Magna Scientia Advanced Biology and Pharmacy

eISSN: 2582-8363

Cross Ref DOI: 10.30574/msabp

Journal homepage: https://magnascientiapub.com/journals/msabp/

(Research Article)

Effect of biochar type on planting media contaminated with heavy metals Pb, Cr, and Hg on growth and yield of rice (*Oryza Sativa* L.) as well as Bioaccumulation Factor and Translocation Factor

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Magna Scientia Advanced Biology and Pharmacy, 2025, 14(02), 034-042

Publication history: Received on 12 January 2025; revised on 26 February 2025; accepted on 01 March 2025

Article DOI: https://doi.org/10.30574/msabp.2025.14.2.0024

Abstract

Soil pollution by heavy metals such as lead (Pb), chromium (Cr), and mercury (Hg) can reduce soil quality and crop productivity, including rice (*Oryza sativa* L.). This study aimed to evaluate the effect of three types of biochar - rice straw and husk biochar (Br), fruit and vegetable waste biochar (Bw), and lignohumic biochar (Bl) - on rice growth, yield, and Bioaccumulation Factor BAF) and Translocation Factor (TF) values. The research was conducted in heavy metal contaminated planting media using a Randomized Group Design (RAK). The results showed that biochar effectively reduced heavy metal levels in soil and increased rice growth and yield. Coir and coconut shell biochar (Bc) showed the lowest BAF and TF values, thus reducing the risk of heavy metal contamination in rice seeds. This study indicates the potential of biochar as an ecological solution for the management of heavy metal contaminated agricultural land.

Keywords: Rice; Heavy metals; Biochar; BAF; TF

1. Introduction

Rice (*Oryza sativa* L.) is the main food crop in Indonesia and the main source of carbohydrates for most people. However, the sustainability of rice production is faced with various challenges, including a decline in soil quality due to heavy metal pollution such as lead (Pb), chromium (Cr), and mercury (Hg). This pollution is mainly caused by industrial waste, mining activities, and the use of fertilizers and pesticides containing heavy metals [5] [6].

The presence of heavy metals in soil not only disrupts soil fertility, but also affects plant productivity and crop quality. Heavy metals can accumulate in soil and plants through root absorption, causing physiological disorders such as decreased enzyme activity, impaired photosynthesis, and reduced plant growth and yield[7]. Heavy metal accumulation in rice plants also poses a risk to human health, especially through the consumption of contaminated rice grains[8].

To overcome this problem, various efforts have been made, including the utilization of organic materials to improve soil quality. One promising solution is the use of biochar, which is activated carbon produced through pyrolysis of organic biomass [9]. Biochar is not only able to improve soil structure and fertility, but is also effective in binding heavy metals.

Thus reducing its availability to plants [10]. Previous research shows that biochar has the ability to increase crop yields and reduce heavy metal levels in soil and plants [11].

Various types of biochar have been evaluated based on their raw materials, such as coconut husk and shell, fruit and vegetable waste, and lignohumic from wood waste. Each type of biochar has unique characteristics that affect its effectiveness in addressing heavy metal pollution and supporting plant growth. However, research related to the effect

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of various types of biochar on important parameters such as Bioaccumulation Factor (BAF) and Translocation Factor (TF) in rice is still limited [12].Therefore, this study was conducted to evaluate the Effect of Biochar Types on Planting Media Contaminated with Heavy Metals Pb, Cr, and Hg on Growth and Yield of Rice (*Oryza Sativa* L.) as well as Bioaccumulation Factor and Translocation Factor. The results of this study are expected to provide ecological and practical solutions to increase the productivity of rice plants in heavy metal contaminated land.

2. Material and methods

2.1. Place and Time of Research

The research was conducted at the Experimental Garden of the Faculty of Agriculture, Udayana University, Pegok, Denpasar. Then further studied in the Agronomy and Horticulture Lab of Udayana University from May to October 2024. This study used Randomized Group Design (RAK) 3 levels of biochar types with 9 replications. The treatment was the type of biochar (B), which consisted of Br: Biochar of rice straw and husk, Bw: Fruit and vegetable waste biochar, Bl: Lignohumic biochar from wood waste.

2.2. Biochar production

Biochar from different raw materials for straw husk biochar (Br) and vegetable fruit biochar (Bw) are made according to the method of Wedayani et al., (2023), while Lignohumic biochar is a biochar that is already available on the market and obtained from Russia under the trade name Lignohumate. Biochar Br and Bw are made by cutting / chopping small pieces of raw material and then dried by drying in the sun until completely . After that, biochar is made by pyrolyzing at 400°C. The pyrolysis process is carried out manually using the pyrolysis method in stages. The charcoal produced from the pyrolysis process is then pulverized by pounding using lumping or using a blender according to the condition of the raw material. The smooth charcoal is then activated using CaCl2 solution so that it becomes biochar. Biochar that is ready is then analyzed using the Scanning Electron Microscope (SEM) test to determine its physico-chemical characteristics, as well as proximate tests (water content, ash content, volatile content and carbon content in biochar). SEM test was conducted on each type of biochar using a voltage of 5.0 kV, an average working distance of 10.5 mm, a current probe of 30 and varying magnification at MERO Foundation. Proximate tests were conducted using ASTM D7582 MVA at the Engineering Laboratory, Faculty of Engineering, Udayana University.

2.3. Seed sowing

Rice seeds before sowing are soaked for 48 hours in a 500 ml water solution that has been mixed with 3 cloves of onion and monosodium glutamate (MSG). The soaked seeds were then drained and tended for 2 days. After maturing, the seeds were sown in a nursery box for 21 days.

2.4. Media preparation

Preparation of planting media for rice plants begins with drying the soil from 5 subak in the downstream Badung river that has been composited, then the dried soil is mashed and sieved. The soil that has been finely then weighed as much as 7 kg and then put into plastic pots. The plastic pots used are 7 kg in size as many as 27 pieces. After that, it was evenly mixed with biochar as much as 52.5 grams per 7 kg of soil in accordance with the recommended use of biochar for rice which is 10-15 tons/ha. After that, a code was given according to the treatment of biochar types with different raw materials. Planting media that are given biochar and given a treatment code are watered with water to field capacity and ready for planting. The planting media was planted with rice seedlings as many as 3 seedlings per plastic pot.

2.5. Maintenance

Rice plants are intensively maintained. Maintenance includes fertilization, watering, soil loosening, pest and disease control, and others to ensure optimal rice growth and yield. Fertilization is carried out according to the recommendations for fertilizing rice plants. The recommended fertilizer dose for rice plants based on MOA No. 22 of 2022 is a single fertilizer dose package of Urea, TSP and KCl at a dose of 300 kg Urea/ha, 50 kg TSP/ha, and 50 kg KCL/ha respectively, or a dose package of Phonska Plus 15-15-15 compound fertilizer at a dose of 400 kg/ha and Urea at a dose of 100 kg/ha. Harvesting is done when the rice seeds are physiologically ripe, characterized by yellowing of the rice grain.

3. Observation Variable

3.1. Results of Analysis of Heavy Metal Content of Pb, Cu and Cd in Soil, Irrigation Water and Rice Plant Organs from Subak in the Lower Badung River

Phase I of the research was carried out surveys and sampling of soil, irrigation water and rice plants in Subak-subak which utilizes the downstream Badung River water as the source of irrigation. The samples were then analyzed for detection. metal content of lead (Pb), cadmium (Cd) and copper (Cu). Testing of heavy metal content was conducted at the Analytical Laboratory of Udayana University.

3.2. Results of Analysis of Heavy Metal Content of Pb, Cd and Cu in Soil and Plant Organs after Biochar Treatment and Heavy Metal Injection

Measurement of heavy metal content in soil, roots, stover and seeds after biochar treatment aims to evaluate the accumulation and distribution of heavy metals and the effectiveness of biochar in reducing pollution during the study.

3.3. Results of the Effect of Biochar Treatment and Heavy Metal Injection on Rice Growth and Yield

3.3.1. Plant height

Plant height was observed every 2 weeks until maximum height. The first observation was 2 weeks after the seedlings were planted (MSBD).

3.3.2. Total grain weight per clump (g)

Total grain weight per clump is the total weight of grain produced by one clump of rice plants, including filled grain (which contains seeds) and empty grain (which does not contain seeds). This parameter is used to measure the productivity of rice plants in a clump as an indicator of yield.

3.3.3. Grain yield at 12% moisture content (g)

Yield of 12% moisture content grain per pot is measured after drying 12% moisture content grain or can be converted from harvested dry grain yield using the formula.

3.4. Bioaccumulation factor (BAF)

$$Bioaccumulation\ factor\ of\ rice\ plants\ to\ heavy\ metals\ Pb, Cr, and\ Hg. Can\ be\ calculated\ by\ formula\ BAF = \frac{Plant\ Heavy\ Metal\ Content}{Soil\ Heavy\ Metal\ Content}$$

3.5. Translocation factor (TF)

$$Translocation\ factors\ of\ rice\ plants\ to\ heavy\ metals\ Pb, Cr, and\ Hg. Available\ at\ calculated\ with)\ formula: TF) = \frac{\text{Heavy Metal Content of stems or seeds}}{Root\ Metal\ Content}$$

4. Results and discussion

4.1. Results of Analysis of Heavy Metal Content of Pb, Cr and Hg in Soil, Irrigation Water and Rice Plant Organs from Subak in the Lower Badung River

The test results of heavy metal content of Pb, Cr, and Hg showed that the highest heavy metal content was found in the soil, namely Pb with an average of 75.43 mg/kg, and the lowest metal content in the soil was Cr with an average of 5.73 mg/kg. In the roots of rice plants, the heavy metal with the highest concentration was Cr with an average of 28.94 mg/kg, and the heavy metal content of Hg was not detected in the soil.

Table 1 Heavy Metal Content of Pb, Cr, and Hg

Type heavy			**Results**			**Average**
No	**metal**	**Unit**	**Sample 1**	**Sample 2**	**Sample 3**	
Rice soil sample						
1.	Pb	mg/kg	73.48	80.15	72.58	75.43
2.	Cr	mg/kg	5.53	5.94	5.71	5.73
3.	Hg	mg/kg	18.28	18.98	15.89	17.72
Sample of rice roots						
1.	Pb	mg/kg	35.38	18.31	11.08	21.59
2.	Cr	mg/kg	36.67	31.40	18.77	28.94
3.	Hg	mg/kg	0.00	0.00	0.00	0.00
Rice straw sample						
1.	Pb	mg/kg	0.00	0.00	0.00	0.00
2.	Cr	mg/kg	13.38	15.71	17.23	15.44
3.	Hg	mg/kg	0.00	0.00	0.00	0.00
Sample of rice seeds						
1.	Pb	mg/kg	0.00	0.00	0.00	0.00
2.	Cr	mg/kg	0.00	0.99	0.82	0.61
3.	Hg	mg/kg	0.00	0.00	0.00	0.00
Rice irrigation water sample						
1.	Pb	mg/kg	0.18	0.14	0.31	0.21
2.	Cr	mg/kg	0.03	0.03	0.06	0.04
3.	Hg	mg/kg	0.02	0.03	0.01	0.02

Description: Spectrometric analysis method, carried out in the Unud Integrated lab Room conditions during analysis: temperature 20 ± 2 0 C, humidity 60 ± 10%; ttd= not detected, Test detection limit for Pb= 0.0170 mg/kg, Cd = 0.0010 mg/kg, Cu = 0.0070 mg/kg, Cr= 0.0019 mg/kg, and Hg= 0.0019 mg/kg

In rice stover, the highest metal content was also found in Cr with an average of 15.44 mg/kg, while heavy metals Pb and Hg were not detected in rice stover. For rice seeds, the highest heavy metal content was Cr with an average of 0.61 mg/kg, and heavy metals Pb and Hg were not detected in rice seeds. In irrigation water, the highest heavy metal was Pb with an average of 0.21 mg/kg, and the lowest metal content was Hg with an average of 0.02 mg/kg. These results show the variation of heavy metal distribution in different media tested, with Pb dominating in soil and water, while Cr accumulated more in plant parts.

Table 3 shows that heavy metals in soil have the potential to be absorbed by plants through the roots, then translocated to other parts of the plant. This is due to the persistent and mobile nature of Pb and Cr, allowing these heavy metals to move from the soil to various organs of rice plants. Meanwhile, heavy metal Hg does not have the potential to be absorbed by the roots.

4.2. Heavy metal content of Pb, Cr and Hg in soil after biochar treatment

The results of the total heavy metal content test of Pb, Cr, and Hg in the soil after the study showed that, biochar coir and coconut shell have the highest heavy metal content in Pb with an average of 9.85 mg/kg, followed by Cr with an average of

5.16 mg/kg, and Hg with an average of 0.24 mg/kg. Fruit and vegetable waste biochar showed the highest heavy metal levels also in Pb with an average of 8.60 mg/kg, followed by Cr with an average of 4.86 mg/kg, and Hg with an average of

0.31 mg/kg. Lignohumic has the highest heavy metal levels in Pb with an average of 8.76 mg/kg, followed by Cr with an average of 5.12 mg/kg, and Hg with an average of 0.25 mg/kg. Of three treatments, Pb had the highest levels in all treatment types, while Hg had the lowest levels. This shows that Pb is the most dominant heavy metal in the post-research soil.

Table 2 Total heavy metal content of Pb, Cd and Cu in the soil after the study planted with rice

Type heavy			**Results**			**Average**
No	**metal**	**Unit**	**Sample 1**	**Sample 2**	**Sample 3**	
Coir and coconut shell biochar (Bc)						
1.	Pb	mg/kg	73.48	80.15	72.58	75.43
2.	Cr	mg/kg	5.53	5.94	5.71	5.73
3.	Hg	mg/kg	18.28	18.98	15.89	17.72
Biochar from fruit and vegetable waste (Bw)						
1.	Pb	mg/kg	9.14	8.27	8.42	8.61
2.	Cr	mg/kg	5.25	4.59	4.75	4.86
3.	Hg	mg/kg	0.29	0.28	0.34	0.31
Lignohumic Biochar (Bl)						
1.	Pb	mg/kg	9.10	8.21	8.96	8.76
2.	Cr	mg/kg	5.20	4.87	5.28	5.12
3.	Hg	mg/kg	0.27	0.24	0.25	0.25

Description: Analyses were conducted at the Soil Science Lab, Faculty of Soil Science, Moscow State University, Russia.

4.2.1. Heavy metal content of Pb, Cr and Hg in rice plant organs after biochar treatment

Rice Root

Table 3 Heavy metal content of Pb, Cr and Hg in rice roots after the study

	BC	**Bw**	**Bl**
Pb	0.00	0.17	5.40
Cr	0.97	1.37	1.14
Hg	0.00	0.00	0.00

Heavy metal Pb content was found in plant roots only in the lignohumic biochar treatment (5.40 mg/kg) and fruit-vegetable waste (0.17 mg/kg). It was not detected in the coir-coconut shell biochar treatment. Cr metal content was detected in all treatments, with the highest levels in fruit-vegetable waste biochar (1.37 mg/kg), followed by lignohumic (1.14 mg/kg) and coconut husk-crack (0.97 mg/kg). No Hg content was found in rice roots in all treatments, indicating that mercury was not absorbed by the roots.

Rice Stover

Table 4 Heavy metal content of Pb, Cr and Hg in rice stover after the study

	BC	Bw	Bl
Pb	0.00	0.00	0.00
Cr	2.43	1.18	0.49
Hg	0.00	0.00	0.00

Heavy metal Pb was not detected in the stover in all biochar treatments. The highest Cr content was found in coir-coconut shell biochar (2.43 mg/kg), followed by fruit- vegetable waste (1.18 mg/kg) and lignohumic (0.49 mg/kg). Hg was not detected in all treatments.

Rice Seeds

Table 5 Heavy metal content of Pb, Cr and Hg in rice seeds after the study

	BC	Bw	Bl
Pb	0.00	0.00	0.00
Cr	1.00	0.68	0.62
Hg	0.00	0.00	0.00

Pb content was not detected in rice seeds in all treatments, indicating low translocation of this heavy metal from roots to seeds.

Cr content was detected in rice seeds with the highest levels in coir-coconut shell biochar (1.003 mg/kg), followed by fruit-vegetable waste (0.68 mg/kg) and lignohumic (0.62). No Hg content was found in rice seeds in all treatments.

4.3. Results of the Effect of Biochar Treatment and Heavy Metal Injection on Rice Growth and Yield

4.3.1. Plant height

Plant height at the Ms, Mf, and Mh levels of planting media treatment was not significantly different with consecutive values of 91.83 cm, 88.50 cm, and 85.71 cm. However, there is a trend in the Ms treatment which has a higher plant height value than Mf and Mh. Plant height in the biochar type treatment Bl has a value of 91.96 cm, significantly different from Bw with a value of 84.89 cm, but in Bc it is not significantly different with a value of 89.20 cm, with a tendency in the Bl treatment which has a higher plant height value than Bc and Bw.

4.3.2. Total grain weight per clump (g)

The treatment of planting media on the variable of total grain weight per clump of Ms, Mf and Mh differed not significantly with values of 71.24 g, 65.64 g, and 66.70 g, respectively. However, there is a tendency in the Ms treatment which has a higher total grain weight per clump compared to other levels.

The total weight of grain per clump in the treatment of biochar types between Bc, Bw and Bl was not significantly different with values of 70.05 g, 65.01 g, and 68.53 g, respectively, with a tendency in the Bc treatment that the total weight of grain per clump was higher than that of Bw and Bl.

4.3.3. Grain yield at 12% moisture content (g)

The grain yield of 12% water content in the treatment of planting media at the Ms level is not significantly different from the Mf and Mh levels with values of 60.47%, 56.01%, 59.78% respectively, but there is a tendency in the Ms treatment which has a higher grain yield of 12% water content compared to other levels. The grain yield of 12% water content in the treatment of biochar types between Bc, Bw and Bl is not significantly different with each value of 60.58%, 55.76%, 59.91%, with a tendency in the Bc treatment which has a higher grain yield of 12% water content than the levels of Bw and Bl.

Table 6 Effect of Biochar Type on Planting Media Contaminated with Heavy Metals Pb, Cr, Hg on Growth

Variables			
Treatment	**Plant Height (cm)**	**Grain Tota Weight per Clump (g)**	**12% Water Content Result (g)**
Biochar Type			
BC	89.20 ab	70.05 a	60.58 a
Bw	84.89 b	65.01 a	55.76 a
Bl	91.96 a	68.53 a	59.91 a
BNT 5%	6.63	10.87	7.82

Notes: Numbers followed by the same letter in the same treatment and column show no significant difference in the least significant difference test (BNT) at the 5% level.

4.4. Bioaccumulation factor (BAF) and translocation factor (TF)

Table 7 BAF and TF values of heavy metals Pb, Cr and Hg in rice plants sampled in farmers' paddy fields

Type Heavy Metals	**Concentration At soil**	**Concentration in plants**	**Concentration At root**	**Concentration At shoots**	**BAF Value**	**TF Value**
	(mg/g)	(mg/g)	(mg/g)	(mg/g)		
Pb	75.43	21.59	21.59	0.00	0.29	0.00
Cr	5.73	44.99	28.94	16.05	7.85	0.55
Hg	17.72	0.00	0.00	0.00	0.00	0.00

Based on Table 7, the highest Bioconcentration Factor (BAF) value was obtained for the heavy metal Cr (Chromium) with a value of 7.85, which indicates the ability of rice plants to absorb large amounts of Cr from the soil. In addition, the highest Translocation Factor (TF) value was also found in Cr with a value of 0.55, which means that most of the Cr accumulated in the roots can be translocated to the top of the plant. This data suggests that rice can act as a strong hyperaccumulator plant for heavy metal Cr. This condition becomes very dangerous for health if rice is grown in soil exposed to heavy metal Cr, because Cr contamination can enter the food chain through consumed rice parts.

Based on Table 8, the highest BAF value in the coir and coconut shell biochar treatment (Bc) was obtained by Cr (Chromium) with a value of 0.85, and the highest TF value was also obtained by Cr with a value of 3.53. In the biochar treatment of fruit and vegetable waste (Bw), the highest BAF value was obtained by Cr with a value of 0.66, and the highest TF value was also obtained by Cr with a value of 1.35. , in the biochar treatmentlignohumic (Bl), the highest BAF value was obtained by Pb (Lead) with a value of 0.62, and the highest TF value was obtained by Cr with a value of 0.97.

Table 8 BAF and TF values of heavy metals Pb, Cr and Hg in rice plants grown with different biochar types

Metal Type Weight	**Concentrati on in soil (mg/g)**	**Concentrati on in the plant (mg/g)**	**Concentrati on in roots (mg/g)**	**Concentrati on in shoots (mg/g)**	**BAF Value**	**TF Value**
Coir and coconut shell biochar (Bc)						
Pb	9.88	0.00	0.00	0.00	0.00	0.00
Cr	5.16	4.40	0,97	3.43	0.85	3.53
Hg	0.24	0.00	0.00	0.00	0.00	0.00
Biochar from fruit and vegetable waste (Bw)						
Pb	8.61	0.17	0.17	0.00	0.02	0.00
Cr	4.86	3.23	1.37	1.86	0.66	1.35

Hg	0.31	0.00	0.00	0.00	0.00	0.00
Lignohumic Biochar (Bl						
Pb	8.76	5.40	5.40	0.00	0.63	0.00
Cr	5.12	2.25	1.14	1.11	0.44	0.97
Hg	0.25	0.00	0.00	0.00	0.00	0.00

The use of coconut husk and shell biochar (Bc) and fruit and vegetable waste biochar (Bw) tended to be more effective in reducing the accumulation of heavy metals Pb and Cr in rice plants compared to lignohumic biochar (Bl). In addition, in all treatments, no accumulation of heavy metal Hg (Mercury) was found in rice plants, indicating that Hg was not absorbed by plants.

5. Conclusion

The use of biochar, especially those made from coconut husks and shells, has been proven effective in reducing heavy metal levels (Pb, Cr, Hg) in soil and plants. This biochar increased the productivity of rice (Oryza sativa L.) by increasing the number of productive tillers, leaf chlorophyll content, and grain weight. In addition, biochar is able to retain heavy metals in the roots with low Bioaccumulation Factor (BAF) and Translocation Factor (TF) values, reducing the risk of contamination in rice seeds. This study shows that biochar can be an ecological solution to manage heavy metal contaminated agricultural land

Compliance with ethical standards

Acknowledgments

All authors from various faculties at Udayana University compiled this journal article based on research on the effect of biochar types on planting media polluted with heavy metals on the growth and yield of rice (Oryza sativa L.). This research was funded by Udayana University and conducted from May to October 2024. The ideas expressed in this article are the result of the collaboration of the research team who have carried out research for 5 months with an ecological approach to overcome heavy metal pollution on agricultural land.

Disclosure of conflict of interest

We have no conflicts of interest to disclose. All authors declare that they have no conflicts of interest.

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