Prediction Models for Evaluating the Uptake of Heavy Metals by the Invasive Grass *Vossia cuspidata* (Roxb.) Griff. in the River Nile, Egypt: A Biomonitoring Approach

Emad A. Farahat 1,*, Waleed F. Mahmoud 2, Hosseim E. A. Awad 2, Hussein F. Farrag 2, Muhammad Arshad 3, Ebrahim M. Eid 4,5, and Gamal M. Fahmy 2

Abstract: This study aimed to develop new prediction models that include sediment properties (pH, organic matter, and silt and clay concentrations) for estimating the potential uptake of heavy metals (HMs) by the invasive grass *Vossia cuspidata*. Plant and sediment samples were collected from the microsites that represent the natural distribution of the species in two Nile islands in Cairo, Egypt. The results show that the root was the main accumulating organ for the analyzed HMs (Fe, Mn, Zn, Cu, Ni, and Pb). The mean concentrations of Fe and Mn and the maximum concentrations of Cu, Ni, and Pb were phytotoxic. The values of the bioconcentration factor were >1, while the translocation factor was >1 for Zn and Cu in rhizome and stem, Mn in leaf, and Ni and Pb in stem and leaf. There were no significant differences between the measured and the predicted HM concentrations in all organs of the species. This indicates the excellent robustness of the developed regression models. Sixteen equations (out of 24) had high $R^2$ values. Thus, *V. cuspidata* could be considered a biomonitor for HM pollution, and the developed equations will benefit the prediction of HM uptake by the species in the River Nile ecosystem.

Keywords: hippo grass; phytoremediation; heavy metals; prediction models; bioindicator; water pollution

1. Introduction

The pollution of terrestrial and aquatic ecosystems by heavy metals (HMs) is attributed to the world’s fast industrialization and urbanization [1]. HMs have negative consequences for the ecosystem, such as pollution and resource depletion [2]. Due to their solubility and mobility, these metals can accumulate in the living organisms, water, and sediment or soil leading to an environmental problem [3]. The bioavailability of HMs to plants is controlled by the adsorption and desorption processes in soil. The adsorption and desorption processes are affected by many other soil factors such as soil pH, organic matter content, silt and clay concentrations, and Fe and Mn oxides [4,5]. For instance, the bioavailability and mobility of HMs in the soil are negatively correlated with high soil pH values [6,7]. In addition, lower percentages of organic matter (OM) and clay in the sediment of *Typha domingensis* were associated with high metal bioavailability and uptake by the plant organs [8].
Aquatic macrophytes are used as a promising green technology to phytoremediate the contaminated watercourses from the HMs [9–12]. Obtaining field data on the uptake and accumulation of HMs by plants is expensive and laborious work. Hence, the establishment of regression models that build relationships between the available HMs in different organs of the plant species and the main soil or sediment variables, e.g., pH, OM, and silt and clay concentrations, is appropriate [7,13]. Some investigators used the bioaccumulation factor (as the content of a metal in plant/its content in soil) to estimate the HM accumulation ability of plants but not to forecast the soil–plant translocation of HMs in different types of soils [14]. Consequently, prediction regression models including soil variables may be beneficial for estimating HM concentrations in aquatic plants [15]. In addition, to choose suitable plants for phytoremediation of aquatic ecosystems, data on their HM accumulation capacity are necessary for developing the regression models [12].

Vossia cuspidata (Roxb.) Griff. (Hippo grass, family Poaceae) is an emergent macrophyte [16]. It is typically found in tropical rainforests such as Southeast Asia and Africa [16]. This species is a new invasive plant in Egypt, having been found in a variety of aquatic and riparian habitats in Cairo and the Nile Delta [17,18]. Before Boulos [19] in 1995, the species had never been recorded in any Egyptian flora or checklist. It reproduces only by rhizomes and spreads horizontally on the water’s surface or along the riverbanks (Shehata, 1996). V. cuspidata can resist drought, high pH, loamy soil, and waterlogged areas [20]. It reaches a height of 3 m above the water surface. In the last decade, V. cuspidata has invaded the River Nile ecosystem leading to a decrease of the native species by about 72% in some places [21]. The distribution of the species is restricted mostly to the main River Nile, its branches (Rosetta and Damietta), and main lateral wide and deep-water canals [17,22]. Galal et al. and Farahat et al. [18,23] studied the phytostabilization potential of V. cuspidata in the polluted water canals and the mainstream of River Nile, Egypt, respectively. They reported that this species had its maximum concentrations of the estimated HMs in their roots, with the transfer of some metals to other plant organs.

To our knowledge, this is the first work to create HM uptake prediction models for V. cuspidata growing in the River Nile ecosystem. The present study is an extension of our previous work on the same species [23]. The main goal of this study was to develop prediction models that include sediment properties (pH, OM, and silt and clay concentrations) for estimating the potential uptake of HMs by V. cuspidata. We think that these models will be useful tools for further ecological studies on the species in the River Nile ecosystem.

2. Materials and Methods

2.1. Study Area

The present study was conducted in two Nile islands, named Bin El Bahrin and Warraq Island, in Cairo, Egypt (latitudes between 30°06′43.47″ and 29°58′45.31″ N, longitudes between 31°13′12.41″ and 31°13′16.30″ E, Figure 1). V. cuspidata grows in the River Nile ecosystem in several microsites along the riverbanks. These sites are known as terraces, slopes, water edges, and open water [21]. The study areas are characterized by hot summer (the maximum air temperature was 33.3 °C in June) and mild winter (the minimum air temperature was 6.2 °C in February). The total yearly precipitation in the study area was less than 60 mm (Egyptian Meteorological Organization, 2020).
2.2. Plant Sampling and Analysis

Plant sampling from the two Nile islands was carried out monthly during one growing season (February 2018 to January 2019) from pure or nearly pure stands of *V. cuspidata*. For sampling, three sites on each island were assigned to represent the plant’s apparent microsites (slopes, water edges, and open water). At each sampling site, the monthly data of the roots, rhizomes, stems, and leaves of *V. cuspidata* were obtained from three randomly distributed quadrats (0.5 m × 0.5 m). The roots and rhizomes were taken at a depth of 0.5 m and washed with Nile water to get rid of sediment. Monthly, quadrats were assigned to be <2 m distance from that previously sampled.

In the laboratory, samples were carefully cleaned with deionized water, sorted into different organs, and then oven-dried at 70 °C until constant weight. Twelve composite samples (six per each island) from each organ of *V. cuspidata* were taken from oven-dried samples each season for chemical analysis (total *n* = 48). One gram was digested in a 15 mL acid mixture of HNO$_3$: HCl (1:1, *v/v*) for each dry and milled sample until a transparent solution was produced. The plant digests were then filtered and diluted to a known volume using deionized water. Microwave Plasma Emission Atomic Spectrometer (Agilent 4210 MP-AES, Agilent Technologies Inc., USA) was used to measure the concentrations of Fe, Mn, Zn, Cu, Ni, and Pb. The manufacturer’s user guide was used to modify the instrument settings and operational instructions. Concentrations of the elements were expressed as mg/kg dry weight.

2.3. Sediment Sampling and Analysis

Twelve composite sediment samples were collected seasonally from the same sampled microsites of the plant from the top of the sediment down to 50 cm, then air-dried and sieved through a 2 mm sieve. Then, sediment samples were mechanically and chemically analyzed, and the following assays were carried out: electrical conductivity (EC) was determined in the sediment paste extract using an electrical conductivity meter (Corning, model 441) and pH values by pH meter (model Orion 9107 BN) in 1:2.5 sediment–water suspension [24]. For additional chemical investigation, sediment–water extracts (1:5, *w/v*) were made [25]. The above-mentioned Agilent 4210 MP-AES was used to analyze the filtered supernatant samples for Fe, Mn, Zn, Cu, Ni, and Pb. The concentrations of sediment samples were expressed in mg/kg dry weight.
2.4. Data Analysis

The data from all the locations were combined and utilized to create a replica. The Shapiro–Wilkinson test ($p = 0.58$) was used to check the data for normality of distribution and homogeneity of variance at the beginning of the analysis. The minimum, maximum, mean, and coefficient of variance (CV) for sediment samples were calculated. To test the significant variation among organs of $V. cuspidata$, ANOVA-1 analysis followed by post hoc tests (Tukey’s HDS) was applied on the mean concentrations of each metal. The ability of $V. cuspidata$ to accumulate HMs from sediment was estimated by the bioconcentration factor (BCF). In addition, the potential translocation of HMs from roots to other plant organs was calculated by the translocation factor (TF) [11,26]. The BCF was calculated as the ratio of HM content in the roots (mg/kg) to that metal in the corresponding sediment (mg/kg). The plant is described as an accumulator to a certain metal when the value of BCF is $> 1$.

Pearson correlation coefficient test was conducted between the HM content of different plant organs and the same HM in sediment, in addition to sediment pH, OM (%), and silt and clay concentrations (%). The sediment pH, OM, silt and clay concentrations, and HM concentrations are the key factors that could be used for explaining the concentrations of HMs in plants [8]. Thus, for many plants, these soil variables were used for developing of the regression equations that were used for prediction of HM concentrations in plants [7,8,15,28].

The basic equation of the model was as follows:

$$C_{\text{plant}} = a + (b \times C_{\text{sediment}}) + (c \times \text{pH}) + (d \times \text{OM}) + (e \times \text{Silt}) + (f \times \text{Clay})$$

where $C_{\text{plant}}$ is the content of a given HM in the organs of $V. cuspidata$; $C_{\text{sediment}}$ is the available HM content in the sediment; OM is the sediment OM content (%); silt and clay are the sediment silt and clay concentrations (%); and $a$, $b$, $c$, $d$, $e$, and $f$ are the regression coefficients. For cross-validation of the models, 12 datasets were randomly selected for each of root, rhizome, stems, and leaves. The other 36 datasets were used to create the regression models to predict the levels of HMs in each of the organs of $V. cuspidata$ as dependent variables based on the measured sediment variables (pH, OM, silt, clay, Fe, Mn, Zn, Cu, Ni, and Pb) as independent variables. The model quality was evaluated based on the coefficient of determination ($R^2$); model efficiency (ME); model strength (i.e., the mean normalized average error, MNAE); and the model bias (i.e., the mean normalized bias, MNB). The calculations of these values were according to Novotná et al. [6]. The resulting regression equations were used to estimate the HM concentrations of the validation dataset ($n = 12$ for each organ). The deviations of the predicted content of a metal in an organ from the measured metal in the same organ were assessed using Student’s $t$-test. SPSS was used to conduct statistical analysis (version 22.0, SPSS Inc. [29]).

3. Results

The analysis of the sediment in the study area showed that silt and clay fractions represent 33.2% and 3.9% of its mechanical analysis. The pH of the sediment was slightly alkaline with a maximum value of 7.9, while the mean OM was 0.44%, with a maximum value of 1.1% and CV = 87.7% (Table 1). The mean concentrations of HM were $4.45$, $0.61$, $0.37$, $0.09$, $0.40$, and $0.45$ mg/kg for Fe, Mn, Zn, Cu, Ni, and Pb. The HMs in the sediment occurred in the following order: Fe $> \text{Mn} > \text{Ni} > \text{Pb} > \text{Zn} > \text{Cu}$.

Table 2 shows the mean concentrations of the investigated HMs in the organs of $V. cuspidata$. The mean Fe and Mn concentrations were significantly different among the organs of the plant ($F$-value $= 10.6$ and 40.8 at $p < 0.001$, respectively). The mean content (mg/kg) of Fe in different organs followed the order: root (2527.4) $> \text{leaf}$ (772.3) $> \text{stem}$ (700.1) $> \text{rhizome}$ (445.3). The root had a phytotoxic content of Fe (i.e., >$1000$ mg/kg) that was significantly different from other organs. In addition, Mn concentrations were in the order root $> \text{leaf} > \text{rhizome} > \text{stem}$ and at a phytotoxic level in the root (i.e., >$400$ mg/kg). Zn recorded considerable low concentrations in different organs of the plant with a maximum
content of 75.6 mg/kg in the stem. Cu concentrations were in the normal range (i.e., <20 mg/kg), but the recorded maximum concentrations in the root, rhizome, leaf, and stem were phytotoxic (43.8, 35.3, 36.7, and 75.6 mg/kg, respectively). The same observation was true in the case of Ni and Pb, where the maximum content values were slightly above the minimum phytotoxic value (i.e., 40 and 30 mg/kg, respectively).

| Value          | pH  | OM (%) | Silt (%) | Clay (%) | Fe   | Mn   | Zn   | Cu   | Ni   | Pb   |
|----------------|-----|--------|----------|----------|------|------|------|------|------|------|
| Mean (n = 48)  | 7.41| 0.44   | 33.24    | 3.91     | 4.45 | 0.61 | 0.37 | 0.09 | 0.40 | 0.45 |
| Minimum        | 6.74| 0.03   | 14.70    | 3.20     | 2.59 | 0.25 | 0.22 | 0.03 | 0.10 | 0.16 |
| Maximum        | 7.97| 1.14   | 39.00    | 5.00     | 6.84 | 1.04 | 0.66 | 0.20 | 0.97 | 1.02 |
| CV (%)         | 4.0 | 87.7   | 17.2     | 12.3     | 30.6 | 35.8 | 33.7 | 49.5 | 62.8 | 56.1 |

Table 1. Minimum, maximum, mean, and coefficient of variance (CV) of pH, organic matter (OM), silt and clay concentrations (%), and heavy metals in the sediment of the River Nile supporting the growth of *Vossia cuspidata* populations in the mainstream of River Nile (Cairo, Egypt) during one growing season.

| Organ        | Value                  | Fe   | Mn   | Zn   | Cu   | Ni   | Pb   |
|--------------|------------------------|------|------|------|------|------|------|
| Root         | Mean (n = 48)          | 2527.4 | 409.2 | 20.6 | 11.4 | 15.2 | 13.9 |
|              | Minimum                | 130.3 | 121.9 | 6.8  | 1.9  | 2.6  | 0.6  |
|              | Maximum                | 5888.6 | 788.2 | 43.8 | 24.4 | 46.0 | 43.6 |
|              | CV (%)                 | 75.2 | 56.4 | 52.2 | 57.1 | 92.2 | 99.2 |
| Rhizome      | Mean (n = 48)          | 445.3 | 36.4  | 20.9 | 9.5  | 15.6 | 14.4 |
|              | Minimum                | 224.7 | 17.1  | 9.1  | 1.1  | 1.7  | 0.2  |
|              | Maximum                | 975.8 | 57.7  | 35.3 | 28.5 | 53.3 | 52.5 |
|              | CV (%)                 | 50.7 | 36.5 | 39.4 | 77.5 | 104.6 | 116.0 |
| Leaf         | Mean (n = 48)          | 772.3 | 54.9  | 13.5 | 8.1  | 17.6 | 17.6 |
|              | Minimum                | 263.7 | 29.8  | 5.8  | 1.2  | 2.6  | 1.3  |
|              | Maximum                | 5021.9 | 117.1 | 36.7 | 17.5 | 71.1 | 66.7 |
|              | CV (%)                 | 148.1 | 44.5 | 58.3 | 65.9 | 120.6 | 112.6 |
| Stem         | Mean (n = 48)          | 700.1 | 25.0  | 21.5 | 14.0 | 17.6 | 16.3 |
|              | Minimum                | 105.4 | 12.5  | 2.8  | 0.6  | 2.6  | 0.7  |
|              | Maximum                | 2877.6 | 44.4 | 75.6 | 85.4 | 50.9 | 50.1 |
|              | CV (%)                 | 91.3 | 37.9 | 85.0 | 142.4 | 84.9 | 92.3 |

The BCF values were >1 for all the analyzed elements, with the highest value for Mn (766.6), followed by Fe (573.0) (Table 3). BCF values for Fe and Mn were significantly different from other elements (*F*-value = 26.4, at *p* < 0.001). TF was >1 for Zn and Cu in rhizome and stem, Mn in leaf, and Ni and Pb in stem and leaf. There were no significant differences in the TF values of the elements/organ (Table 3).
Table 3. Mean ± standard error of the bioconcentration factors (BCFs) of heavy metals from sediment to *Vossia cuspidata* roots and the translocation factors (TFs) of heavy metals from *Vossia cuspidata* roots to rhizomes (TF<sub>rhizome</sub>), stems (TF<sub>stem</sub>), and leaves (TF<sub>leaf</sub>) in the mainstream of River Nile (Cairo, Egypt) during one growing season.

| Element | BCF     | TF<sub>rhizome</sub> | TF<sub>stem</sub> | TF<sub>leaf</sub> |
|---------|---------|----------------------|------------------|------------------|
| Fe      | $573.0 \pm 115.0$<sup>a</sup> | $0.54 \pm 0.1$<sup>a</sup> | $0.82 \pm 0.2$<sup>a</sup> | $0.73 \pm 0.1$<sup>a</sup> |
| Mn      | $766.6 \pm 111.2$<sup>a</sup> | $0.81 \pm 0.3$<sup>a</sup> | $0.56 \pm 0.2$<sup>a</sup> | $1.11 \pm 0.4$<sup>a</sup> |
| Zn      | $57.6 \pm 8.1$<sup>b</sup>   | $1.14 \pm 0.1$<sup>a</sup> | $1.21 \pm 0.3$<sup>a</sup> | $0.72 \pm 0.1$<sup>a</sup> |
| Cu      | $147.9 \pm 24.6$<sup>b</sup> | $1.02 \pm 0.4$<sup>a</sup> | $1.28 \pm 0.4$<sup>a</sup> | $0.79 \pm 0.2$<sup>a</sup> |
| Ni      | $60.1 \pm 25.3$<sup>b</sup>  | $0.98 \pm 0.5$<sup>a</sup> | $1.12 \pm 0.3$<sup>a</sup> | $1.15 \pm 0.7$<sup>a</sup> |
| Pb      | $41.9 \pm 15.8$<sup>b</sup>  | $0.97 \pm 0.5$<sup>a</sup> | $1.16 \pm 0.3$<sup>a</sup> | $1.21 \pm 0.7$<sup>a</sup> |
| F-value | $26.2$** | $0.3$ns              | $0.9$ns          | $0.2$ns          |

Means followed by different letters are significantly different at *p* < 0.05 according to Tukey’s HSD test. **:** *p* < 0.001, *ns* : not significant (i.e., *p* > 0.05).

The Pearson correlation analysis between HM in plant organs and sediment variables showed non-significant correlations between HM in the root and sediment HM, pH, OM, and silt and clay concentrations at *p* < 0.05 (Figure 2, Table S1). For rhizome, Ni and Pb had a significant positive correlation with Fe (*r* = 0.59 and 0.60, respectively) and Mn (*r* = 0.57 and 0.56, respectively) and Cu with Zn (*r* = 0.54). In leaf, Ni and Pb showed a highly significant positive correlation with sediment HM except for Mn and Cu. In addition, Ni and Pb in stem had a significant positive correlation with sediment Ni and Pb at *p* < 0.05 and between sediment Ni and stem Cu (*r* = 0.49, at *p* < 0.05).

Regression models were developed to help with the prediction of the HM concentrations in the organs of *V. cuspidata* dependent on their concentrations in the sediment, using sediment pH, OM, and sediment silt and clay concentrations as cofactors. The developed model equations and their prediction accuracies are presented in Table 4. The cross-validation of the models was augmented by the associations between the measured and the predicted HM concentrations, concerning the peak of both $R^2$, ME, and low MNAEs. Based on the output of the Student’s *t*-tests, there were no significant differences between the actual and the expected HM concentrations in different organs, i.e., the models perform effectively.

Sixteen models out of 24 established models had high significant $R^2$, with a range from 0.26 for Fe in rhizomes to 0.66 for Ni in leaves (Table 4). The ME values ranged from 0.112 for Pb in rhizomes to 0.873 for Ni in leaves. In addition, the regression models had low MNAE values ranging from 0.058 for Ni in leaves to 0.284 for Cu in roots. In the roots and rhizomes, the equations that generated the largest $R^2$ (0.558 and 0.413, respectively) were for Mn, followed by Zn ($R^2 = 0.36$) in roots and Fe in rhizomes ($R^2 = 0.26$). The values of $R^2$ were linked with high ME and small MNAE values. In leaves, the equations with the highest $R^2$ values (0.663, 0.578, and 0.497) were for Ni, Pb, and Zn, respectively, and were associated with remarkably high ME and low MNAE values. The same was true in stems for Ni and Pb equations ($R^2 = 0.539$ and 0.536, respectively).
Figure 2. Pearson correlation coefficient ($r$-values, $n = 48$) between heavy metals in the different organs of *Vossia cuspidata* (roots, rhizomes, leaves, and stems) and their concentrations in the sediment, pH, organic matter (OM), and silt and clay concentrations (%).

Regression models were developed to help with the prediction of the HM concentrations in the organs of *V. cuspidata* dependent on their concentrations in the sediment, using sediment pH, OM, and sediment silt and clay concentrations as cofactors. The developed model equations and their prediction accuracies are presented in Table 4. The cross-validation of the models was augmented by the associations between the measured and the predicted HM concentrations, concerning the peak of both $R^2$, ME, and low MNAEs.

Based on the output of the Student's $t$-tests, there were no significant differences between...
Table 4. Regression models between heavy metal concentrations in *Vossia cuspidata* organs (mg/kg) and sediment heavy metals (mg/kg), pH, organic matter (OM), and silt and clay concentrations (%).

| Equation | R²   | ME    | MNAE  | MNB  | Student's t-Test |
|----------|------|-------|-------|------|------------------|
| **Roots** |      |       |       |      |                  |
| Cu = −103.826 + (42.112 × Cu_{soil}) + (0.657 × OM) + (11.677 × pH) + (0.178 × Silt) + (4.882 × Clay) | 0.332 ** | 0.604 | 0.284 | 0.072 | 0.771 | 0.457 |
| Fe = −24977.674 + (479.263 × Fe_{soil}) − (953.986 × OM) + (2069.725 × pH) + (156.277 × Silt) + (1349.333 × Clay) | 0.325 ** | 0.581 | 0.302 | 0.079 | 0.798 | 0.442 |
| Mn = 5767.464 + (303.339 × Mn_{soil}) + (148.907 × OM) − (593.591 × pH) − (22.251 × Silt) − (120.727 × Clay) | 0.558 *** | 0.842 | 0.146 | 0.024 | 0.288 | 0.779 |
| Ni = 283.880 − (22.644 × Ni_{soil}) + (21.063 × OM) − (28.277 × pH) + (0.419 × Silt) − (18.777 × Clay) | 0.320 ** | 0.565 | 0.338 | 0.088 | 0.841 | 0.418 |
| Pb = 287.031 − (26.505 × Pb_{soil}) + (24.511 × OM) − (28.310 × pH) + (0.422 × Silt) − (19.545 × Clay) | 0.349 ** | 0.623 | 0.246 | 0.071 | 0.642 | 0.534 |
| Zn = 192.147 + (9.272 × Zn_{soil}) + (5.322 × OM) − (20.812 × pH) + (0.289 × Silt) − (8.481 × Clay) | 0.360 ** | 0.663 | 0.237 | 0.066 | 0.618 | 0.549 |
| **Rhizomes** |      |       |       |      |                  |
| Cu = −35.775 + (26.287 × Cu_{soil}) − (4.728 × OM) + (6.582 × pH) − (0.808 × Silt) − (0.335 × Clay) | 0.196 | 0.355 | 0.468 | 0.159 | 2.611 | 0.024 |
| Fe = −339.958 + (40.843 × Fe_{soil}) + (312.337 × OM) + (50.835 × pH) + (2.193 × Silt) + (4.202 × Clay) | 0.260 * | 0.464 | 0.362 | 0.113 | 1.323 | 0.213 |
| Mn = −56.183 − (6.640 × Mn_{soil}) + (14.654 × OM) + (7.736 × pH) + (1.109 × Silt) + (1.021 × Clay) | 0.413 *** | 0.785 | 0.218 | 0.048 | 0.515 | 0.617 |
| Ni = −144.065 + (15.105 × Ni_{soil}) + (1.163 × OM) + (17.499 × pH) + (0.308 × Silt) + (3.349 × Clay) | 0.098 | 0.228 | 0.530 | 0.226 | 3.080 | 0.010 |
| Pb = −143.622 + (15.696 × Pb_{soil}) + (0.370 × OM) + (17.568 × pH) + (0.282 × Silt) + (2.868 × Clay) | 0.091 | 0.112 | 0.817 | 0.295 | 3.739 | 0.003 |
| Zn = −87.860 + (0.076 × Zn_{soil}) − (2.994 × OM) + (8.907 × pH) + (0.405 × Silt) + (7.822 × Clay) | 0.198 | 0.423 | 0.421 | 0.129 | 2.453 | 0.032 |
| **Leaves** |      |       |       |      |                  |
| Cu = −108.796 + (6.862 × Cu_{soil}) − (3.108 × OM) + (11.248 × pH) + (0.248 × Silt) + (6.660 × Clay) | 0.378 ** | 0.695 | 0.223 | 0.065 | 0.593 | 0.565 |
| Fe = 16156.157 − (323.433 × Fe_{soil}) + (1374.681 × OM) − (1217.499 × pH) − (18.437 × Silt) − (1256.146 × Clay) | 0.396 ** | 0.785 | 0.219 | 0.051 | 0.520 | 0.613 |
| Mn = −16.556 + (7.443 × Mn_{soil}) − (21.895 × OM) + (5.149 × pH) + (1.009 × Silt) + (1.241 × Clay) | 0.213 | 0.442 | 0.368 | 0.118 | 1.457 | 0.173 |
| Ni = 281.695 + (51.451 × Ni_{soil}) − (0.513 × OM) − (30.920 × pH) + (0.264 × Silt) − (16.449 × Clay) | 0.663 *** | 0.873 | 0.058 | 0.003 | 0.041 | 0.968 |
| Pb = 233.578 + (42.096 × Pb_{soil}) + (2.603 × OM) − (27.027 × pH) + (0.250 × Silt) − (11.302 × Clay) | 0.578 ** | 0.849 | 0.106 | 0.022 | 0.147 | 0.886 |
| Zn = 151.569 − (4.207 × Zn_{soil}) + (11.465 × OM) − (15.815 × pH) − (0.056 × Silt) − (5.768 × Clay) | 0.497 *** | 0.812 | 0.187 | 0.039 | 0.423 | 0.681 |
### 4. Discussion

Low OM and clay concentrations were obtained in the sediment samples that can help in the accumulation of HMs in the organs of *V. cuspidata*. It was reported that low percentages of OM and clay are associated with an increase in the bioavailability of HMs in the sediment of watercourses [8,30]. The pH of the sediment was slightly alkaline but not high, i.e., it probably had no significant influence on the bioavailability mobility of HMs in the sediment. Chaudri et al. [31] in 2007 and Eid et al. [7,8] in 2018 and 2020a reported that high sediment pH is known to enhance the adsorption of many HMs and decrease its solubility in the sediment solutions. Low concentrations of most of the HMs were detected in the sediment. These low HM concentrations in the river’s sediment and water may be ascribed to the frequent discharging of water into the river leading to some improvement in its quality [23,32] (Abdel-Shafy and Aly, 2002; Farahat et al., 2021). The presence of excessive Fe and Mn concentrations in the sediment samples as compared to other HMs was reported for the River Nile in other studies [11,23].

This study indicated that the root of *V. cuspidata* was the main organ for the accumulation of the estimated HMs. Similar findings were reported for the accumulation ability of roots to HMs in *V. cuspidata* and other aquatic macrophytes such as *T. domingensis* and *Phragmites australis* [8,18,23]. The presence of phytotoxic concentrations of Fe in the roots may be due to the presence of iron plaque on the root surfaces that plays an important role in the stabilization of HMs [33]. In addition, the existence of phytotoxic Mn concentrations in the root could be due to its aggregation in the root plaque with Fe and other HMs, as several aquatic macrophytes have been known to do [34]. The presence of low mean or phytotoxic maximum concentrations of Cu, Ni, and Pb in the organs of *V. cuspidata* depends on the monthly availability of these metals in sediments and on the accumulation ability of the plant to these metals [18,23].

The existence of BCF values >1 for all metals reflects the high bioaccumulation potential of *V. cuspidata* when compared to other Egyptian aquatic plants. For example, the BCF value for Fe in this study was nearly two times and 7.4 times that reported for *T. domingensis* and *P. australis*, respectively [8,28]. The same was true in the case of Zn and Cu. On the contrary, the BCF values for Ni and Pb in the roots of *V. cuspidata* represented about 2.4 and 4.1 folds, respectively, of that reported for *P. australis* [27]. *T. domingensis* had BCF values 0.55 for Ni and 6.74 for Pb [8] compared to 60.1 and 41.9, respectively, in the present study. This study indicated that *V. cuspidata* is a good candidate for phytoremediation use.

### Table 4. Cont.

| Equation | $R^2$ | ME | MNAE | MNB | Student’s $t$-Test |
|----------|------|----|------|-----|-------------------|
| **Stems** |      |    |      |     |                   |
| Cu = 60.375 – (83.621 × Cu$_{soil}$) + (25.657 × OM) – (0.459 × pH) – (0.432 × Silt) – (8.238 × Clay) | 0.279 * | 0.483 | 0.341 | 0.103 | 0.860 | 0.408 |
| Fe = 1048.276 + (214.361 × Fe$_{soil}$) + (491.823 × OM) – (224.471 × pH) + (14.344 × Silt) – (85.061 × Clay) | 0.208 | 0.425 | 0.401 | 0.124 | 1.878 | 0.087 |
| Mn = −80.752 – (1.617 × Mn$_{soil}$) – (4.740 × OM) + (11.067 × pH) + (0.344 × Silt) + (3.921 × Clay) | 0.113 | 0.293 | 0.527 | 0.225 | 3.022 | 0.012 |
| Ni = −227.257 + (67.746 × Ni$_{soil}$) – (24.093 × OM) + (27.663 × pH) + (0.497 × Silt) + (1.298 × Clay) | 0.539 *** | 0.840 | 0.163 | 0.036 | 0.349 | 0.734 |
| Pb = −238.447 + (68.242 × Pb$_{soil}$) – (25.226 × OM) + (27.505 × pH) + (0.589 × Silt) + (2.955 × Clay) | 0.536 *** | 0.817 | 0.171 | 0.037 | 0.368 | 0.720 |
| Zn = −71.786 + (33.922 × Zn$_{soil}$) + (7.227 × OM) + (10.965 × pH) + (0.903 × Silt) – (8.627 × Clay) | 0.162 | 0.297 | 0.508 | 0.212 | 2.801 | 0.017 |

$R^2$: coefficient of determination, ME: model efficiency, MNAE: mean normalized average error, MNB: mean normalized bias. *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$. 
The translocations and accumulation of HMs from roots to other plant organs depend on many factors such as water transport, chemical speciation, climate factors, physiology and phenology of plants, solubility, and availability of each metal ion [10,35,36]. Accordingly, the translocation of HMs from roots to different organs of the *V. cuspidata* was not even. Although the emergent macrophytes are characterized by low mobility and translocation of HMs from roots to above-ground organs, according to Rezania et al. [37], the TF for Zn, Cu (in stems), Ni and Pb (in leaf and stem), and Mn in leaf was >1. This means that *V. cuspidata* can either exclude the HMs through its accumulation in the roots or by transferring to physiologically active above-ground organs but in low amounts. According to Lago-Vila et al. [38], one of the main requirements for a candidate species for phytoremediation is its ability to translocate HMs to the shoot system.

There were some significantly positive correlations between the HMs in the plant and other metals in the sediment. These correlations reflect the direct association between HMs in the sediment and the organs of *V. cuspidata* and the cumulative effect of the pollution of the river Nile. These findings are in line with those reported for other aquatic macrophytes in Egypt [18,39].

Sediment pH, OM, and silt and clay concentrations play an important role in the availability and solubility of HMs in the sediment. As previously mentioned, the existence of low values of pH, OM, and clay concentrations in the sediment increased the bioavailability of HMs and consequently their absorption by plants. These sediment variables are usually included in the prediction models of HMs due to their direct effect on their bioavailability [8,40]. The inclusion of sediment pH, OM, and silt and clay concentrations led to the development of 16 equations with statistically significant $R^2$ values with high ME and low MNAE values. The application of these regression models will help in the prediction of HMs in plant organs. The maximum $R^2$ values were 0.539 and 0.663 for leaf and stem Ni, respectively, and 0.588 and 0.413 for rhizome Mn, respectively. Eid et al. [8,28] found that the maximum $R^2$ values for *T. domingensis* were 0.558 for shoot Pb, 0.751 for rhizome Cd, and 0.763 for root Zn, while they were 0.698 for leaf Cu, 0.629 for stem Fe, and 0.434 for below-ground organs in young populations of *P. australis* in Lake Burullus, Egypt. The existence of low MNAE values with the developed models refers to non-significant differences between the predicted and measured data, i.e., high performance of the models. Since these are the first developed prediction models for the uptake of HMs by *V. cuspidata*, we could not compare our results with previous related studies on the same species. The existence of some low values for $R^2$ may be attributed to the pooling of variable sediment and HM data collected throughout the year. This could be improved in the future by increasing the number of replicas for some metals in the sediment and plant organs.

5. Conclusions

This was the first attempt, to our knowledge, to create regression models that predict HM uptake in different organs of the invasive grass *V. cuspidata* in the Nile River ecosystem. This study identified a significant correlation between the concentrations of some HMs in organs of *V. cuspidata* and the supporting sediment. *V. cuspidata* is a good candidate for biomonitoring the contamination of watercourses by HMs and helps in its phytoremediation. In this study, the BCF values of all HMs were more than the unity indicating that this species is a good phytoextractor for HMs. The presence of TF >1 for some HMs in a certain organ means that this plant has also the ability to phytoextract these elements. Most of the developed regression models for HM concentrations in the organs of *V. cuspidata* performed well, with high $R^2$ and Me and low MNAEs. These models will represent a good tool for environment-related studies on this species, particularly ecological risk assessment. In addition, they overcome the restriction of using bioaccumulation factors since these models are using the most influential sediment variables that affect the uptake of HMs by *V. cuspidata*. 
Supplementary Materials: The following are available online at http://www.mdpi.com/1099-4300/13/19/10558/s1, Table S1: Pearson correlation coefficient (r-values, n = 48) between heavy metals in Vossia cuspidate organs and some sediment variables.

Author Contributions: E.A.F.: conceptualization, methodology, formal analysis, investigation, drafting the original manuscript, writing—review and editing, and visualization; W.F.M.: field work, laboratory chemical analysis; H.E.A.A.: writing—review and editing; H.F.F.: writing—review and editing; M.A.: writing—review and editing; E.M.E.: conducting the modeling analysis, writing—review and editing; G.M.F.: supervision, writing—review and editing. All authors have read and agreed to the published version of the manuscript.

Funding: Arshad M. extends his appreciation to the Deanship of Scientific Research at King Khalid University for funding this work through the Research Group Project under grant number RGP1/301/42.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data are contained within the article and Supplementary Materials File.

Conflicts of Interest: The authors declare no conflict of interest.

References
1. Chakravarty, P.; SenSarma, N.; Sarma, H.P. Biosorption of cadmium (II) from aqueous solution using heartwood powder of Areca catechu. Chem. Eng. J. 2010, 162, 949–955. [CrossRef] [PubMed]
2. Oyuela Leguizamo, M.A.; Gómez, W.D.F.; Sarmiento, M.C.G. Native herbaceous plant species with potential use in phytoremediation of heavy metals, spotlight on wetlands: A review. Chemosphere 2017, 168, 1230–1247. [CrossRef] [PubMed]
3. Chandra, R.; Yadav, S.; Yadav, S. Phytoextraction potential of heavy metals by native wetland plants growing on chlorolignin containing sludge of pulp and paper industry. Ecol. Eng. 2017, 98, 134–145. [CrossRef]
4. Krishnamurti, G.S.; Huang, P.M.; Kozak, L.M. Sorption and desorption kinetics of cadmium from soils: Influence of phosphate. Soil Sci. 1999, 164, 888–898. [CrossRef]
5. Usman AR, A.; Kuzyakov, Y.; Stahr, K. Sorption, Desorption, and Immobilization of Heavy Metals by Artificial Soil; University of Hohenheim: Stuttgart, Germany, 2008.
6. Novotná, M.; Mikeš, O.; Komprdová, K. Development and comparison of regression models for the uptake of metals into various field crops. Environ. Pollut. 2015, 207, 357–364. [CrossRef]
7. Eid, E.M.; Alrumman, S.A.; Farahat, E.A.; El-Bebany, A.F. Prediction models for evaluating the uptake of heavy metals by cucumbers (Cucumis sativus L.) grown in agricultural soils amended with sewage sludge. Environ. Monit. Assess. 2018, 190, 501. [CrossRef] [PubMed]
8. Eid, E.M.; Galal, T.M.; Shaltout, K.H.; El-Sheikh, M.A.; Asaeda, T.; Alfarhan, A.H.; Alharthi, A.; Alshehri, A.M.A.; Picó, Y.; et al. Biomonitoring potential of the native aquatic plant Typha domingensis by predicting trace metals accumulation in the Egyptian Lake Burullus. Sci. Total Environ. 2020, 714, 136603. [CrossRef] [PubMed]
9. Eid, E.M.; Shaltout, K.H. Monthly variations of trace elements accumulation and distribution in above- and below-ground biomass of Phragmites australis (Cav.) Trin. Ex Steudel in Lake Burullus (Egypt): A biomonitoring application. Ecol. Eng. 2014, 73, 17–25. [CrossRef]
10. Galal, T.M.; Farahat, E.A. The invasive macrophyte Pistia stratiotes L. as a bioindicator for water pollution in Lake Mariut, Egypt. Environ. Monit. Assess. 2015, 178, 701. [CrossRef] [PubMed]
11. Farahat, E.A.; Galal, T.M. Trace metal accumulation by Ranunculus sceleratus: Implications for phytostabilization. Environ. Sci. Pollut. Res. 2018, 25, 4214–4222. [CrossRef] [PubMed]
12. Eid, E.M.; Shaltout, K.H.; Moghamm, F.S.; Youssef, M.S.; El-Mohsnawy, E.; Haroun, S.A. Bioaccumulation and translocation of nine heavy metals by Eichhornia crassipes in Nile Delta, Egypt: Perspectives for phytoremediation. Int. J. Phytoremediat. 2019, 21, 821–830. [CrossRef] [PubMed]
13. Odoh, C.K.; Zabney, N.; Sam, K.; Eze, C.N. Status, progress and challenges of phytoremediation—An African scenario. J. Environ. Manag. 2019, 237, 365–378. [CrossRef] [PubMed]
14. Augustsson, A.L.; Uddh-Soderberg, T.E.; Hogalm, K.J.; Filipsson, M.E. Metal uptake by homegrown vegetables—The relative importance in human health risk assessments at contaminated sites. Environ. Res. 2015, 138, 181–190. [CrossRef] [PubMed]
15. Zhang, S.; Song, J.; Gao, H.; Zhang, Q.; Lv, M.; Wang, S.; Liu, G. Improving prediction of metal uptake by Chinese cabbage (Brassica pekinensis L.) based on a soil-plant stepwise analysis. Sci. Total Environ. 2016, 569, 1595–1605. [CrossRef] [PubMed]
16. Boulos, L. Flora of Egypt: Vol. 4 (Alismataceae–Orchidaceae); Al Hadara Publishing: Cairo, Egypt, 2005; p. 617.
17. Shehata, M.N. Ecological Studies on Vossia cuspidate, Roxb Griff in the Nile Delta of Egypt. Egypt. J. Bot. 1996, 36, 37–51.
18. Galal, T.M.; Gharib, F.A.; Ghazi, S.M.; Mansour, K.H. Phytostabilization of heavy metals by the emergent macrophyte Vossia cuspidata (Roxb.) Griff.: A phytoremediation approach. Int. J. Phytoremediat. 2017, 19, 992–999. [CrossRef]
19. Boulos, L. Flora of Egypt. Cairo, Egypt: Al Hadara Publishing; Cairo, Egypt, 1995.
20. Raphael, E.; Momoh, S.; Kayode, D.; Gideon, A.; Friday, E.T. The phytochemical constituents of Vossia cuspidata and Synedrella nodiflora. Int. J. Curr. Res. Biosci. Plant Biol. 2016, 3, 53–57. [CrossRef]
21. Mahmoud, W.F.; Farahat, E.A.; Fahmy, G.M.; Farghaly, H.F.; Awad, H.E. Monthly and seasonal variations of biomass partitioning and macronutrients in the invasive grass Vossia cuspidata (Roxb.) Griff. Aquat. Bot. 2021, 172, 103999. [CrossRef]
22. Mahmoud, W.F.; Farahat, E.A.; Fahmy, G.M.; Farghaly, H.F.; Awad, H.E. Impact of the invasive species Vossia cuspidata (Roxb.) Griff. on the diversity and temporal changes of the native flora of the River Nile in Egypt. Taenckholmia 2021, 41, 1–17.
23. Farahat, E.A.; Mahmoud, W.F.; Fahmy, G.M. Seasonal variations of heavy metals in water, sediment, and organs of Vossia cuspidata (Roxb.) Griff. in River Nile ecosystem: Implication for phytoremediation. Environ. Sci. Pollut. Res. 2021, 28, 32626–32633. [CrossRef]
24. Chapman, H.D.; Pratt, R.E. Methods of Analysis for Soil, Plants and Water; Department of Soil, Plant, and Nutritional Sciences, University of California: Los Angeles, CA, USA, 1961; p. 309.
25. Allen, S.E. Chemical Analysis of Ecological Materials; Blackwell Science; Oxford, UK, 1989.
26. Xiao, R.; Bai, J.; Zhang, H.; Gao, H.; Liu, X.; Wilkes, A. Changes of Fe, Al and Fe contents in fringe marshes along a pedogenic chronosequence in the Pearl River estuary, South China. Cont. Shelf Res. 2011, 31, 739–747. [CrossRef]
27. Gupta, S.; Nayek, S.; Saha, R.N.; Satpati, S. Assessment of heavy metal accumulation in macrophyte, agricultural soil, and crop plants adjacent to discharge zone of sponge iron factory. Environ. Geol. 2008, 55, 731–739. [CrossRef]
28. Eid, E.M.; Shaltout, K.H.; Al-Sodany, Y.M.; Haroun, S.A.; Galal, T.M.; Ayed, H.; Khedher, K.M.; Jensen, K. Common reed (Phragmites australis (Cav.) Trin. ex Steudel) as a candidate for predicting heavy metal contamination in Lake Burullus, Egypt: A biomonitoring approach. Ecol. Eng. 2020, 148, 105787. [CrossRef]
29. SPSS IBM. IBM SPSS Statistics software for Windows; 2013.
30. Liu, Z.L.; He, X.Y.; Chen, W.; Yuan, F.H.; Yan, K.; Tao, D.L. Accumulation and tolerance characteristics of cadmium in a potential hyperaccumulator-Lonicera japonica Thunb. J. Hazard. Mater. 2009, 169, 170–175. [CrossRef] [PubMed]
31. Chaudri, A.; McGrath, S.; Gibbs, P.; Chambers, B.; Carlton-Smith, C.; Godley, A.; Bacon, J.; Campbell, C.; Aitken, K. Cadmium availability to wheat grain in soils treated with sewage sludge or metal salts. Chemosphere 2007, 66, 1415–1423. [CrossRef] [PubMed]
32. Abdel-Shafy, H.I.; Aly, R.O. Water issue in Egypt: Resources, pollution and protection endeavors. Cent. Eur. J. Occup. Environ. Med. 2002, 8, 3–21.
33. Ma, H.; Gao, F.; Fan, X.; Cui, B.; Liu, C.; Cui, E.; Zhang, Z.; Hu, C.; Mo, Y. Vinasxe affects the formation of iron plaque on roots of Acorus calamus and immobilization of lead, cadmium, copper, zinc by this plant. J. Water Process. Eng. 2020, 38, 101587. [CrossRef] [PubMed]
34. St-Cyr, L.; Campbell, P.G.C. Metals (Fe, Mn, Zn) in the root plaque of submerged aquatic plants collected in situ: Relations with metal concentrations in the adjacent sediments and in the root tissue. Biogeochemistry 1996, 33, 45–76. [CrossRef]
35. Bonanno, G.; Borg, J.A.; DiMartino, V. Levels of heavy metals in wetland and marine vascular plants and their biomonitoring potential: A comparative assessment. Sci. Total Environ. 2017, 576, 796–806. [CrossRef]
36. Christou, A.; Theologides, C.P.; Costa, C.; Kalavrouziotis, I.K.; Varnavas, S.P. Assessment of toxic heavy metal concentrations in soils and wild and cultivated plant species in Limni abandoned copper mining site, Cyprus. J. Geochem. Explor. 2017, 178, 16–22. [CrossRef]
37. Rezania, S.; Park, J.; Rupani, P.F.; Darajeh, N.; Xu, X. Phytoremediation potential and control of Phragmites australis as a green phytomass: An overview. Environ. Sci. Pollut. Res. 2019, 26, 7428–7441. [CrossRef]
38. Lago-Vila, M.; Arenas-Lago, D.; Rodriguez-Seijo, A.; Andrade, M.L.; Vega, F.A. Ability of Cytisus scoparius for phytoremediation of soils from a Pb/Zn mine: Assessment of metal bioavailability and bioaccumulation. J. Environ. Manag. 2019, 235, 152–160. [CrossRef] [PubMed]
39. Eid, E.M.; Shaltout, K.H.; El-Sheikh, M.A.; Aseada, T. Seasonal courses of nutrients and heavy metals in water, sediment and above- and below-ground Typha domingensis biomass in Lake Burullus (Egypt): Perspective for phytoremediation. Flora 2012, 207, 783–794. [CrossRef]
40. Lopes, C.; Herva, M.; Franco-Uria, A.; Roca, E. Multicorrelation models and uptake factors to estimate extractable metal concentrations from soil and metal in plants in pasturelands fertilized with manure. Environ. Pollut. 2012, 166, 17–22. [CrossRef] [PubMed]