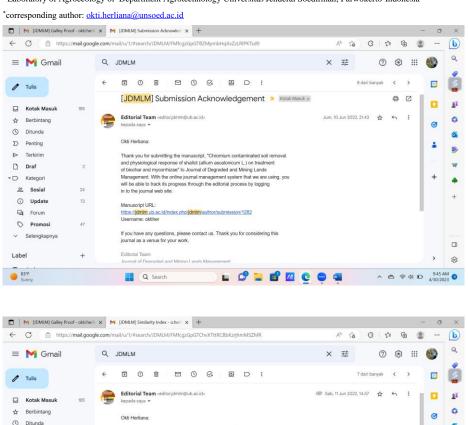
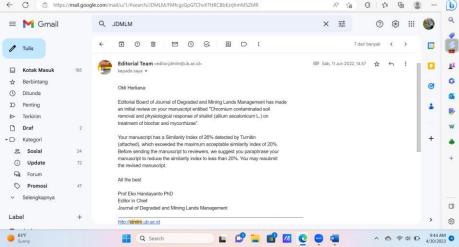
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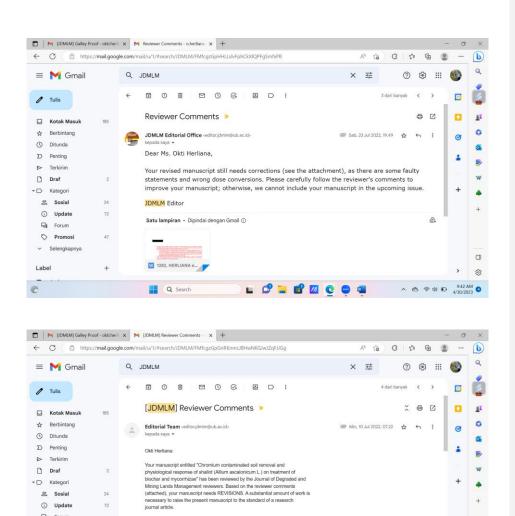
Removal of chromium from chromium-contaminated soil and physiological response of shallot ($allium\ ascalonicum\ L.$) on treatments of biochar and mycorrhizae

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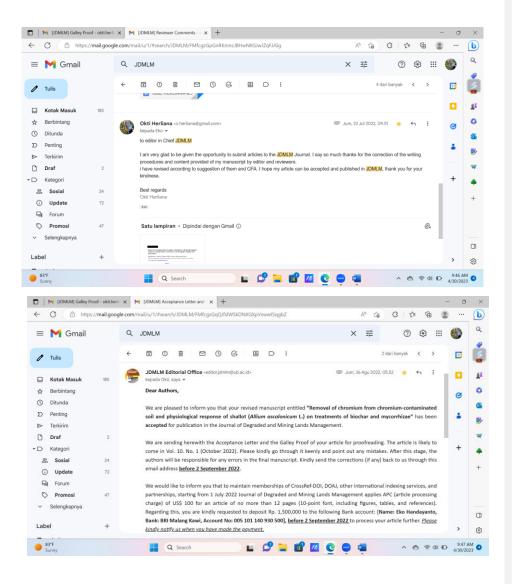
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Prof Eko Handayanto PhD



Research Article

- Polybag is not a standard weight or area unit, so the mycorrhiza dose of 1 g polybag⁻¹ should be converted to the
 mycorrhiza dose of 210 kg ha⁻¹, or 0.21 t ha⁻¹, and the mycorrhiza dose of 2 g polybag⁻¹ should be converted to
 the mycorrhiza dose of 420 kg ha⁻¹, or 0.42 t ha⁻¹.
 - The weight of soil from 1 ha of land is about 2,100,000 kg (20 cm depth; bulk density of 1.1 g cm⁻³), in your manuscript, 1 polybag contains 10 kg of soil, → then the mycorrhiza dose of 1 g polybag⁻¹ (on in another word, 1 g 10 kg⁻¹ of soil) is equivalent to the mycorrhiza dose of 210 kg ha⁻¹, or 0.21 t ha⁻¹, and the mycorrhiza dose of 2 g polybag⁻¹ (on in another word, 2 g 10 kg⁻¹ of soil) is equivalent to the mycorrhiza dose of 420 kg ha⁻¹, or 0.42 t ha⁻¹

- 2. In the Abstract of your manuscript, you wrote the biochar dosages of 0 t ha⁻¹, 2.5 t ha⁻¹, 5 t ha⁻¹, 5 t ha⁻¹, and 10 t ha⁻¹, but in the Method section, you wrote the biochar doses of B1 = 45 g polybag⁻¹, B2 = 90 g polybag⁻¹ and B3 = 180 g polybag⁻¹. Please calculate again.
 - The weight of soil from 1 ha of land is about 2,100,000 kg (20 cm depth; bulk density of 1.1 g cm⁻³); in your manuscript, 1 polybag contains 10 kg of soil → then (a) the dose of 2.5 t ha⁻¹ is equivalent to 2,500 kg 2,100,000 kg⁻¹ of soil, or 12 g 10 kg⁻¹ of soil, or 12 g polybag⁻¹ (not 45 g polybag⁻¹, as this is equivalent to 9.5 t ha⁻¹), (b) the dose of 5 t ha⁻¹ is equivalent to 5000 kg 2,100,000 kg⁻¹ of soil, or 24 g 10 kg⁻¹ of soil, or 24 g polybag⁻¹ (not 90 g polybag⁻¹, as this is equivalent to 19 t ha⁻¹), and (c) the dose of 10 t ha⁻¹ is equivalent to 10000 kg 2,100,000 kg⁻¹ of soil, or 48 g 10 kg⁻¹ of soil, or 48 g polybag⁻¹ (not 180 g polybag⁻¹, as this is equivalent to 38 t ha⁻¹).

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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Article history:

Received Day Month 20xx Accepted Day Month 20xx Published Day Month 20xx

Keywords:

shallot; physiological character; chromium; biochar; mychorriza

Abstract

The food safety and soil degradation were the reasons to treat pollouted soil. Shallots are high value commodities so that cultivation is carried out intensively. Continuous use of agrochemicals can caused heavy metal contamination. This study aimed to investigate chromuium removal, physiological characters and yield of shallot (Allium ascalonicum L.) on biochar and mycorrhiza application on the chromium contaminated soil. The pot experiment was conducted from April to September 2019, at screen house ex-farm of the Faculty of Agriculture, Jenderal Soedirman University. The treatments tested consisted of two factors. The first factor was mycorrhiza dosages of 0 g polybag-1, 1 g polybag-1 and 2 g polybag-1, and the second factor was biochar dosages of 0 t ha⁻¹, 2.5 t ha⁻¹, 5 t ha⁻¹ and 10 t ha⁻¹. The twelve treatments were arranged in a factorial, completely randomized block design with three replications. The results showed that application of biochar 2.5 t ha⁻¹, 5 t ha⁻¹ and 10 t ha⁻¹ had been able to increase plant height and percentage of root infection. The application of mycorrhiza 1 g polybag-1 and 2 g polybag-1 had been able to increase plant height, percentage of root infection and plant tissue P uptake. The both applications of biochar and mycorrhiza had been able to increase plant height and percentage of root infection.

To cite this article: Herliana, O. Ahadiyat, Y.R. Cahyani, W. Anwar, A.H.S. 2022. Removal of chromium from chromium-contaminated soil and physiological response of shallot (*Allium ascalonicum* L.) on treatments biochar and mycorrhizae. Journal of Degraded and Mining Lands Management 9(0):0000-0000, doi:10.15243/jdmlm. 2022.000.0000.

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 (Saleh and Atmaja, 2017). Shallot cultivation is prone to failure due to environmental factors and pests, so the use of fertilizers and pesticides is massive. Increased public awareness aspects

Commented [Ir1]: PLEASE convert the unit of g/polybag to kg/ha→ polybag is not a standard unit. 1 ha is about 2,100,000 kg soil (20 cm depth, bulk density of 1.1 g/cm²), if 1 polybag contain: 10 kg soil, then the dose of 1 g/polybag is equivalent to 210 kg mycornhiza/ha, or 0.21 t/ha

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 that 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 (Mulyadi, 2013). 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. The threshold for Cr in the soil according to SNI is 5 ppm (Puspita et al., 2011). According to Bruce (2002), the soil is considered polluted by Cr if the concentration is above 10 ppm in the soil. The increasing Cr piles 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 (Susanti et al., 2018). Consumption of chromium that can be tolerated by the human body is a maximum of 0.07 ppm per day (Nanik, 2008). The accumulation of Cr in the human body can cause damage to the respiratory organs and cause cancer in humans (Palar, 1994).

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 °C (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 °C, so that agricultural waste can be useful for remediation of contaminated soil

Mycorrhizae have been reported to be potential for remediating heavy metalcontaminated soils (Riri et al., 2011). 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 Glomus genus associated with plants are effective in absorbing heavy metals, such as Cd, Zn, and Pb (Musfal, 2010). Shallots are the main kitchen spices, so the demand is quite high, effort to reduce heavy metal residue on this commodity is interesting issue to be studied. This research was aimed to investigate chromuium removal, physiological characters and yield of shallot (Allium ascalonicum L.) upon biochar and mycorrhiza 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 soil taken from shallot cultivation Incepticol soil type obtained in Songgom Village, Songgom District, Brebes Regency with location maps was - 7.019176926846719, 108.982348621595, pH of 5.27 and Cr content of 18.78 ppm. According to the SNI of Indonesia, this Cr content is above the maximum residual limit of Chromium, 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. According to Saraswati et al (2007), soil sampling can be done using a composite method that aims to obtain soil in a relatively homogeneous area.

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 °C 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 is 110 m above sea level, the average daily temperature is 27.5 °C, 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 10 days after planting. During the experiment, 500 mL of water was supplied everyday.

Experimental design

The treatments tested consisted of two factors. The first factor was biochar dosage (B) consisting of 4 levels, i.e., B0 = without biochar, B1 = 45 g biochar polybag⁻¹, B2 = 90 g biochar polybag⁻¹ and B3 = 10 biochar g polybag⁻¹. The second factor was mycorrhizae inoculation consisting of 3 levels, i.e., M0 = without mycorrhizae, M1 = 1 g mycorrhizae polybag⁻¹. The twelve treatments were arranged a randomized bock design with three replications

Variables observed

Commented [Ir2]: Please calculate again. 1 ha of topsoil (20 cm depth; bulk density of 1.1 g/cm²) is about 2,100,000 kg of soil. If one 1 polybag contains 10 kg soil, then the dose of 2.5 t/ha should be equivalent to 2,500 kg/2,100,000 kg soil, or 12 g/10 kg soil, or 12 g/polybag (not 45 g/polybag) → 45 g/ha is equivalent to 9.5 t/ha

Commented [Ir3]: PLEASE convert the unit of g/polybag to kg/ha → polybag is not a standard unit. 1 ha is about 2,100,000 kg soil (20 cm depth; bulk density of 1.1 g/cm²), if 1 polybag contains 10 kg soil, then the dose of 1 g/polybag is equivalent to 210 kg mycorrhiza/ha, or 0.21 t/ha

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.

Cr plant absorption and Cr soil removal

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 following formula (Hardiani et al., 2009),

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

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 heavy metal Cr are shown in

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

No	Observed Variable —	H	Effect			
110	Observed variable —	В	M	B x M		
1.	Plant height	S	S	S		
2.	Total Root Length	ns	ns	ns		
3.	Root Dry Weight	ns	ns	ns		
4	Crop Growth Rate	ns	ns	ns		
5	Net Assimilation Ratio	ns	ns	ns		
6	Leaf Chlorophyll Content	ns	ns	ns		
7	Percentage of Root Infection	S	s	S		
8	P uptake by Plant Tissue	ns	s	ns		
9	Chromium content in plant tissue	S	s	ns		
10	Chromium Removal from Soil	s	s	ns		
11	Number of tubers per clump	ns	ns	S		
12	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

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.

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

Biochar	SL	sw	CGR	NAR	CC	P uptake	Cr PT	Cr R
Treatment	(cm)	(g)	(g day ⁻¹)	(g cm ⁻² day ⁻¹)	(mg L ⁻¹)	(ppm)	(ppm)	(%)
0 g polybag ⁻¹	307.29	0.07	0.071	0.04	12.72	139.45	1.21 a	26.97a
45 g polybag ⁻¹	296.76	0.06	0.106	0.02	12.55	179.27	1.07 b	35.82a
90 g polybag ⁻¹	293.34	0.07	0.145	0.029	12.66	155.47	0.96 bc	50.77b

180g polybag	334.04	0.06	0.09	0.058	12.48	162.82	0.94 bc	53.34b
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.

Results of the analysis of variance showed that the application of biochar had no significant effect on plant growth and physiological aspects such as root length, plant growth rate, net assimilation rate, leaf chlorophyll content, and plant tissue P uptake. This indicates that shallot plants can adapt to media contaminated with chromium. According to Cobbett (2000), plant cells respond to heavy metal stress using various defense mechanisms, including exclusion, immobilization, chelation, and compartmentalization of metal ions. According to Liu and Konteke (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 45 g biochar polybag¹, which was 1.07 ppm. The addition of 90 g biochar polybag¹ and 180 g biochar polybag¹ was not significantly different from the application of 45 g biochar polybag¹, 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. Results of this experiment showed that the application of 90 g biochar polybag¹ and 180 g biochar polybag¹ 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 a.l, 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, particularly immobile elements 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 Tn table 3.

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

Biochar Treatment	SL (cm)	SW (g)	CGR (g day ⁻¹)	NAR (g cm ⁻² day ⁻¹)	CC (mg L ⁻¹)	P uptake	Cr PT (ppm)	Cr R (%)
0 g polybag ⁻¹	295.4	0.07	0.096	0.048	13.12	132.48 a	1.17 a	23.76a
1 g polybag ⁻¹	292.52	0.06	0.085	0.044	11.84	167.68 b	0.79 b	47.62b
2 g polybag ⁻¹	335.65	0.07	0.129	0.018	12.8	177.25b	0.71 b	56.25c
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 1 g mycorrhizae polybag⁻¹ and 2 g polybag⁻¹ 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 1 g mycorrhizae polybag⁻¹ and 2 g mycorrhizae polybag⁻¹ 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 mycorrhiza, 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 contaminant in soil; application of 2 g mycorrhizae polybag⁻¹ showed the best treatment, which can reduce

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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 so as 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 absorption of substances that are toxic to plants (Suharno and Sancayaningsih, 2013).

Effect of the interaction between mycorrhiza 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 the height of the plants treated with 1 g mycorrhizal polybag⁻¹ with 45 g biochar polybag⁻¹, 90 g biochar polybag⁻¹, 180 g biochar polybag⁻¹and 2 g mycorrhiza polybag⁻¹ with 45 g biochar polybag⁻¹, 90 g biochar polybag⁻¹. 180 g biochar polybag⁻¹did not show a significant difference with a plant height range of 33.57 cm - 38.07 cm. However, the treatment was significantly different from the treatment without mycorrhizal with 10 t biochar ha⁻¹ and 1 g mycorrhizal polybag⁻¹ without biochar which resulted in plant heights of 32.62 cm and 32.66 cm. Treatments without mycorrhizal and with 45 g biochar polybag⁻¹ and 90 g biochar polybag⁻¹ and 2 g mycorrhizal polybag⁻¹ with treatment without biochar showed no significant difference with the plant height range of 29.78 cm - 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.

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

		Variable				
Mycorrhiza dose	Biochar Dosage	Plant height (cm)	Root Infection Percentage (%)	Number of Tuber	Weight of Tuber	
	0 g polybag ⁻¹	26.81 d	18.56 d	5.00 c	18.56 d	
without	45 g polybag ⁻¹	29.78 с	24.31 b	6.41 b	24.31 b	
mycorrhizae	90 g polybag ⁻¹	31.78 с	28.40 a	7.28 b	28.40 a	
	180g polybag ⁻¹	32.62 b	29.76 a	9.50 a	29.76 a	
	0 g polybag ⁻¹	32.66 b	22.22 c	7.33 b	22.22 c	
1 g polybag ⁻¹	45 g polybag ⁻¹	34.17 a	27.03 a	7.72 a	27.03 a	
i g polybag	90 g polybag ⁻¹	33.97 a	28.06 a	8.72 a	28.06 a	
	180g polybag ⁻¹	38.07 a	29.61 a	8.94 a	29.61 a	
	0 g polybag ⁻¹	31.20 с	25.03 a	7.00 b	25.03 a	
2 g polybag ⁻¹	45 g polybag ⁻¹	33.57 a	26.76 a	8.25 a	26.76 a	
2 g polyoag	90 g polybag ⁻¹	34.65 a	27.69 a	7.42 b	27.69 a	
	180g polybag ⁻¹	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.

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 elements 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 results, and the results increased, ranging from 18.56-29.76 g. The treatment of 1 g mycorrhizae polybag-1 with biochar had a significant effect. The total weight increased, ranging from 22.62 – 29.61 cm. However, the treatment of mycorrhizal 1 g polybag⁻¹ on biochar 45, 90, and 180 g polybag-1 did not have a significant effect, and the treatment without biochar got the lowest results. Mycorrhizal treatment of 2 g/polybag 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 90 g polybag-1 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 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. According to Satriawan and Handayanto (2015), plants need P for root development, accelerating flowering and maturation, as well as root and seed formation. 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.

Conclusions

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 5 t biochar ha-1 could reduce the accumulation of chromium in shallot plant tissues by 20.66% and remove soil chromium content by 50.77%. Mycorrhiza 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 45 biochar g-1 and 1 g mycorrhizae polybag⁻¹ could increase plant height and percentage of root infection, number of tubers, and tuber weight of shallot Acknowledgements

The authors thank the LPPM of Universitas Jenderal Soedirman for funding this research in the competency improvement scheme year of 2019, the Balingtan Ministry of Agriculture Pati. The kind help of Dianawati, Anggraria Dyah, M. Minanur, and Gilang Aji to carry out this research is highly appreciated

References → there are TOO MANY LOCAL/DOMESTIC REFERENCES, please replace them with articles from international journals

- 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
- Audet P. and Charest C. 2006. Effects of AM colonization on "wild tobacco" plants grown in zinc-contaminated soil. Mycorrhiza 16: 277-283
- 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, https://doi.org/10.1104/pp.123.3.825
- Ferreiro, J., Gascó G, Méndez, A and Reichman, S.M. 2018 Soil Pollution and Remediation Internatonal 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 (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. https://doi.org/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
- 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.
- 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
- 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)
- Lehmann, J. and Joseph, S. 2009. Biochar for Environmental Management: Journal of 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
- 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)
- Mulyadi. 2013. Pb heavy metal in rice fields and grain in Juwana Sub-watershed, Central Java. *Jurnal Agrologi*.2(2): 95-101 (in Indonesian)
- Musfal, M. 2017. Potential of Arbuscular mycorrhizal fungi to increase corn crop. *Jurnal Penelitian Dan Pengembangan Pertanian* 29(4):154-158. (in Indonesian) doi: http://dx.doi.org/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.
- Nanik. H.S. 2008. Chromium content in agriculture, sediment and blood shellfish (*Andara granosa*) in the Coastal area around the estuary of the Sayung River, Morosari Village, Demak Regency, Central Java.. *Jurnal Bioma* 10 (2): 53-56. (in Indonesian)
- 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, https://doi.org/10.1016/j.aoas.2019.12.006.
- Palar, H. 1994. Heavy Metal Pollution and Toxicology. Jakarta: PT Rineka Cipta. (in Indonesian)
- 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. (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.
- 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.
- 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 liquid waste of the batik. *Penelitian Berkala Perikanan Terubuk*. 39(1): 60-63. (in Indonesian)
- Putri, V.C., Mukhlis., 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* FP USU, 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.
- Riri, R.C., Guchi, H. and Rauf, A. 2013. Bioremediation of Cd, Cu, and Pb polluted soils using endomycorrhizae. Jurnal Online Agroekoteknologi 2(1): 348-361, (in Indonesian)
- Rokhminarsi, E., Darini S.U. and Begananda. 2019. The effectiveness of mycorrhizal biofertilizer based on azolla (mycola) on shallots. *Jurnal Hortikultura*, 29(1): 45-52. (in Indonesian)
- Saleh, I and Atmaja, I.S.W. 2017. Effectiveness of Arbuscular Mycorrhizal Fungi (AMF) inoculation on shallot production with different watering techniques. *Jurnal Hortikultura Indonesia*, 8(2): 120-127 (in Indonesian)
- Saleh, S., Anshary, A., Made, U., Mahfudz, and Basir-Cyio, M. 2021. Application of Mycorrhizae and Beauveria in organic farming system effectively control leafminers and enhance shallot production. AGRIVITA Journal of Agricultural Science, 43(1): 79–88. https://doi.org/10.17503/agrivita.v1i1.2831
- Saraswati, R., Husen, E., and Simanungkalit, R.D.M. 2007. Soil Biological Analysis Method. Balai Besar Litbang Sumberdaya Lahan Pertanian, BPPP Departemen Pertanian. (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:https://doi.org/10.21107/agrovigor.v5i2.310

- Satriawan, B.D. and Handayanto E. 2015. Effect of biochar and crop residues 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 Bhumika 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*. Fakultas Teknologi Pertanian IPB. Bogor. (in Indonesian)
- Suharno and Sancayaningsih P, 2013. Arbuscular Mycorrhizal Fungi: Potential of heavy metal mycorrhizoremediation technology in mine site rehabilitation. *Jurnal Bioteknologi* 10 (1): 37-48. (in Indonesian)
- Susanti, A., Rohmat H., and Hari P. 2018. Implementation of mycorrhizae as a means of knowledge on independent conservation of marginal land in Kabuh District, Jombang. *Jurnal Agroradix* 1(2): 9-17 (in Indonesia)
- 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.