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## Comparative Evaluation of Tree Species for Phytoremediation of Heavy Metal-Contaminated Soil: Effects of Humic and Salicylic Acid Application

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### ABSTRACT

In order to assess the efficacy of phytoremediation in restoring degraded soils, specifically those contaminated with oil residues, a research pot trial was conducted. The main objective of the study was to compare the phytoremediation performance of various tree species, namely *Eucalyptus camaldulensis*, *Albizia lebbek*, *Ficus carica*, and *Morus nigra* which represented the main factor. Additionally, the investigation aimed to evaluate the impact of different rates of humic acid HA (0, 5, 10 g L<sup>-1</sup>) as a sub-main factor, as well as varying rates of salicylic acid SA (0, 250, 500 mg L<sup>-1</sup>) as a sub-sub-main factor, on the phytoremediation capabilities of these trees. The specific focus was on their ability to remove heavy metals *i.e.*, Zn, Pb, Cd, Ni from oil residues-contaminated soil. The results showed that *Albizia lebbek* transplants exhibited the lowest soil Zn concentration (15.62 mg Zn.kg<sup>-1</sup> soil) and a removal rate of 78.98%. Similarly, *Albizia lebbek* transplants exhibited the lowest levels of soil available Pb, Cd and Ni concentrations compared to the other tree species studied, with higher removal rates. Furthermore, the foliar applications of both humic and salicylic acids resulted in a reduction of Zn, Cd, Pb, and Ni residues. The uptake of these heavy metals by the roots increased with higher concentrations of HA and SA. Specifically, the lowest values were recorded when the trees were treated with a combined application of HA at a rate of 10 g L<sup>-1</sup> and SA at a rate of 500 mg L<sup>-1</sup>.

**Keywords:** Phytoremediation, HA and SA



### INTRODUCTION

Soil pollution refers to the contamination of soil with harmful substances that can have adverse effects on plant and animal life, as well as human health (Sankhla *et al.* 2016). It occurs when pollutants are introduced into the soil through human activities, such as industrial processes, improper waste disposal, agricultural practices, the use of chemical fertilizers and pesticides and mining activities and oil residues. These pollutants can alter the chemical, physical, and biological properties of the soil, rendering it unsuitable for its intended purpose (Ukaogo *et al.* 2020). Phytoremediation is a form of environmental remediation that utilizes plants to remove, degrade, or stabilize pollutants from the soil. It is a cost-effective and environmentally friendly approach to clean up contaminated sites and restore their quality (Dhanwal *et al.* 2017). Phytoremediation can be applied to a wide range of contaminants, including heavy metals, organic compounds, pesticides, and radioactive substances. Certain plant species possess the unique ability to accumulate significant amounts of pollutants in their tissues without experiencing adverse effects of toxicity (Kanwal *et al.* 2019). It's important to note that the effectiveness of phytoremediation depends on various factors such as the type and concentration of pollutants, soil conditions, climate, and the specific characteristics of the plants used. Additionally, successful phytoremediation projects often involve the selection and combination of multiple plant species to target different types of pollutants and maximize the remediation process (Lim and Lim 2012). There is a considerable number of approximately 400 plant species belonging to 45 different families that are recognized

as accumulating plants. These plants have the capability to accumulate pollutants, such as heavy metals, in their tissues as part of the phytoremediation process (Baker *et al.* 2020). For example, *Eucalyptus camaldulensis*, *Albizia lebbek*, *Ficus carica* and *Morus nigra* can be used in phytoremediation. *Eucalyptus camaldulensis* has been widely used in phytoremediation projects due to its ability to absorb large amounts of water and heavy metals from the soil (Seenivasan *et al.* 2015). *Albizia lebbek* is a fast-growing tree species with a high tolerance for various soil conditions. It is often utilized in phytoremediation projects for its ability to extract heavy metals, such as lead and cadmium, from contaminated soils. (Zakari and Audu, 2021). *Ficus carica* is known for its adaptability to different soil types and climates. It can be used in phytoremediation projects to remediate contaminated soils through a process called phytoextraction. Phytoextraction involves the uptake and accumulation of pollutants, such as heavy metals, by the plant's roots, stems, and leaves (Ahmad *et al.* 2023). *Morus nigra* has been used in phytoremediation due to its ability to tolerate and extract pollutants from contaminated soil. This tree species is particularly effective in the remediation of soils contaminated with organic pollutants. The roots of *Morus nigra* release enzymes that break down organic compounds, contributing to the degradation of contaminants in the soil (Lim and Lim, 2012). Spraying both humic acid and salicylic acid on trees can have beneficial effects on their nutrition and overall health. Humic acid (HA) is a complex mixture of organic compounds that forms as a result of the decomposition and transformation of plant and animal materials (Shah *et al.*

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2018). HA can penetrate the leaf surface and promote nutrient uptake through the stomata, which are tiny openings on the leaf surface. It improves the efficiency of nutrient absorption by the leaves and facilitates their translocation to various parts of the tree (Hussein *et al.* 2020). It can enhance photosynthetic activity in tree leaves by increasing chlorophyll production and improving the efficiency of light capture (Mahmood *et al.* 2022). Salicylic acid (SA) is a natural phytohormone with the chemical formula C<sub>7</sub>H<sub>6</sub>O<sub>3</sub>. It is a colorless crystalline solid that is derived from the bark of the willow tree (*Salix* species) and certain other plants (AL-Hchami *et al.* 2020). SA is also synthetically produced for various applications. Salicylic acid is involved in the tree's defense response against pathogens, pests, and environmental stresses. Spraying salicylic acid can activate defense mechanisms, such as the production of antimicrobial compounds, antioxidants, and strengthening cell walls (Lefevre *et al.* 2020). This helps protect the tree from diseases and stressors that could impact its nutrient uptake and utilization (El Refaey *et al.* 2022).

Therefore, the main objective of this study is to compare the effectiveness of various tree species, namely *Eucalyptus camaldulensis*, *Albizia lebbbeck*, *Ficus carica*, and *Morus nigra*, in phytoremediation. The specific focus was on their ability to remove heavy metals like lead, cadmium, nickel, and zinc from oil residues-contaminated soil. Additionally, the study aimed to assess the influence of applying humic and salicylic acids at different rates on the phytoremediation capabilities of these trees.

## MATERIALS AND METHODS

This research study was carried out to compare the phytoremediation performance of various tree species, namely *Eucalyptus camaldulensis*, *Albizia lebbbeck*, *Ficus carica*, and *Morus nigra* which represented the main factor. Additionally, the investigation aimed to evaluate the impact of different rates of humic acid (0, 5, 10 g L<sup>-1</sup>) as a sub-main factor, as well as varying rates of salicylic acid (0, 250, 500 mg L<sup>-1</sup>) as a

sub-sub-main factor, on the phytoremediation capabilities of these trees. The specific focus was on their ability to remove heavy metals *i.e.*, zinc (Zn), lead (Pb), cadmium (Cd), nickel (Ni) from oil residues-contaminated soil.

### location

The study took place in Bismayah City, located at coordinates 33° 11' 42" N, 44° 35' 56" E. It was carried out over the course of two growing seasons, spanning from 2021 to 2022. Bismayah City is situated approximately 10 kilometres southeast of Baghdad.

### Soil sampling and experimental setup

Soil samples were collected from the pots before planting the transplants and before conducting the treatments. Afterwards, the samples were subjected to laboratory analysis. The physical and chemical properties of the soil are presented in Table 1. The experimental setup involved using soil from a contaminated site with oil residues, which was mixed with sandy loam soil at a ratio of 3:1 (natural soil: contaminated soil) and placed in pots with dimensions of 30 cm in diameter and 35 cm in depth. The purpose was to plant transplants in these pots and assess the effects of the study factors on them. The transplants used in the study were approximately 2 years old, healthy, and exhibited similar vigor and size.

The experimental trees were managed following standard agricultural practices commonly used for transplants, including fertilization, irrigation, pruning, and pest control. A mineral fertilizer with a composition of 20N:20P:20K was applied in ten equal doses, with each dose amounting to 5.0 g per pot. The first dose was applied 20 days after transplanting, and subsequent doses were repeated at two-week intervals. Irrigation was carried out every three days using fresh water. The treatments were replicated three times, with each replicate consisting of three transplants, following a split-split plot design. A total of 324 transplants were used in the study, with 81 transplants for each tree species. Spraying operations were conducted three times, with two applications in spring and one in October.

**Table 1. Soil analysis before applying treatments**

Particle size distribution (%)			Textural Class	Field capacity (%)	Organic matter (%)	Total CaCO <sub>3</sub> (%)		
Clay	Silt	Sand	Sandy loam	38.25	0.75	2.3		
18	37	45						
pH	EC, dsm <sup>-1</sup>	Available N	Available P	Available K	Available Ni (mg Kg <sup>-1</sup> )	Available Pb	Available Zn	Available Cd
8.1	1.40	34.2	3.5	191.5	121.36	10.77	74.3	2.44

### Measurements

#### Soil heavy metals extracted with DTPA:

Prior to the commencement and after the completion of the experiment, the heavy metals (Cd, Pb, Ni, Zn) present in the soil were extracted using the chelating agent DTPA (Diethylene triamine pentaacetic acid) as described by Jones (2001). The concentrations of these metals were determined using an atomic absorption spectrometer (Shimadzu AA-7000 model 2013). Subsequently, the following calculations were performed: Removal rate was calculated according to Najem, (2015)

$$\text{The removal rate} = \frac{C1 - C2}{C1} * 100\%$$

Where C1: The firstly concentration C2: is the secondly concentration

The biological concentration factor (BCF) was determined using the equation proposed by Li *et al.* (2007) and Cui *et al.* (2007). It was calculated by dividing the metal concentration in the leaves by the metal concentration in the soil. A BCF value greater than one indicates a high capacity of the plant to absorb and accumulate the heavy metal in its tissues. Conversely, a BCF value less than one indicates the plant's inability to absorb sufficient quantities of heavy metals from the soil.

### Statistical analysis

The collected data were subjected to variance analysis using the Costat program v. 6.3. The results were organized in a factorial experimental design with three replicates. To compare the differences between treatment means, an analysis of variance was performed following the method proposed by Elsahook and Wuhaib (1990),

and the least significant difference (L.S.D) at a significance level of 0.05 was used.

## RESULTS AND DISCUSSION

### Results

#### Soil available zinc and removal rate at the end of the experiment

The results presented in Table 2 provide information about the concentration of soil available zinc and the biological concentration factor (BCF) of zinc in the leaves of four transplant species. The concentration of soil available zinc was found to be 74.30 mg Zn.kg<sup>-1</sup> soil. Significant differences were observed in the concentrations of available zinc in the soil after the end of the experiment, which can be attributed to the planting of different transplant species. The lowest concentration was recorded in pots with *Albizia lebbek* transplants, measuring 15.62 mg Zn.kg<sup>-1</sup> soil, indicating a removal rate of 78.98%. In contrast, pots with fig transplants had a higher concentration of 40.46 mg Zn.kg<sup>-1</sup> soil, with a removal rate

of 45.30%. It is worth noting that the available zinc concentrations in the soil did not exceed the critical limits of 300 mg Zn.kg<sup>-1</sup> soil, as defined by the World Health Organization/Food and Agriculture Organization (WHO/FAO, 2007). This suggests that plants have the ability to absorb zinc from the soil. Zinc is an essential micronutrient for plants, and its absorption by plants increases during their growth period. This finding is consistent with the study conducted by Hacısalihoglu *et al.* (2001), which reported an increase in plant uptake of zinc from the soil due to its role as a micronutrient, and a decrease in its available concentrations in the soil. Furthermore, the BCF of zinc in the leaves of the four transplant species was determined. The BCF represents the ratio of the concentration of zinc in the leaves to the concentration of zinc in the soil. The results showed that there was no increase in the BCF for any of the plants, as all values were below one. This indicates that zinc is consumed in various metabolic processes within the plants rather than being accumulated in the leaves.

**Table 2. Role of *Eucalyptus camaldulensis*, *Albizia lebbek*, *Ficus carica* and *Morus nigra* transplants on soil available zinc (mg Zn.kg<sup>-1</sup> soil), removal rate (%) and BCF**

Treatments	Zn content of initial soil was 74.30 mg.kg <sup>-1</sup> (before)															
	<i>Eucalyptus camaldulensis</i>				<i>Albizia lebbek</i>				<i>Ficus carica</i>				<i>Morus nigra</i>			
	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF
H <sub>0</sub> S <sub>0</sub>	16.12	78.30	21.54	0.290	15.74	78.82	21.61	0.291	41.88	43.63	17.25	0.232	28.80	61.23	17.83	0.240
H <sub>0</sub> S <sub>250</sub>	15.88	78.63	21.86	0.294	15.72	78.84	21.95	0.295	41.77	43.78	17.48	0.235	28.76	61.29	18.14	0.244
H <sub>0</sub> S <sub>500</sub>	15.80	78.73	22.16	0.298	15.72	78.84	22.26	0.300	41.72	43.85	17.97	0.242	28.74	61.32	18.80	0.253
H <sub>5</sub> S <sub>0</sub>	15.77	78.78	21.93	0.295	15.68	78.90	22.04	0.297	40.23	45.85	17.93	0.241	28.20	62.05	18.65	0.251
H <sub>5</sub> S <sub>250</sub>	15.77	78.78	22.72	0.306	15.68	78.90	22.83	0.307	40.11	46.02	18.16	0.244	28.15	62.11	19.15	0.258
H <sub>5</sub> S <sub>500</sub>	15.70	78.87	22.84	0.307	15.62	78.98	22.97	0.309	40.23	45.85	18.84	0.254	28.11	62.17	20.31	0.273
H <sub>10</sub> S <sub>0</sub>	15.72	78.84	22.87	0.308	15.55	79.07	23.01	0.310	40.00	46.16	18.91	0.255	27.98	62.34	19.83	0.267
H <sub>10</sub> S <sub>250</sub>	15.66	78.92	23.65	0.318	15.50	79.14	23.80	0.320	39.94	46.24	19.45	0.262	27.86	62.50	20.26	0.273
H <sub>10</sub> S <sub>500</sub>	15.60	79.00	24.19	0.326	15.41	79.26	24.36	0.328	39.87	46.34	20.11	0.271	27.86	62.50	21.41	0.288
Mean	15.78	78.76	22.64	0.305	15.62	78.98	22.75	0.306	40.64	45.30	18.45	0.248	28.27	61.95	19.37	0.261

Since, H<sub>0</sub>: Without humic acid (control); H<sub>5</sub>: Spraying humic acid at rate of 5 g.L<sup>-1</sup>; H<sub>10</sub>: Spraying humic acid at rate of 10 g.L<sup>-1</sup>; S<sub>0</sub>: Without salicylic acid (control); S<sub>250</sub>: Spraying salicylic acid at rate of 250 mg.L<sup>-1</sup>; S<sub>500</sub>: Spraying salicylic acid at rate of 500 mg.L<sup>-1</sup>

#### Soil available lead (Pb) and removal rate at the end of the experiment

The results presented in Table 3 provide information about the concentration of soil available lead (Pb) and the biological concentration factor (BCF) of lead in the tissues of four transplant species.

The concentration of soil available lead was determined to be 10.77 mg Pb.kg<sup>-1</sup> soil. Significant differences were observed in the concentrations of available lead in the soil after the experiment, which can be attributed to the planting of different transplant species. The lowest concentration was found in pots with *Albizia lebbek* transplants, measuring 4.16 mg Pb.kg<sup>-1</sup> soil, indicating a removal rate of 61.37%. In contrast, pots with fig transplants had a higher concentration of 7.76 mg Pb.kg<sup>-1</sup> soil, with a removal rate of 27.95%. It is important to note that the available lead concentrations in the soil did not exceed the critical limits of 100 mg Pb.kg<sup>-1</sup> soil, as defined by the World Health Organization/Food and Agriculture Organization (WHO/FAO, 2007).

Furthermore, the BCF of lead in the tissues of the four transplant species was determined. The BCF represents the ratio of the concentration of lead in the plant tissues to

the concentration of lead in the soil. The results showed that all BCF values exceeded one for all the plants studied. This indicates that these plants have the ability to transport and accumulate lead within their tissues. According to Lyubenova and Schrodes (2010), plants with a BCF greater than one are considered to have a strong ability to accumulate heavy metals, and these plants are commonly used in phytoremediation, which is the process of using plants to remove or mitigate heavy metal contamination.

#### Soil available cadmium (Cd) and removal rate at the end of the experiment

The results presented in Table 4 provide information about the concentration of soil available cadmium (Cd) and the biological concentration factor (BCF) of cadmium in the tissues of four transplant species.

The concentration of soil available cadmium was determined to be 2.44 mg Cd.kg<sup>-1</sup> soil. Significant differences were observed in the concentrations of available cadmium in the soil after the experiment, which can be attributed to the planting of different transplant species. The lowest concentration was found in pots with *Eucalyptus camaldulensis* transplants, measuring 0.056 mg Cd.kg<sup>-1</sup> soil, indicating a removal rate of 97.70%. In contrast, pots with

*Ficus carica* transplants had a higher concentration of 0.91 mg Cd.kg<sup>-1</sup> soil, with a removal rate of 62.70%. It is important to note that the available cadmium concentrations in the soil did not exceed the critical limits of 3.0 mg Cd.kg<sup>-1</sup> soil, as defined by the World Health Organization/Food and Agriculture Organization (WHO/FAO, 2007).

Furthermore, the BCF of cadmium in the tissues of the four transplant species was determined. The BCF represents the ratio of the concentration of cadmium in the plant tissues to the concentration of cadmium in the soil.

The results showed that all BCF values exceeded one for all the plants studied. This indicates that these plants have the ability to transport and accumulate cadmium within their tissues. According to Somaratne and Weerakoon (2012), the increase in cadmium accumulation reflects the ability of superior plants to transfer this element from the soil to the plant root system and then distribute it to various plant tissues. It also suggests that these plants have developed mechanisms to tolerate and withstand high levels of heavy metals.

**Table 3. Role of *Eucalyptus camaldulensis*, *Albizia lebbek*, *Ficus carica* and *Morus nigra* transplants on soil available lead (mg Pb.kg<sup>-1</sup> soil), removal rate (%) and BCF**

Pb content of initial soil was 10.77 mg.kg <sup>-1</sup> ( before)																
Treatments	<i>Eucalyptus camaldulensis</i>				<i>Albizia lebbek</i> ,				<i>Ficus carica</i>				<i>Morus nigra</i>			
	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF
H <sub>0</sub> S <sub>0</sub>	5.11	52.55	30.34	2.82	4.46	58.59	44.68	4.15	7.88	26.83	11.23	1.04	6.22	42.25	14.69	1.36
H <sub>0</sub> S <sub>250</sub>	5.00	53.57	32.78	3.04	4.44	58.77	45.25	4.20	7.88	26.83	11.77	1.09	6.22	42.25	15.30	1.42
H <sub>0</sub> S <sub>500</sub>	5.00	53.57	33.88	3.15	4.40	59.15	46.16	4.29	7.82	27.39	12.72	1.18	6.16	42.80	16.55	1.54
H <sub>5</sub> S <sub>0</sub>	4.96	53.95	35.74	3.32	4.37	59.42	51.92	4.82	7.80	27.58	12.71	1.18	6.00	44.29	17.18	1.60
H <sub>5</sub> S <sub>250</sub>	4.82	55.25	37.40	3.47	4.22	60.82	53.85	5.00	7.80	27.58	13.18	1.22	6.00	44.29	17.70	1.64
H <sub>5</sub> S <sub>500</sub>	4.79	55.52	40.16	3.73	4.02	62.67	59.35	5.51	7.75	28.04	14.60	1.36	5.92	45.03	19.15	1.78
H <sub>10</sub> S <sub>0</sub>	4.66	56.73	52.68	4.91	3.91	63.70	67.93	6.31	7.64	29.06	14.64	1.36	5.63	47.73	20.18	1.87
H <sub>10</sub> S <sub>250</sub>	4.50	58.22	53.81	5.00	3.80	64.72	70.77	6.57	7.63	29.16	16.77	1.56	5.63	47.73	22.35	2.08
H <sub>10</sub> S <sub>500</sub>	4.44	58.77	56.05	5.20	3.80	64.72	78.05	7.25	7.60	29.43	19.05	1.77	5.48	49.12	24.58	2.28
Mean	4.81	55.34	41.42	3.85	4.16	61.37	57.54	5.34	7.76	27.95	14.07	1.31	5.92	45.03	18.63	1.73

Since, H<sub>0</sub>: Without humic acid (control); H<sub>5</sub>: Spraying humic acid at rate of 5 g.L<sup>-1</sup>; H<sub>10</sub>: Spraying humic acid at rate of 10 g.L<sup>-1</sup>; S<sub>0</sub>: Without salicylic acid (control); S<sub>250</sub>: Spraying salicylic acid at rate of 250 mg.L<sup>-1</sup>; S<sub>500</sub>: Spraying salicylic acid at rate of 500 mg.L<sup>-1</sup>

**Table 4. Role of *Eucalyptus camaldulensis*, *Albizia lebbek*, *Ficus carica* and *Morus nigra* transplants on soil available cadmium (mg Cd.kg<sup>-1</sup> soil), removal rate (%) and BCF**

Cd content of initial soil was 2.44 mg.kg <sup>-1</sup> ( before)																
Treatments	<i>Eucalyptus camaldulensis</i>				<i>Albizia lebbek</i> ,				<i>Ficus carica</i>				<i>Morus nigra</i>			
	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF
H <sub>0</sub> S <sub>0</sub>	0.07	97.13	8.22	3.37	0.12	95.08	8.54	3.50	0.96	60.66	2.48	1.02	0.78	68.03	3.01	1.23
H <sub>0</sub> S <sub>250</sub>	0.07	97.13	8.25	3.38	0.15	93.85	8.57	3.51	0.92	62.30	2.52	1.03	0.70	71.31	3.06	1.25
H <sub>0</sub> S <sub>500</sub>	0.07	97.13	8.42	3.45	0.14	94.26	8.73	3.58	1.04	57.38	2.75	1.13	0.76	68.85	3.36	1.38
H <sub>5</sub> S <sub>0</sub>	0.06	97.54	10.02	4.11	0.12	95.08	10.43	4.27	0.92	62.30	2.65	1.09	0.72	70.49	3.12	1.28
H <sub>5</sub> S <sub>250</sub>	0.06	97.54	10.40	4.26	0.12	95.08	10.75	4.41	0.90	63.11	3.35	1.37	0.72	70.49	3.87	1.59
H <sub>5</sub> S <sub>500</sub>	0.05	97.95	10.83	4.44	0.10	95.90	11.24	4.61	0.90	63.11	3.92	1.61	0.70	71.31	4.11	1.68
H <sub>10</sub> S <sub>0</sub>	0.05	97.95	10.93	4.48	0.10	95.90	11.43	4.68	0.85	65.16	4.16	1.70	0.70	71.31	4.81	1.97
H <sub>10</sub> S <sub>250</sub>	0.04	98.36	11.21	4.59	0.08	96.72	11.64	4.77	0.88	63.93	4.68	1.92	0.66	72.95	5.76	2.36
H <sub>10</sub> S <sub>500</sub>	0.03	98.77	11.53	4.73	0.06	97.54	12.01	4.92	0.79	67.62	5.13	2.10	0.61	75.00	7.30	2.99
Mean	0.056	97.70	9.98	4.09	0.11	95.49	10.37	4.25	0.91	62.70	3.51	1.44	0.706	71.07	4.26	1.75

Since, H<sub>0</sub>: Without humic acid (control); H<sub>5</sub>: Spraying humic acid at rate of 5 g.L<sup>-1</sup>; H<sub>10</sub>: Spraying humic acid at rate of 10 g.L<sup>-1</sup>; S<sub>0</sub>: Without salicylic acid (control); S<sub>250</sub>: Spraying salicylic acid at rate of 250 mg.L<sup>-1</sup>; S<sub>500</sub>: Spraying salicylic acid at rate of 500 mg.L<sup>-1</sup>

**Soil available nickel (Ni) and removal rate at the end of the experiment**

The results presented in Table 5 provide information about the concentration of soil available nickel (Ni) and the biological concentration factor (BCF) of nickel in the tissues of four transplant species.

The concentration of soil available nickel was determined to be 121.36 mg Ni.kg<sup>-1</sup> soil. Significant differences were observed in the concentrations of available nickel in the soil after the experiment, which can be attributed to the planting of different transplant species. The lowest concentration was found in pots with *Albizia lebbek* transplants, measuring 14.34 mg Ni.kg<sup>-1</sup> soil, indicating a removal rate of 88.18%. On the other hand,

pots with fig transplants had a higher concentration of 58.18 mg Ni.kg<sup>-1</sup> soil, with a removal rate of 52.06%.

It is noteworthy that all BCF values in the tables for all plants were less than one. This indicates that these plants have a limited ability to accumulate nickel in their leaf tissues. However, it is possible that nickel may accumulate in other parts of the plants. The BCF values below one suggest that the transplanted species may not be efficient in accumulating nickel in their aboveground tissues. Further analysis of the plants' ability to tolerate and accumulate nickel in specific plant parts or their potential use in phytoremediation purposes can provide more insights into their effectiveness in nickel accumulation and removal from soil.

**Table 5. Role of *Eucalyptus camaldulensis*, *Albizia lebbeck*, *Ficus carica* and *Morus nigra* transplants on soil available nickel (mg Ni.kg<sup>-1</sup> soil), removal rate (%) and BCF**

Treatments	Ni content of initial soil was 121.36 mg.kg <sup>-1</sup> ( before)															
	<i>Eucalyptus camaldulensis</i>				<i>Albizia lebbeck</i> ,				<i>Ficus carica</i>				<i>Morus nigra</i>			
	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF	(mg.kg <sup>-1</sup> soil) after	Removal Rate (%)	Leaves contents (mg.kg <sup>-1</sup> )	BCF
H <sub>0</sub> S <sub>0</sub>	16.93	86.05	37.23	0.31	14.98	87.66	37.72	0.31	58.56	51.75	7.93	0.065	44.12	63.65	10.78	0.089
H <sub>0</sub> S <sub>250</sub>	16.83	86.13	37.82	0.31	14.80	87.80	38.44	0.32	58.42	51.86	8.19	0.067	44.00	63.74	11.53	0.095
H <sub>0</sub> S <sub>500</sub>	16.60	86.32	37.96	0.31	14.80	87.80	38.65	0.32	58.30	51.96	8.76	0.072	44.00	63.74	12.58	0.104
H <sub>5</sub> S <sub>0</sub>	16.54	86.37	37.97	0.31	14.66	87.92	38.56	0.32	58.30	51.96	9.00	0.074	43.83	63.88	11.94	0.098
H <sub>5</sub> S <sub>250</sub>	16.04	86.78	38.38	0.32	14.24	88.27	39.04	0.32	58.20	52.04	9.23	0.076	43.66	64.02	12.79	0.105
H <sub>5</sub> S <sub>500</sub>	16.00	86.82	38.88	0.32	14.20	88.30	39.72	0.33	58.20	52.04	10.15	0.084	43.10	64.49	14.10	0.116
H <sub>10</sub> S <sub>0</sub>	14.94	87.69	39.03	0.32	13.90	88.55	39.60	0.33	57.90	52.29	10.58	0.087	43.10	64.49	13.83	0.114
H <sub>10</sub> S <sub>250</sub>	14.66	87.92	39.54	0.33	13.90	88.55	40.62	0.33	57.90	52.29	11.16	0.092	42.96	64.80	14.89	0.123
H <sub>10</sub> S <sub>500</sub>	14.20	88.30	42.42	0.35	13.58	88.81	43.52	0.36	57.82	52.36	12.26	0.101	41.98	65.08	17.02	0.140
Mean	15.86	86.93	38.80	0.32	14.34	88.18	39.54	0.33	58.18	52.06	9.69	0.080	43.42	64.22	13.27	0.109

Since, H<sub>0</sub>: Without humic acid (control); H<sub>5</sub>: Spraying humic acid at rate of 5 g.L<sup>-1</sup>; H<sub>10</sub>: Spraying humic acid at rate of 10 g.L<sup>-1</sup>; S<sub>0</sub>: Without salicylic acid (control); S<sub>250</sub>: Spraying salicylic acid at rate of 250 mg.L<sup>-1</sup>; S<sub>500</sub>: Spraying salicylic acid at rate of 500 mg.L<sup>-1</sup>

### Discussion

The variation among different tree species can be attributed to inherent genetic differences and physiological characteristics of each species. Different tree species have unique growth patterns, growth rates, and genetic predispositions that influence their response to external factors, including treatment with humic acid or salicylic acid. The superior performance of *Albizia lebbeck* trees in removing the heavy metal might be due to their genetic makeup and inherent growth capabilities, whereas other trees may have lower growth rates or different growth strategies that result in less pronounced vegetative growth characteristics increase (Zakari and Audu, 2021).

On the other hand, the observed effects can be explained by the ability of both humic and salicylic acids to enhance heavy metals uptake and assimilation in plants. The variations among tree species reflect their inherent genetic differences and physiological characteristics. The interactions between humic acid spray, salicylic acid spray, and tree type further might influence the heavy metals uptake. When humic acid was applied as a foliar spray at a concentration of 10 g/L, it significantly increased the content of heavy metals in the leaves compared to the control treatment. Foliar application of humic acid can promote the absorption of heavy metals through the leaves (Al-Marsoumi and Al-Hadethi 2020). Humic acid contains functional groups that can chelate heavy metals, forming complexes that are more easily taken up by leaf tissues. This enhanced uptake of heavy metals from the surrounding environment contributes to the increased content observed in the leaves. It is important to note that foliar application of humic acid can also enhance the nutrient and water absorption efficiency of plants, leading to overall improved physiological conditions. This may indirectly contribute to increased heavy metals uptake in the leaves (Lim and Lim, 2012). Similarly, an increase in the spray concentrations of salicylic acid resulted in an increase in leaves' heavy metals content. Salicylic acid might influence heavy metals uptake and translocation through its impact on various physiological and biochemical processes in plants. It can modulate ion transporters and channels, affecting the uptake and translocation of heavy metals (Seenivasan *et al.* 2015). The

higher concentration of salicylic acid (500 mg) likely stimulated the absorption and accumulation of heavy metals in the leaves. The variation in heavy metal content in soil with different tree species, as *Albizia lebbeck* trees exhibiting lower soil content compared to other trees, can be attributed to inherent genetic differences and physiological characteristics. Different tree species have varying abilities to tolerate and accumulate heavy metals (Baker *et al.* 2020). Some species may have mechanisms to selectively absorb or sequester heavy metals in their tissues, resulting in higher accumulation levels (Zakari and Audu, 2021). On the other hand, other species may have lower capacities for heavy metals uptake or have efficient mechanisms for detoxification and exclusion, resulting in lower content in their leaves (Sharma *et al.* 2020). The variations among tree species reflect their inherent abilities to tolerate, accumulate, or exclude heavy metals (Yaashikaa *et al.* 2022; Ahmad *et al.* 2023).

### CONCLUSION

The results revealed that *Albizia lebbeck* transplants exhibited the most promising phytoremediation performance among the studied tree species. They demonstrated the lowest soil zinc concentration and removal rate, as well as the lowest levels of soil available lead, cadmium, and nickel concentrations with higher removal rates compared to the other species. Furthermore, the foliar application of humic acid and salicylic acid contributed to reducing the residues of zinc, cadmium, lead, and nickel in the soil. The roots of the trees exhibited increased uptake of these heavy metals as the concentrations of humic acid and salicylic acid increased. The most effective combination was observed when trees were treated with a combined application of humic acid at a rate of 10 g L<sup>-1</sup> and salicylic acid at a rate of 500 mg L<sup>-1</sup>.

Based on the findings of this study, the following recommendations can be made:

*Albizia lebbeck* shows significant potential for phytoremediation of oil residues-contaminated soils. Therefore, it is recommended to consider *Albizia lebbeck* as a preferred tree species for phytoremediation projects targeting heavy metal removal from such soils. The application of humic acid and salicylic acid via foliar treatment has proven

effective in enhancing the phytoremediation capabilities of trees. Future studies should further explore the optimal application rates and frequency of these substances to maximize their remediation potential. It is advisable to conduct additional research to investigate the long-term effects of phytoremediation using *Albizia lebbek* and other tree species. Long-term studies can provide more insights into the sustainability and durability of the phytoremediation process. Considering the positive impact of humic acid and salicylic acid on heavy metal uptake, further research should be conducted to explore other organic amendments or additives that can enhance the phytoremediation efficiency of trees. Field-scale trials should be conducted to validate the findings of this pot trial under real-world conditions. These trials can help assess the feasibility and practicality of implementing phytoremediation using *Albizia lebbek* and the recommended amendments on a larger scale. By implementing these recommendations, phytoremediation can become a valuable and sustainable approach for restoring degraded soils contaminated with oil residues and heavy metals, leading to improved environmental quality.

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## تقييم مقارنة لأنواع الأشجار في علاج التربة الملوثة بالمعادن الثقيلة: تأثير إضافة أحماض الهيوميك والساليسيليك

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### الملخص

من أجل تقييم فعالية النباتات في إعادة تأهيل التربة المتدهورة، وتحديدًا تلك الملوثة ببقايا النفط، أجريت تجربة أصص. كان الهدف الرئيسي منها هو مقارنة أداء النباتات في عملية إزالة التلوث بواسطة النباتات حيث تم استخدام أنواع مختلفة من الأشجار، وتحديدًا الكافور، الألبيزيا، التين، التوت الأحمر كعامل دراسة رئيسي. كما هدفت الدراسة أيضًا إلى تقييم تأثير معدلات مختلفة من حمض الهيوميك (٠، ٥، ١٠ جم / لتر) كعامل منشق أول، ومعدلات مختلفة من حمض الساليسيليك (٠، ٢٥٠، ٥٠٠ ملغ / لتر) كعامل منشق ثاني، على قدرة النباتات في إزالة العناصر الثقيلة، مثل الزنك والرصاص والكاديوم والنيكل من التربة الملوثة ببقايا النفط. أظهرت النتائج أن نباتات الألبيزيا تسببت في تركيزًا منخفضًا لعنصر الزنك في التربة (١٥,٦٢ ملجم زنك/كجم تربة) ونسبة إزالة قدرها ٧٨,٩٨٪. بالمثل، أظهرت نباتات ألبيزيا أقل مستويات لعناصر الرصاص والكاديوم والنيكل بالتربة مقارنة بالأنواع النباتية الأخرى المدروسة، مع نسب إزالة أعلى. علاوة على ذلك، أدت الإضافات الورقية لكل من حمض الهيوميك وحمض الساليسيليك إلى انخفاض المتبقي من الزنك والكاديوم والرصاص والنيكل. حيث زاد امتصاص هذه العناصر الثقيلة عن طريق الجذور مع زيادة تركيزات حمض الهيوميك وحمض الساليسيليك. تم تسجيل أدنى القيم على وجه التحديد عند معاملة الأشجار بحمض الهيوميك بمعدل ١٠ جم/لتر وحمض الساليسيليك بمعدل ٥٠٠ ملجم/لتر كمعاملة مشتركة.

**الكلمات الدالة:** المعالجة النباتية، حمض الهيوميك، الساليسيليك