizal fungi in sustainable maintenance of plant health and soil fertility. *Biology and Fertility of Soils* 37:1-16, doi:10.1007/s00374-002-0546-5.

Komárek M., Vanek, A. and Ettler, V. 2013. Chemical stabilization of metals and arsenic in contaminated soils using oxides -a review. *Journal of Environmental Pollution* 172:9-22, doi:10.1016/j.envpol.2012.07.045.

Kusuma, M.E. 2018. Response of *Brachiaria decumbens* grass to the application of biochar and organic fertilizer on sandy soil. *Jurnal Ilmu Hewani Tropika* 7(2):33-38 (*in Indonesian*).

Laoli, B.M.S, Kisworo and Raharjo, D. 2021. Accumulation of chromium (Cr) pollutants in rice plants along the Opak River Basin, Bantul Regency. *Jurnal Biospecies* 14(1):59-66 (*in Indonesian*).

Leelarungrayub, N., Rattanapanone, V., Chanarat, N. and Gebicki, J.M. 2006. Quantitative evaluation of the antioxidant properties of garlic and shallot preparations. *Nutrition* 22: 266-275, doi:10.1016/j.nut.2005.05.010.

Lehmann, J. and Joseph, S. 2009. Biochar for Environmental Management. In: Lehmann, J. and Joseph, S. (eds), *Science and Technology*. Earthscan-UK. pp. 71-78.

Liu, D. and Kottke, I. 2003. Subcellular localization of chromium and nickel in root cells of *Allium cepa* by EELS and ESI. *Cell Biology and Toxicology* 19:299-311, doi:10.1023/B:CBTO.0000004984.87619.15.

Miskiyah and Mnarso, S.J. 2009. Contamination of pesticide residues in red chili, lettuce, and shallots (case studies in Bandungan and Brebes, Central Java and Cianjur, West Java). *Jurnal Hortikultura* 19(1):101-111 (*in Indonesian*).

Mohammadi-Motlagh, H.R., Mostafaie, A. and Mansouri, K. 2011. Anticancer and anti-inflammatory activities of shallot (*Allium ascalonicum*) extract. *Archives of Medical Science* 7(1):38-44, doi:10.21082/jp3.v29n4.2010.p154-158.

Naher, U.A., Othman, R. and Panhwar, Q.A. 2013. Beneficial effects of mycorrhizal association for crop production in the tropics - a review. *International Journal of Agriculture and Biology* 15(5):1021-1028.

Nurida, N.L. 2014. Potential use of biochar to rehabilitation of upland in Indonesia. *Jurnal Sumberdaya Lahan* 8(3):57-68 (*in Indonesian*).

Oni, B.A, Oziegbe, O. and Olawole, O.O. 2019 Significance of biochar application to the environment and economy, *Annals of Agricultural Sciences* 64(2):222-236, doi:10.1016/j.aoas.2019.12.006.

Owoyele, B.V., Alabio, T., Adebayo, J.O., Soladoye, A.O., Abioye, A.I. and Jimoh, S.A. 2004. Haematological evaluation of ethanolic extract of Allium ascalonicum in male albino rats. *Fitoterapia* 75:322-330, doi:10.1016/j.fitote.2004.02.006.

Permanasari, I., Kartika, D., Irfan, M. and Ahmad, T.A. 2016. Increased efficiency of phosphate fertilizers through the application of mycorrhizae on soybeans. *Jurnal Agroteknologi* 6(2):23-30, doi:10.24014/ja.v6i2.2237 (*in Indonesian*).

Plenchette, C., Clermont-Dauphin, C, Meynard, J.M. and Fortin, J.A. 2005. Managing arbuscular mycorrhizal fungi in cropping systems. *Canadian Journal of Plant Science* 85:31-40, doi:10.4141/P03-159

Puga, A.P., Abreu, C.A., Melo, L.C.A. and Beesley, L. 2015. Biochar application to contaminated soil reduces the availability and plant uptake of zinc, lead and cadmium. *Journal of Environmental Management* 159:86-93, doi:10.1016/j.jenvman.2015.05.036.

Puspita U.R., Siregar, A.S. and Hidayati, N.V. 2011. The ability of aquatic plants as phytoremediator agents of heavy metal chromium (Cr) found in the liquid waste of the batik. *Penelitian Berkala Perikanan Terubuk* 39(1):60-63 (*in Indonesian*).

Putri, V.C., Mukhlis, M. and Benny H. 2017. Provision of several types of biochar to improve the chemical properties of ultisol soil and the growth of corn plants. *Jurnal Agroteknologi* 5(4):824-828 (*in Indonesian*).

Ratnasari, I.F.D., Hadi, S.N., Suparto, S.R., Herliana, O. and Ahadiyat, Y.R. 2020. Phytoremediation of cadmium-contaminated soil using terrestrial kale (*Ipomoea reptans* Poir) and corncob biochar. *Journal of Degraded and Mining Lands Management* 7(4):2313-2318, doi:10.15243/jdmlm.2020.074.2313.

Riaz, M., Kamran, M., Fang, Y., Wang, Q., Cao, H., Yang, G., Deng, L., Wang, Y., Zhou, Y., Anastopoulos, I. and Wang, X. 2021. Arbuscular mycorrhizal fungi-induced mitigation of heavy metal phytotoxicity in metal contaminated soils: A critical review. *Journal of Hazardous Materials* 402, doi:10.1016/j.jhazmat.2020.123919.

Riri, R.C., Guchi, H. and Rauf, A. 2013. Bioremediation of Cd, Cu, and Pb polluted soils using endo mycorrhizae. *Jurnal Online Agroekoteknologi* 2(1):348-361 (*in Indonesian*).

Rokhminarsi, E., Darini S.U. and Begananda. 2019. The effectiveness of mycorrhizal biofertilizers based on Azolla (mycola) on shallots. *Jurnal Hortikultura* 29(1):45-52, doi:10.21082/jhort.v29n1.2019.p45-52 (*in Indonesian*).

Saleh, S., Anshary, A., Made, U., Mahfudz, and Basir-Cyio, M. 2021. Application of mycorrhizae and Beauveria in organic farming systems effectively control leaf miners and enhance shallot production. *AGRIVITA Journal of Agricultural Science* 43(1):79-88, doi:10.17503/agrivita.v1i1.2831.

Saraswati, R., Husen, E. and Simanungkalit, R.D.M. 2007. *Soil Biological Analysis Method.* Center for Research and Development of Agricultural Land Resources, Ministry of Agriculture, Indonesia (*in Indonesian*).

Sasli, I.and Ruliansyah, A. 2012. Utilization of Location-Specific Arbuscular Mycorrhizae for Fertilization Efficiency in Corn Plants in Tropical Peatlands. *Agrovigor: Jurnal Agroekoteknologi* 5(2):65-74, doi:10.21107/agrovigor.v5i2.310 (*in Indonesian*).

Satriawan, B.D. and Handayanto, E. 2015. Effect of biochar and crop residue application on chemical properties of a degraded soil of South Malang, and P uptake by maize. *Journal of Degraded and Mining Lands* 2(2):271-281.

Shekhawat, K., Sreemoyee Chatterjee, S. and Joshi, B. 2015. Chromium toxicity and its health hazards. *International Journal of Advanced Research* 3(7):167-172.

Shenbagavalli, S. and Mahimairaja, S. 2012. Production and characterization of biochar from different biological wastes. *International Journal of Plant, Animal, and Environmental Sciences* 2(1):197-201.

Shofiyanto, M.E. 2008. *Hydrolysis of Corn Cobs by Cellulolytic Bacteria for Bioethanol Production in Mixed Culture*. Faculty of Agricultural Technology, IPB. Bogor (*in Indonesian*).

Suharno and Sancayaningsih, P. 2013. Arbuscular mycorrhizal fungi: the potential of heavy metal mycorrhizoremediation technology in mine site rehabilitation. *Jurnal Bioteknologi* 10(1):37-48 (*in Indonesian*).

Stoica, C., Dinu, L., Lucaciu, I., Nita-Lazar, M. and Oncu, V. 2020. The toxic effect of conventionally treated mine water on aquatic organisms. *Revista de Chimie* 71:67-71, doi:10.37358/RC.20.1.7813.

Tasneem, A., Kafeel, A., Zafar, I.K., Zunaira, M., Ahlam, K., Al-Qthanin, R., Alsubeie, M., Alamri, S., Hashem, M., Farooq, S., Maqboolm M.M., Hashim, S. and Wang, Y. 2021. Chromium accumulation in soil, water and forage samples in automobile emission areas. Saudi *Journal of Biological Sciences* 28(6):3517-3522, doi:10.1016/j.sjbs.2021.03.020.

Thakur, R., Sharma, G.D., Dwivedi, S. and Khatik, S.K. 2007. Chromium: as a pollutant. *Journal of Industrial Pollution Control* 23(2):197-203.

Unver, M.C., Ugulu, I., Durkan, N., Baslar, S. and Dogan, Y. 2015. Heavy metal contents of *Malva sylvestris* sold as edible greens in the local markets of Izmir. *Ekoloji* 24:13-25, doi:10.5053/ekoloji.2015.01.

Vance, P.C., Uhde-Stone, C. and Allan, D.L. 2002. Phosphorus acquisition and use: critical adaptations by plants for securing a non renewable resource. *New Phythologist* 157:423-447, doi:10.1046/j.1469-8137.2003.00695.x.

Yang, Y., Khan, Z.I., Ahmad, K., Arshad, N., Rehman, S.U., Ullah, M.F., Wajid, K., Mahpara, S., Bashir, H. and Nadeem, M. 2020. Does the chromium element in forages and fodders grown in contaminated pasture lands cause toxicity in livestock: assessing the potential risk. *Revista de Chimie* 71:397-405, doi:10.37358/RC.20.7.8257.

Yin, M.C, and Tsao, S.M. 1999. Inhibitory effect of seven Allium plants upon three Aspergillus species. *International Journal Food Microbiology* (49):49-56, doi:10.1016/S0168-1605(99)00061-6.