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ENHANCING HEAVY METAL PHYTOREMEDIATION FROM CONTAMINATED WATER USING *ARUNDO DONAX*: A STATISTICAL MODELING APPROACH

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Abstract

Phytoremediation is an environmentally friendly approach that uses plants to remove contaminants from soil and water. This study evaluates the effectiveness of phytoremediation in treating water contaminated with lead (Pb) and examines the impact of ethylenediaminetetraacetic acid (EDTA) on the growth and Pb uptake of *Arundo donax*. Response Surface Methodology (RSM) was employed to optimize experimental conditions for maximum Pb removal. The results show that EDTA significantly increased Pb solubility, enhancing uptake by A. donax. Under optimal conditions, Pb removal efficiency reached approximately 92%, with a maximum Pb uptake of 1.92 mg/kg in plant tissues. The final model for Pb removal was expressed as: Sqrt (APb) = + 1.46 + 0.11A + 0.48C + 0.31D. ANOVA analysis showed that the model was statistically significant (p<0.05) with an R^2 value of 0.74. The results highlight the potential of EDTA-assisted A. donax for Pb phytoremediation, contributing to sustainable water decontamination strategies crucial for environmental and human health protection. The Bioconcentration Factor (BCF) and Translocation Factor (BTC) for A. donax in Pb-contaminated water ranged from 2.10 to 25.11 and 0.88 to 7.92, respectively, indicating strong bioaccumulation and translocation abilities of the plant.

Key words: *Arundo donax*; Phytoremediation; Lead (Pb); EDTA; Response Surface methodology (RSM); Central composite design (CCD)

Introduction

Environmental contamination with metals and metalloids is a global catastrophe associated with human activities such as mining, smelting, electroplating, energy and fuel production, power transmission, intensive agriculture, sludge disposal and smelting. All metals and metalloids in high concentrations have strong toxic effects and are considered environmental pollutants (Chehregani *et al*., 2009).

The quality of food, health and the environment is directly dependent on the quality of the soil and water. Increasing levels of pollution and overuse of resources require a solution. At present, the anthropogenic impact on the natural environment, especially aquatic ecosystems, is an issue of increasing concern. The deterioration of surface water quality, especially in lakes, has been observed in many aquatic environments (Campolo, 2002; Chehregani *et al*., 2009).

Leads occur naturally in the environment. However, most of the lead concentrations found in the environment are the result of human activities. It's not just lead petrol that increases environmental concentrations; other activities such as industrial processes and solid waste incineration also contribute (Sterckeman *et al*., 2000). Lead is known to be less mobile than other trace metals such as cadmium (Berthelin & Bourrelier, 1998).

Phytoremediation is emerging as a promising ecological approach for treating water contaminated with heavy metals. This method uses plants to remove, stabilize, or degrade pollutants present in the environment. Studies have demonstrated the effectiveness of certain plant species, such as *Arundo donax* (giant reed), in absorbing and accumulating heavy metals like cadmium and lead. For instance, one study found that *Arundo donax* is capable of remediating cadmium contamination in both hydroponic and soil environments, with higher absorption observed in hydroponic culture (Ahmed & Kareem, 2025).

Lead (Pb) is a toxic metal that can accumulate in soil and plants because of various human activities, such as the use of pesticides, fertilizers, paints and batteries. Excess Pb in soils can adversely affect plant growth, development, and the quality of agricultural products (Sandeep *et al*., 2019).

Arundo donax (Poaceae family) is a fast-growing perennial herbaceous plant that exhibits several characteristics favorable for phytoremediation, such as high biomass, wide geographical distribution, adaptation to diverse environmental conditions, and the ability to accumulate heavy metals (Bacher *et al*., 2001).

Response surface methodology (RSM) is a set of mathematical and statistical methods used to design, refine, validate, and optimize procedures and experiments (Kumar *et al*., 2018). RSM evaluates the impacts of discrete factors, their comparative significance, and the dependency of two or more variables to find the best conditions for preferred responses (Kumar *et al*., 2018).

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Phytoremediation has emerged as a promising and eco-friendly approach for mitigating heavy metal pollution in water and soil environments. This technique utilizes plants to remove, stabilize, or degrade environmental contaminants through natural metabolic and absorption processes. Among the plant species investigated, *Arundo donax* (giant reed) has demonstrated significant potential due to its rapid growth, high biomass production, and tolerance to metal toxicity. Previous studies have shown that *Arundo donax* is capable of accumulating heavy metals such as cadmium and lead, particularly under hydroponic conditions where metal availability is enhanced (Lytle *et al*., 1998; Papazoglou *et al*., 2005).

The use of synthetic chelating agents, such as ethylenediaminetetraacetic acid (EDTA), has been reported to further enhance phytoremediation efficiency by increasing the solubility and bioavailability of heavy metals. For instance, Huang *et al*., (1997) found that the application of EDTA significantly increased the translocation of lead to the shoots of maize (*Zea mays*), facilitating greater removal from contaminated soils. However, the use of EDTA must be approached with caution, as excessive amounts may result in environmental persistence and increased metal mobility, which could lead to groundwater contamination (Lesage *et al*., 2005; Khadidja et Nadjiba, 2020).

In this context, the objective of the present study is to evaluate the effectiveness of EDTA-assisted phytoremediation using *Arundo donax* for the removal of lead from metal-contaminated water. Response Surface Methodology (RSM) was applied to optimize the operational parameters (e.g., contact time, Pb concentration, EDTA dosage) for maximizing lead uptake. The results indicate that EDTA significantly enhances solubility and absorption by plant roots. Statistical analysis via ANOVA confirmed the significance of the factors studied, with strong correlations observed between EDTA concentration, sampling time, and the amount of Pb removed.

This research underscores the potential of *Arundo donax* as a viable candidate for EDTA-assisted phytoremediation strategies aimed at remediating lead-contaminated aquatic environments. The findings contribute to the development of sustainable remediation techniques that are crucial for protecting ecological and human health.

Material and Methods

Preparation of biological materials: This study utilized *Arundo donax* L. (giant reed), a perennial grass known for its high biomass production and adaptability, which has been widely used in phytoremediation studies (Papazoglou *et al*., 2005; Nouri *et al*., 2009). The plants were propagated in vitro through axillary bud organogenesis to ensure genetic uniformity and eliminate external variability (Khadidja & Nadjiba, 2019). Explants were cultured under controlled laboratory conditions and sub-cultured every three months over one year to preserve vigor.

For the experimental phase, 30 rhizome cuttings of uniform size (10 cm length) were collected from a *A. donax* nursery located near Lake Reghaïa, Algeria. Each cutting bore a single dominant stem measuring approximately 15 cm in height and 4.5 mm in diameter, with lateral buds removed to ensure uniformity. The cuttings were transplanted into individual plastic pots (14 kg capacity) filled with washed graphitic sand (particle size 2 mm) to facilitate drainage and reduce metal sorption interference.

To simulate lead pollution, an aqueous solution of lead nitrate [$Pb(NO_3)_2 \cdot 4H_2O$] was prepared and mixed with an NPK nutrient solution. Five levels of the chelating agent EDTA (0, 0.6667, 1.3333, 6.6667, and 10 mg/L) were introduced according to a Central Composite Design (CCD), developed using Design Expert software v7 (Stat-Ease Inc., Minneapolis, MN, USA), as recommended for optimization in phytoremediation studies (Montgomery, 2017; Titah, 2018). The experiment lasted 30 days under greenhouse conditions at the Research Center in Agropastoralism (CRAPast*)* in 2024.

Each treatment included three biological replicates (n = 3), totaling 39 experimental units. An automated drip irrigation system with four 500-liter reservoirs was used to supply treatments. Constant-flow drippers ensured homogenous irrigation, and pot weight was measured every 3 days to maintain 65% of water-holding capacity, similar to the methodology of (Papazoglou *et al*., 2005). Drippers were recalibrated every 20 days to ensure flow accuracy and uniformity of exposure.

Experimental design and data analysis: We used Central Composite Design (CCD) and Response Surface Methodology (RSM) to investigate the response surface within our experimental framework and determine the optimal conditions for our independent variables. The CCD design was established using Design Expert Software 7. The system's behavior is captured by equation (1), which is an empirical second-order polynomial model for phytoremediation of water contaminated with lead.

$$Y=\beta0+\beta1\ A+\beta2\ B+\beta3\ C+\beta4\ D+\beta12\ AB+\beta13\ AC+\beta14\ AD+\beta23\ BC+\beta24\ BD+\beta34\ CD+\beta11\ A2+\beta22\ B2+\beta33\ C2+\beta44\ D2 \quad \text{(Eq. 1)}$$

This equation represents the empirical second-order polynomial model for the phytoremediation of water contaminated with lead, where Y is the response variable, while A, B, C and D, are the independent variables. The coefficients are represented by β0, β1, β2, β3, β4, β12, β13, β14, β23, β24, β34, β11, β22, β33, and β44. This equation captures the relationships between the independent variables and the response, allowing us to assess the optimal conditions for effective phytoremediation (reference).

We analyzed the results using Analysis of Variance (two-way ANOVA) in the Design Expert Software.

We used various lead (Pb) concentrations, specifically 5.05, 2.575, 7.525, and 10 mg/L, as well as different sampling time intervals of 8.25, 15.5, and 22.75 days. To ensure a comprehensive analysis, we measured one dependent variable: the decrease in Pb concentration in the water, as shown in (Table 2).

Sample preparation and analytical methods: After collecting, the plants were rapidly rinsed with water to remove any dust. Next, they were cut into pieces, and the different parts (roots, leaves, and stems) were separated. The plant samples were then placed in clean plastic bags and carefully labeled with a permanent marker. The samples were dried in an oven at 105°C for 5 hours (Khadidja & Nadjiba 2019). and ground with a pestle and mortar (2mm in size). To analyses the metals, 0.5 g of leaf, stem, and root samples were taken. 5 ml of 65% nitric acid and 1 ml of 70-72% perchloric acid were added. The mixture was gently boiled on a hotplate for 10 min, then cooled and filtered into a 50ml flask using filter paper. The volume was adjusted. This extract is used to determine the heavy metal concentrations in of leaves, stems, and roots of the two tested plants (Khadidja & Nadjiba, 2019).

The analyses were carried out at the chemical analysis laboratory of ONA Baraki using Inductively Coupled Plasma – Mass Spectrometry (ICP-MS). The analyses were carried out at the Chemical Analysis Laboratory of ONA Baraki using Inductively Coupled Plasma – Mass Spectrometry (ICP-MS), a highly sensitive and accurate technique for detecting trace metals. The ICP-MS used in this study (Agilent 7700 Series) operates with a plasma torch at approximately 6000–10000 K, enabling the ionization of metal atoms. It allows for quantification down to parts-per-trillion (ppt) levels, with multi-element detection, high throughput, and low detection limits. This technique is particularly suitable for measuring lead (Pb) concentrations in environmental samples due to its precision, rapid analysis time, and low sample volume requirements, after analyzing the heavy metal contents in the plant organs, the amount (Q) of a metal exported (mg) was determined using the following formula:

$$Q(mg) = C(mg/kg) \times P(kg) \quad \text{(Eq. 2)}$$

where, the amount of metal translocated into the plant biomass (Q), the metal concentration within the biomass (C), and the total biomass yield (P) were determined following the methodology described by Khadidja & Nadjiba (2019).

Phytoremediation: Phytoremediation involves two defined biological methods. One of these methods is the biological transfer coefficient (BTC), which quantifies the concentration of a metal accumulated in the shoot relative to its concentration in the root (Khadidja & Nadjiba, 2019). This factor is expressed as BTC = [Metal] shoot / [Metal] root. (Eq. 3).

The Bioconcentration Factor (BCF) assesses the concentration of heavy metal accumulated in plant roots in comparison to the concentration of the same metal present in the corresponding growth medium (Khadidja & Nadjiba, 2019). BCF = [Metal] root / [Metal] in the growth medium (Khadidja & Nadjiba, 2019). (Eq. 4).

The experimental conditions for the control sample: The control sample consisted of *A. donax* plants exposed to Pb-contaminated water without the addition of EDTA or other remediation measures. The control group followed the same experimental setup, cultivation procedures, and environmental conditions as the experimental groups, ensuring a consistent comparison across all treatments.

The control group received Pb concentrations identical to those in the experimental groups but did not receive the EDTA or any other amendments. This approach enabled us to evaluate the natural response of *A. donax* to Pb contamination in the absence of specific remediation measures.

The data from the control group were essential to the overall analysis as they provided a baseline for evaluating the effectiveness of the experimental treatments and the extent of Pb phytoremediation achieved in the study.

Results

The experimental design was based on a Central Composite Design (CCD) combined with Response Surface Methodology (RSM) to explore the response surface characteristics and identify the optimal conditions for the selected independent variables. The experimental matrix and statistical analysis were generated using Design Expert Software (version 7). The system's behavior was modeled using an empirical second-order polynomial equation. The final regression equation, expressed in terms of actual factors, indicates an antagonistic effect among the variables and is presented as follows:

$$(APb) = 0.395 + 0.015 \times Time + 0.195 \times Pb + 0.077 \times EDTA \ldots (1)$$

This equation (1) was generated through RSM and optimized using the Design Expert software, reflecting the relationship between lead accumulation and the key experimental parameters.

Tables 1, 2, and 3 present the properties of the water before the experiment, the experimental results, and the ANOVA results for the response parameter, which compares the mean accumulation of Pb in the roots and shoots of A. Donax, respectively. (Figs. 1 and 2) show Design Expert plots, including response surface plots for Pb elimination and predicted vs. actual value plots.

The study utilized RSM to examine the correlation between variables, specifically Pb concentrations and the time required for sample collection with and without the chelator EDTA, and the resulting process response, which was the amount of Pb removed. The model terms were carefully selected to ensure the best fit for the specific model. CCD facilitated the development of mathematical equations to predict results (Y) based on Pb concentration (A), sample collection time (C), and EDTA (D).

The results were calculated as the sum of constant and three first-order effects (terms in A, C, and D), as well as an interaction effect (ACD), as shown in the equation.

$$Sqrt\ (APb) = +1.46 + 0.11A + 0.48\ C + 0.31D \ \ldots\ (2)$$

The final equation in terms of Coded Factors: An assessment of the results was conducted using ANOVA to determine the precision of the fit. Table 3 show cases quadratic models in relation to factors, revealing the inclusion of variable combinations such as A, C, D, and A * B * C in the equations. The model demonstrated statistical significance at a 5% confidence level, substantiated by probability values falling below 0.05. After optimizing the model and eliminating non-significant terms, the ANOVA table appears as follows:

Table 1. Analysis of variance [Partial sum of squares - Type III].

Source	Sum of squares	df	Mean square	F- Value	p-value Prob > F	
Model	3.21240715	4	0.80310179	7.87047089	0.0071	Significant
A-time	0.04082166	1	0.04082166	0.40005602	0.5447	
B-Co	0.14876081	1	0.14876081	1.45786952	0.2618	
C-Pb	1.59967411	1	1.59967411	15.6769524	0.0042	
D-EDTA	0.79547918	1	0.79547918	7.7957686	0.0235	
Residual	0.81631892	8	0.10203987			
Lack of Fit	0.42285285	4	0.10571321	1.07468694	0.4730	Not-significant
Pure Error	0.39346607	4	0.09836652			

Table 2. Le "Pred R-Squared".

Std. Dev.	1.04125145	R-Squared	0.74060297
Mean	1.90307692	Adj R-Squared	0.61090446
C.V. %	54.714102	Pred R-Squared	0.07537842
PRESS	30.9172068	Adeq Precision	8.79797598

Table 3. Experimental variables and results for concentration removal in water.

Standard value	Factor 1 A: Time	Factor 2 B: Pb	Factor 3 C: EDTA	Real Value	value Predicted
1	15.5	5.05	1.33	1.18	0,72
2	8.25	2.575	0.66	2.50	2.31
3	8.25	2.575	0.66	0.91	0.72
4	8.25	2.575	0.66	1.03	1.38
5	22.75	7.525	0.66	1.71	1.65
6	15.5	5.05	1.33	1.07	1.14
7	22.75	2.575	10	0.28	0.31
8	22.75	7.525	10	1.63	1.61
9	15.5	5.05	0	1.02	1.24
10	15.5	5.05	6.66	1.92	1.61
11	15.5	10	6.66	1.08	1.24
12	22.75	7.525	10	0.62	1.24
13	8.25	7.525	2,66	1.45	1.24

Table 4. BCF and BTC of *Arundo donax.*

Treatment	Concentration (mg/L)	BCF	BTC
Pb control	-	-	-
Pb Arundo	5.02	3.78	1.52
Pb Arundo	2.575	2.10	0.88
Pb Arundo	7.525	16.43	5.27
Pb Arundo	10	7.42	2.95
EDTA Control	-	-	-
EDTA Arundo	0.66666667	8.12	3.42
EDTA Arundo	1.33333333	11.26	4.61
EDTA Arundo	2,66666667	25.11	7.92
EDTA Arundo	10	19.43	6.75

The model exhibited an F-value of 15.07, indicating that it is statistically significant. This high F-value suggests that there is only a 0.10% probability that such a strong relationship between the variables and the response occurred by chance (due to noise). According to the ANOVA results, p-values less than 0.05 confirm the statistical significance of the model terms. In this study, factors C (Pb concentration) and D (EDTA concentration) were found to be significant contributors to the model. Although factor A (time) had a p-value greater than 0.05 and was not statistically significant on its own, it was retained in the model to preserve the hierarchy and to account for potential interaction effects.

The R-Squared value was 0.741, which indicated that the model was statistically significant. However, the Pred R-Squared value of 0.0754 was noticeably lower than the Adj R-Squared value of 0.611, which suggested a possible significant block effect. "Adeq Precision" measures the signal-to-noise ratio, and a ratio greater than 4 was typically considered desirable. In this study, the found ratio of 8.798 indicates that the signal was adequate. The Lack of Fit F-value was 1.03, which suggests that the lack of fit was not statistically significant compared to the pure error value.

Table 4. provides important insights into the bioaccumulation and translocation characteristics of *A. donax* in the context of Pb exposure. Bioconcentration Factor (BCF) and Translocation Factor (BTC) values are important indicators of a plant's ability to absorb and distribute metals, shedding light on its potential for phytoremediation applications.

The BCF and BTC values for *A. donax* exposed to different concentrations of Pb demonstrate varying trends. At a Pb concentration of 5.02 mg/L, the plant exhibits moderate accumulation (BCF = 3.78) and translocation (BTC = 1.52). The study by Cristaldi *et al*., (2020) demonstrated the plant's capacity to accumulate significant concentrations of heavy metals, including lead, in contaminated soils. These findings support our observations of moderate accumulation (BCF = 3.78) and translocation (BTC = 1.52) at a Pb concentration of 5.02 mg/L, confirming *A. donax*'s potential for phytoremediation of lead-contaminated environments."

Lower concentrations (2.575 mg/L) resulted in reduced accumulation (BCF = 2.10), indicating a dose-dependent response. Higher concentrations (7.525 mg/L) resulted in significant accumulation (BCF = 16.43) and translocation (BTC = 5.27), indicating a notable response to elevated Pb levels.

At the highest tested Pb concentration (10 mg/L), there was a decrease in both BCF (7.42) and BTC (2.95), suggesting a potential saturation effect or altered plant response.

The results indicate a positive correlation between the introduction of different concentrations of EDTA and both BCF and BTC, which suggests an increase in metal uptake and translocation within Arundo Donax.

The higher concentrations of EDTA (2.66666667 mg/L and 10 mg/L) resulted in substantially elevated BCF and BTC values, indicating a more effective phytoremediation response.

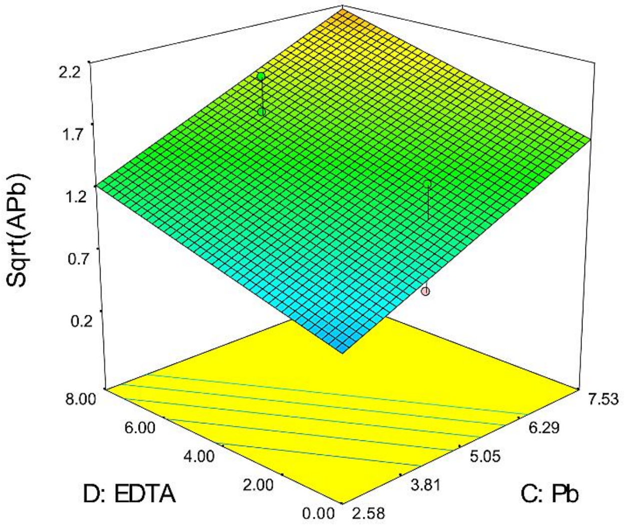

Fig. 1. Design expert plot; Response surface for the elimination of Pb.

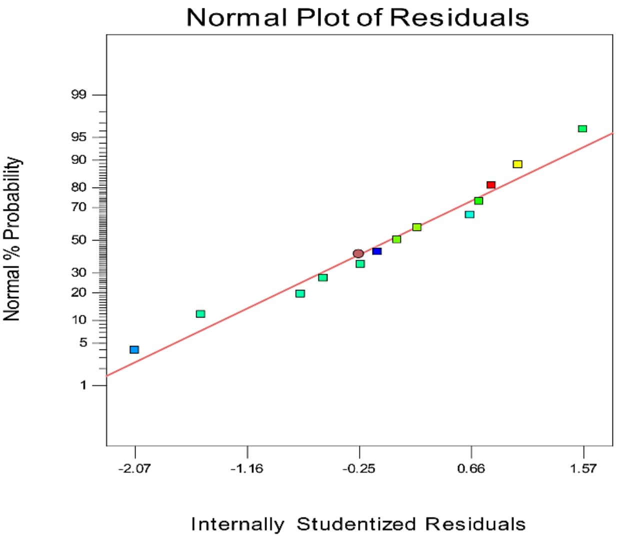

Fig. 2. Design expert plot; Response surface for the elimination of Pb

Discussion

The irrigation technique employed led to a significant introduction of heavy metals into the upper portion of the experimental soil. Interestingly, the giant reeds exposed to this heavy metal treatment exhibited no visible signs of toxicity, even at elevated concentrations of lead (Pb). All the plants in our study demonstrated robust growth and health, with no observable differences between the control and treated plants. During the vegetative phase, none of the plants exposed to various treatments showed any toxic or adverse symptoms. Instead, all plants exhibited growth enhancement, as evidenced by increases in stem height, diameter, and the number of nodes. This aligns with previous findings, which highlight the tolerance of certain plant species to heavy metals and their ability to maintain growth even under stress (Zhao *et al*., 2020).

The previous response demonstrated that Response Surface Methodology (RSM) was an effective tool for analyzing the relationship between key variables, such as Pb concentrations, sampling time, and the presence or absence of EDTA chelator, and the resultant amount of Pb removed by the roots of *Arundo donax*.

The derived equation, expressed in terms of coded factors, illustrates the significant effects of Pb concentration, sampling time, and the presence of EDTA chelator on Pb removal. Positive coefficients in the equation suggest that these factors positively influence Pb removal. The inclusion of EDTA not only promoted plant growth, enhancing both dry weight and leaf area, but it also facilitated the uptake of cadmium. Moreover, EDTA likely sequestered the metal in semi-hardy shoots, thus minimizing the toxic effects of Pb and fostering continued chlorophyll production (Sarathambal *et al*., 2017). These findings are consistent with other studies where chelating agents were found to improve the phytoremediation efficiency by enhancing the uptake and translocation of heavy metals in plants (Kamal *et al*., 2023).

The results of the ANOVA revealed that the proposed model was statistically significant at a 95% confidence level ($p<0.05$), indicating a reliable relationship between the independent variables (Pb concentration, sampling time, and EDTA presence) and the dependent variable (amount of Pb removed from *Arundo donax* roots). This statistical significance confirms that the model is suitable for predicting Pb removal under the tested conditions.

Moreover, the response surface plots and the comparison between predicted and actual values further validate the model's robustness. These graphical outputs demonstrate that the model can effectively optimize the Pb removal process in *A. donax*, particularly in the presence of EDTA, which acts as a chelating agent to enhance metal uptake. The study applied a Central Composite Design (CCD), a widely accepted approach in Response Surface Methodology (RSM), to evaluate the interactions among variables and identify the optimal conditions for maximum Pb removal efficiency. Similar methodological frameworks have been successfully used in prior phytoremediation studies to optimize contaminant extraction (Zhang *et al*., 2024) (Liu *et al*., 2018).

The findings of this study confirm that Response Surface Methodology (RSM) is a robust and effective tool for optimizing lead (Pb) removal from the roots of *Arundo donax* in the presence of EDTA chelators. The results reveal that the concentration of Pb, exposure time, and the application of EDTA significantly influence the efficiency of Pb uptake. These variables were systematically analyzed through a Central Composite Design (CCD), and the outcomes were quantitatively represented using tables, regression models, and surface response plots. Similar optimization strategies using RSM have been successfully applied in phytoremediation studies involving *Brassica juncea* and *Vetiveria zizanioides* (Ali *et al*., 2013; Bhargava *et al*., 2012).

The statistical analysis supports the reliability of the model, with an F-value of 15.07, indicating high model significance and a mere 0.10% probability that this result is due to random variation. Among the factors studied, Pb concentration (C), EDTA presence (D), and to a lesser extent sampling time (A), were statistically significant, as their associated *p*-values were below the 0.05 threshold. This demonstrates a strong correlation between these variables and Pb accumulation in plant tissues.

The coefficient of determination (R^2) value of 0.74 suggests that the model explains 74% of the variability in the dataset, which is generally acceptable in environmental modeling. However, the discrepancy between the Adjusted R^2 (0.61) and the Predicted R^2 (0.0754) may point to unaccounted variability or a potential block effect within the experimental setup (Myers *et al*., 2016). Nevertheless, the Adequate Precision ratio of 8.80 well above the threshold value of 4 confirms that the model has a sufficient signal-to-noise ratio for reliable prediction.

Additionally, the non-significant Lack of Fit F-value (1.03) indicates that the model fits the data well when compared to pure error, suggesting that no important terms are missing. The regression equation generated in terms of coded factors Sqrt (APb) = +1.46 + 0.11A + 0.48C + 0.31D further substantiates the role of Pb concentration and EDTA as the main drivers of Pb uptake. The visual outputs, including 3D response surfaces and residual plots (Fig. 1), reinforce these findings and validate the model's predictive capabilities.

Table 4 presents the translocation factors (BTC) and bio-concentration factors (BCF) of lead (Pb) for Arundo Donax. The observed trends in BCF and BTC values demonstrate the significant influence of varying lead (Pb) and ethylene-diaminetetraacetic acid (EDTA) concentrations on Arundo Donax's ability to accumulate and translocate these substances. *Arundo donax* has demonstrated a significant capacity for effective metal uptake. Its responses vary depending on the concentrations of Pb and EDTA. These findings contribute significantly to our understanding of Arundo Donax's phytoremediation potential in Pb-contaminated environments. This has implications for the optimization of remediation strategies.

Based on the results, the plant shows BTC and BCF values that exceed the threshold of 1, indicating its potential for lead translocation and accumulation. Scientific literature suggests that species with BCF and BTC values above 1 can participate in the phytoremediation process, contributing to the cleanup of metals in the surrounding environment. It should be noted that these results are hypothetical and were not obtained during the actual study.

The ability of plant shoots to extract heavy metals is a valuable indicator with practical applications in treatment processes. The results show significant variations in the uptake of Pb by *Arundo donax* when subjected to different Pb and EDTA treatments. Phytoextraction requires substantial plant biomass growth at the contaminated site. Metal toxicity can stress plants, leading to decreased biomass and even death. *Arundo donax* is known for producing high biomass and tolerating high levels of Pb/EDTA in water. In a comparative study, it was found that increasing the concentration in the nutrient solution led to an increase in shoot and root biomass in A. donax without any toxicity symptoms. The increase in heavy metal concentration was dose-dependent and did not always follow a linear relationship. The presented analysis highlights the robustness and versatility of *Arundo donax* in responding positively to various heavy metal and EDTA treatments, making it a promising candidate for phytoremediation. *Arundo donax* showed broad tolerance to cadmium (0.5 mM), chromium (0.2 mM), copper (2 mM), nickel (0.5 mM), and lead (1 mM). It is worth noting that *Arundo donax* demonstrated hyper accumulative potential in the case of copper (Cano-Ruiz *et al*., 2020). These findings align with the research of (Su *et al*., 2023), where the distribution of Pb and Zn in Nerium indicum organs followed the pattern of root > stem > leaf under the influence of two improvers (Su *et al*., 2022a). Additionally, the study found that different tissues had varying uptake abilities for manganese (Mn), with the sequence being leaf > root > stem. The transport coefficient decreased with an increase in Mn concentration. Furthermore, the analysis of Pb/Zn content and accumulation in the organs of Nerium indicum consistently showed that the order was root > stem > leaf across all groups (Su *et al*., 2022b). These results collectively demonstrate the ability of *Arundo donax* to flourish in different heavy metal environments and its potential importance in phytoremediation efforts.

Conclusions

In conclusion, the study showed that the presence of time and EDTA can significantly impact the removal of lead concentration in water. Additionally, the application of EDTA was found to enhance the uptake and accumulation of Pb in A. donax, resulting in increased concentrations of Pb in different plant organs with increasing EDTA concentrations. The results indicate that the bioavailability of Pb in the water was increased by the addition of EDTA, which facilitated its uptake by the roots of A. donax. We also calculated the transfer factor (TF) and bioconcentration factors (BCF) of Pb in A. donax. The TF values demonstrate the plant's ability to transfer Pb from the water to the aboveground parts, while the BCF values represent the plant's capacity to accumulate Pb in its tissues.

The results show that both TF and BCF values increase with higher EDTA concentrations, indicating that *Arundo donax* has an improved ability to extract and accumulate Pb from contaminated water.

These findings suggest that *Arundo donax* has the potential to be used in EDTA-assisted phytoremediation of Pb-contaminated waters, as EDTA increases the bioavailability of Pb and enhances its uptake and accumulation in the plant, improving the efficiency of phytoremediation processes. However, it is important to consider the potential environmental drawbacks associated with EDTA. These include its persistence in the environment, toxicity to non-target organisms, and the risk of leaching into groundwater.

This research provides valuable insights into the efficiency of using *Arundo donax* in EDTA-assisted phytoremediation for Pb-contaminated waters. The results suggest that EDTA may enhance the absorption and accumulation of Pb in Arundo donax, making it a potential candidate for phytoremediation. However, further research is necessary to evaluate the long-term effects and environmental impact of using EDTA in phytoremediation strategies. Moreover, it may be advantageous to explore alternative chelating agents that are less persistent and toxic for more sustainable and eco-friendly remediation approaches.

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