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## Absorption Efficiency of Heavy Metals by Water Spinach with Rice Husk Char in Soils

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## Abstract

Asian rice is the most widely produced crop in Taiwan. Its husks derived from milling can serve as fuel, compost, as well as pyrolysis-generated biochar to create economic value. In this study, the researchers added rice husk char (RHC) at different concentrations (i.e., 0%, 10%, 15%, and 20%) to soil contaminated by heavy metals. Next, a pot experiment was implemented to observe changes in heavy metal concentrations within the aboveground parts of water spinach and tested soil. Regarding the efficiency of different soil in absorbing heavy metals, soil with high RHC volume outperformed that with less RHC. Concerning the efficiency of edible plant parts in absorbing heavy metals at different growth stages, the water spinach plants absorbed more Cd and Cu than other metal elements. Moreover, soil added with RHC proved to effectively absorb heavy metals with less interference, allowing the spinach plant to grow healthily in a favorable environment. The soil porosity also increased following RHC addition, resulting in a more developed root system. This study serves as a reference for researchers to determine the effects of biochar presence on ion transmission of heavy metals and their application in soil improvement.

Keyword: Rice Husk Char, Water Spinach, Biochar, Pot Experiment.

## Introduction

Deficiency and excess of nutrient metal elements in plants will cause metabolism disorders [1]. In addition to the properties of heavy metals and the plant in question, soil quality and environmental status can affect heavy metal accumulation [2-4]. In everyday life, heavy metals are recognized depending on the environmental pollution level; generally, elements that are evidently toxic to living organisms are considered heavy metals, such as Cd, Cr, Cu, Ni, Pb, Zn, Mn, Co, and Hg. Specifically, arsenic, despite not being a metal element, has a specific gravity of 5.7 and is noticeably toxic to living organisms. Moreover, many of its chemical properties are similar to those of general heavy metals. Therefore, arsenic is categorized as a heavy metal. By contrast, plants have lower demand for micronutrients such as Fe, Cu, Zn, and Mn. When the increase in concentrations remains constant, enhancing crop's amount of growth helps create more benefits than does enhancing macronutrients, while achieving the highest yield more quickly. Adding a small concentration of micronutrients allows crop growth to peak and start to fall faster than that in the macronutrient case [5,6]. However, excessive macronutrient or micronutrient consumption tends to reduce the amount of growth due to imbalanced nutrition. Particularly, only a little excess of micronutrients can cause toxic injuries.

Carbonized rice husks are the most common agricultural biochar. Biochar is obtained using various raw materials through pyrolysis in an anoxic state [7]. Its high specific surface area and porous nature allows the material to absorb pollutants and lock them in the contaminated soil [8,9]. The straw and husks of rice after harvesting are desirable biochar

sources [10,11]. Numerous studies have explored the absorbability of different biochar against heavy metals in a solution or soil. Furthermore, biochar helps reduce greenhouse gas emissions in soil [12] and enhance soil fertility and crop productivity [6,13]. Dense soil tends to inhibit the root growth and reduce biological activities within soil [14]. To solve this problem, adding biochar can effectively improve soil porosity and stimulate plant growth [15]. In addition, the oxides (e.g., Na, K, Mg, and Ca) or carbonates within biochar allows the material to turn into an alkaline material capable of pH adjustment for acidic soil [16].

Considering the aforementioned properties, this study explores the efficiency of two types of contaminated soil, added with rice husk char (RHC) at different concentrations, in absorbing heavy metals at the aboveground parts of water spinach (*Ipomoea aquatica*). A pot experiment was conducted to determine concentration changes in heavy metals within the aboveground parts of plants and tested soil.

## Materials and methods

**Sources of the soil samples:** The Soil-A and Soil-B adopted in this study originate from adjacent farmlands contaminated by heavy metals in central Taiwan; the two places have similar soil quality but different heavy metal concentrations. After soil samples were obtained, they were separately screened using a 2mm-mesh sieve and mixed evenly. The samples were then subjected to a basic property analysis.

**Source of the rice husk char:** The RHC used in this study originates from an agricultural material supplier in Taiwan. The biochar was formed by implementing pyrolysis on rice husks at 350°C following the husk removal process.

**Source of the fertilizer:** In this study, the fertilizer administered during the pot experiment is an organic fertilizer available on the market, Taiwan. It consists of 3.5% total nitrogen, 3.0% total phosphorus pentoxide, 3.0% total potassium oxide, 1.5% water-soluble magnesium oxide, and 65.0% organic substances with a pH of 6.0.

Analysis of soil quality: The researcher placed 50 g of dried soil samples in a beaker and added 300 mL of reagent water and 10 mL of  $(NaPO_3)_6$  solution to it, after which the mixture was left to stand still for 10 min, allowing the soil to disperse. Next, the researcher stirred the mixture for 10 min using a stirrer and poured it into a graduated cylinder before adding reagent water to 1,000 mL. Subsequently, the cylinder was sealed and evenly shaken; after standing still for 1 h, the ratio among sand grain, silt, and clay within the soil was determined.

**pH measurement:** The soil samples were first dried and screened (using a 20-mesh sieve). A total of 20±0.2 g of each sample was placed in a beaker and added with 20 mL of reagent water. Subsequently, a watch glass was adopted to cover the beaker, in which the sample and water were stirred continually for 5 min. When a complex matrix containing water-absorbing soil or salt was identified, reagent water was added in turn to dilute the mixture until the matrix in question disappeared. After most solids deposited within the mixture, the pH level at the aqueous phase layer was measured.

**Pot experiment and experimental procedures:** Soil-A and Soil-B were separately mixed with RHC at different concentrations and divided into Group Soil-A and Group Soil-B, after which water spinach was planted on them and went through germination to harvest. Specifically, 1000, 900, 850, and 800 mL of Soil-A and Soil-B were added with 0, 100, 150, 200 mL of RHC, respectively. Next, 5 g of fertilizer was added to each control group and experimental group before

the soil sample was placed in a pot. Ten water spinach seeds were then sown into each pot; they were gently pressed into topsoil and covered up. Next, the pots were moved to an outdoor space with sufficient sun exposure and watered until water came out the drain holes.

#### **Results and discussion**

**Code definitions:** This section defines the codes and parameters associated with experimental results in this study, as summarized in Table 1.

### Basic properties of the tested soil and RHC

Table 2 presents the basic properties of Soil-A, Soil-B, and RHC. The pH values of Soil-A and Soil-B were 7.08 and 7.10, respectively, signifying their neutrality; the quality of the two samples was both silty clay loam. The pH of RHC was 7.46. The heavy metal Cd concentrations within Soil-A and Soil-B were 7.12 mg/kg and 12.8 mg/kg, respectively, surpassing the 5-mg/kg limit stipulated by the Environmental Protection Administration, Taiwan, for farmlands growing edible crops in the Soil Pollution Control Standards. Moreover, the Pb concentration within Soil-B was 763 mg/kg, much higher than the stipulated 500 mg/kg limit. Similarly, the Zn concentrations within Soil-A and Soil-B were 2,250 mg/kg and 4,280 mg/kg, respectively, both exceeding the standard limit of 2,000 mg/kg.

## Changes in pH levels of soils

Figure 1 illustrates the pH changes in the soil samples. Following the water spinach harvest, the pH levels of Soil-A samples climbed from 7.08 to 7.20–7.60, with A1 having the highest value. Those of Soil-B escalated from 7.10 to 7.49–7.97, with B1 having the highest value. The results revealed that adding more than 10% RHC to soil for growing water spinach converted the soil from a weak alkali to a neutral substance. Nuckols et al. [17] indicated that silty clay loam has a moderately fine texture, desirable water holding and nutrient preserving capacities, and preferable soil structure, thereby facilitating the growth of plants with rich organic substances and nutrients, hence its high suitability for growing general vegetables. This study demonstrated that adding a certain amount of RHC to soil helps improve the yield and quality of cash crops.

#### Changes in heavy metal concentrations within soil

Changes in heavy metal concentrations within Soil-A and Soil-B samples, added with RHC at different concentrations, were analyzed after the edible aboveground parts of water spinach were harvested. According to the results shown in Table 3 (Groups 1, 2, and 3 have been defined in Table 1), both Soil-A and Soil-B samples with higher RHC content delivered greater performance in absorbing heavy metals than did those with lower RHC content. Specifically, the Soil-A subgroups with RHC achieved the optimal performance by absorbing up to 66.7% of Cd, followed by their performance in absorbing Pb (66.0%). The Soil-B subgroups with RHC exhibited highest absorbability of Pb, up to 65.9% of the heavy metal, followed by that of Zn, 63.3%.

The aforementioned results demonstrated that adding RHC to soil helped effectively reduce the amount of heavy metals absorbed by the aboveground parts of water spinach plants, which was consistent with Li et al. [18]. A plant with excessive unnecessary heavy metal uptake may be poisoned; for example, a high Cu concentration can kill the root cells

within a short period of time mainly because the heavy metal provokes plant cells, generates free radicals, and forces the cells to remain in an oxidation state, consequently inhibiting plant growth [18,19]. Thanks to its high specific surface area and porous nature, the RHC adopted in this study could absorb and lock pollutants within the soil in question, thereby reducing the amount of heavy metals adsorbed by the water spinach plant.

Changes in heavy metal concentrations within the aboveground parts of water spinach

Changes in heavy metal concentrations within the harvested edible aboveground parts of water spinach plants grown on Soil-A and Soil-B groups (administered with different volumes of RHC) were determined (Table 4); the heavy metal absorbability of the harvests was also analyzed (Figure 2). The results in Table 4 revealed that concentrations of several heavy metals decreased among plants grown on soil with high RHC content. However, concentrations of other heavy metals increased with the RHC content.

As the results in Figure 2 displayed, water spinach plants grown on the two types of RHC-added soil performed most desirably in absorbing Cd, followed by Cu, Ni, Zn, Cr, and Pb, with their absorbability being particularly strong for Cd and Cu. A possible reason was that the electron configuration of Cd made the metal prone to be absorbed by plants and thus travelled to their aboveground parts [20]. The study results suggested that adding RHC to soil enabled a water spinach plant to effectively concentrate Cd, resulting in greater absorbability. Additionally, a previous study proved the presence of selective adsorption of Cu in general crops, particularly water spinach [17], which is consistent with the present study; that is, adding a certain amount of RHC allows water spinach to more desirably absorb Cu compared with other metals.

## **Pot experiment**

#### Growth process of water spinach

Figures 3 to 7 record the growth of water spinach plants, of which the seeds were soaked in water before being sown into Soil-A and Soil-B, on Day 1, Day 14, and Day 30. The ambient temperature range was 15–27°C, with day and night temperatures averaging 19–23°C. The aboveground parts of plants grown on Soil-A were 12.7–16.4 cm tall on Day 30, with the WA2 subgroup being the tallest, followed by WA1, WA3, and WA-CG; those of plants grown on Soil-B were 11.7–17.2 cm tall on Day 30, with the WB2 subgroup being the tallest, followed by WB3, WB1, and WB-CG. The statistics indicated that plants grown on soil without RHC were the shortest in both groups. Accordingly, adding RHC enabled the soil to effectively absorb heavy metals [3,17], providing a favorable environment for water spinach to grow without much interference, hence the preferable growth status.

Growth of root system in soil

Figure 8 depicts the root system growth in the four Soil-A subgroups. Specifically, the A-CG root system was the least developed, whereas A3 was the most developed, demonstrating that adding RHC to soil enabled an increase in porosity of clay loam [21], stimulating mature development of the water spinach root system.

Ching-Lin Ho, et al. International Journal of Engineering Research and Generic Science (IJERGS)

#### Conclusion

In this study, the researchers added RHC at different concentrations to soil contaminated by heavy metals and compared the heavy metal concentrations between the aboveground water spinach parts grown on the soil by implementing a pot experiment. The pH measurement results showed that adding >10% RHC to soil would convert it from a weak alkali to a neutral element. Regarding efficiency of the two soil types in absorbing heavy metals, this study discovered that soils added with more RHC outperformed its counterparts with less RHC. Concerning changes in heavy metal concentrations within the aboveground parts of water spinach, a specific volume of RHC enabled water spinach to effectively concentrate Cd and Cu, namely more desirable performance in absorbing Cd and Cu compared with other metals. After conducting a pot experiment on the growth status of water spinach plants, this study identified that soil with RHC effectively absorbed heavy metals with reduced interference, creating a favorable environment for the plants to grow and leading to more desirable growth status. Additionally, the soil became more porous, which was conducive to root system development for water spinach. The pot experiment was performed in an outdoor space, where climate conditions were unpredictable. Future studies could regard influential factors (e.g., sunshine hours, temperature, humidity, and pests and diseases) as control variables to improve research efficiency and reliability.

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Ching-Lin Ho, et al. International Journal of Engineering Research and Generic Science (IJERGS)

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# Legend Tables and Figures

Table 1:Definitions of codes adopted in this study

Code	Definition	RHC concentration (% v/v)
A soil	Basic properties of Soil-A	-
B soil	Basic properties of Soil-B	-
A-CG	Soil-A:rice husk char = 1000:0 mL; the soil following water spinach	0
	harvest (excluding RHC and root system)	
A1	Soil-A:rice husk char = 900:100 mL; the soil following water spinach	10
	harvest (excluding RHC and root system)	
A2	Soil-A:rice husk char = 850:150 mL; the soil following water spinach	15
	harvest (excluding RHC and root system	
A3	Soil-A:rice husk char = 800:200 mL; the soil following water spinach	20
	harvest (excluding RHC and root system)	
B-CG	Soil-B:rice husk char = 1000:0 mL; the soil following water spinach	0
	harvest (excluding RHC and root system)	
B1	Soil-B:rice husk char = 900:100 mL; the soil following water spinach	10
	harvest (excluding RHC and root system)	
B2	Soil-B:rice husk char = 850:150 mL; the soil following water spinach	15
	harvest (excluding RHC and root system)	
B3	Soil-B:rice husk char = 800:200 mL; the soil following water spinach	20
	harvest (excluding RHC and root system)	
WA-CG	The aboveground part of water spinach grown on A-CG soil	-
WA1	The aboveground part of water spinach grown on A1 soil	-
WA2	The aboveground part of water spinach grown on A2 soil	-
WA3	The aboveground part of water spinach grown on A3 soil	-
WB-CG	The aboveground part of water spinach grown on B-CG soil	-
WB1	The aboveground part of water spinach grown on B1 soil	-
WB2	The aboveground part of water spinach grown on B2 soil	-
WB3	The aboveground part of water spinach grown on B3 soil	-

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Substance	Soil-A	Soil-B	rice husk char
Indicator			
Sand%	18	16	-
Silt%	50	48	-
Clay%	32	36	-
Texture	Silty clay loam	Silty clay loam	-
рН	7.08	7.10	7.46
EC(µs/cm)	86.6	90.3	872
Cd(mg/kg)	7.12	12.8	0.15
Cr(mg/kg)	97.8	97.9	5.53
Cu(mg/kg)	108	146	6.81
Pb(mg/kg)	385	763	2.41
Ni(mg/kg)	38.4	77.3	4.30
Zn(mg/kg)	2250	4280	61.6

Table 2: Basic property analysis of the tested soil and RHC

Table 3: Changes in heavy metal concentrations within soil

	Soil	CG	1	2	3
Cd(mg/kg)	)		I	I	
Soil-A	7.12	2.63	3.03	2.53	2.37
Soil-B	12.7	6.82	8.22	6.88	6.47
Cr(mg/kg)					L
Soil-A	94.8	62.8	56.9	66.6	54.9
Soil-B	97.9	77.6	75.9	73.3	71.2
Cu(mg/kg)	)		I	I	L
Soil-A	108	61.4	67.0	64.6	60.5
Soil-B	146	104	105	101	91.4
Pb(mg/kg)			I	I	
Soil-A	385	112	177	136	131
Soil-B	763	417	463	400	260
Ni(mg/kg)	I		L.	- I	
Soil-A	87.4	72.5	72.0	73.7	73.1
Soil-B	77.3	72.2	70.2	73.5	71.5
Zn(mg/kg)	·		1	1	L

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Soil-A	2250	743	982	799	813
Soil-B	4280	2510	2620	2360	1570
Table 4: Ch	nanges in heavy	y metal concentr	rations within th	e aboveground j	parts of water spina
	Soil	CG	1	2	3
Cd(mg/kg	)				
WA	-	1.69	1.47	1.27	1.35
WB	-	3.05	2.68	1.99	1.87
Cr(mg/kg)	)				
WA	-	3.29	3.05	3.23	3.15
WB	-	4.17	3.42	3.28	3.96
Cu(mg/kg	)				
WA	-	29.1	26.9	25.0	26.8
WB	-	28.8	27.1	23.4	25.3
Pb(mg/kg)	)				
WA	-	4.79	4.59	4.69	4.15
WB	-	6.78	6.15	6.26	4.96
Ni(mg/kg)	)				
WA	-	4.53	3.26	3.30	3.19
WB	-	4.47	3.23	3.02	3.04
Zn(mg/kg	)			I	I
WA	-	119	91.8	88.7	97.9
WB	-	156	114	102	96.0

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Figure 1: Changes in pH values within soil following water spinach harvest



Figure 2: Comparison between water spinach harvests in absorbing different heavy metals



Soil-A



Figure 3: Water spinach plants grown on Soil-A and Soil-B on Day 1



Figure 4: Water spinach plants grown on Soil-A on Day 14



Figure 5: Water spinach plants grown on Soil-B on Day 14



Figure 6: Water spinach plants grown on Soil-A on Day 30



Figure 7: Water spinach plants grown on Soil-B on Day 30









Figure 8: Root development of water spinach grown on Soil-A