



## EFFECTS OF LEAD AND CHROMIUM ON GERMINATION AND ROOT ARCHITECTURE OF *TYPHA LATIFOLIA* (TYPHACEAE) SEEDLINGS

### EFFECTOS DEL PLOMO Y CROMO EN LA GERMINACIÓN Y ARQUITECTURA DE RAÍZ DE PLÁNTULAS DE *TYPHA LATIFOLIA* (TYPHACEAE)

Renato Oquendo\*<sup>1</sup>, Loiret Fernández\*<sup>2</sup>, Galo Pabón-Garcés\*<sup>1</sup>  
and Lucía Vásquez-Hernandez<sup>1</sup>

<sup>1</sup>Universidad Técnica del Norte, 100104, Ibarra, Ecuador. [<https://ror.org/049784n50>]

<sup>2</sup>Universidad de La Habana, 10400, La Habana, Cuba. [<https://ror.org/04204gr61>]

\*Corresponding author: [ldvasquez@utn.edu.ec](mailto:ldvasquez@utn.edu.ec)

Article received on July 21st, 2021. Accepted, after review, on July 11th, 2023. Published on March 1st, 2026.

#### Abstract

This research evaluates the effect of different concentrations of lead and chromium on root architecture and germination percentage of *Typha latifolia*. Seeds of *Typha latifolia* were collected from Yahuarcocha Lagoon (Ibarra-Ecuador). Pre-germinative treatments and exposure to lead concentrations of 50, 100, and 200 ppm, as well as 10, 20, and 40 ppm of chromium concentrations were applied to the seeds to measure the germination percentage. Regarding root architecture analysis, *T. latifolia* seedlings were grown for four weeks in 50% Hoagland solution and then transferred to rhizotrons, where the heavy metals were added to concentrations like the germination test. The seedlings were kept in these conditions for 15 days and variables such as number of roots, total area, and total length were recorded. Photographic records of the parameters under study were taken and analyzed in Image J and its SmartRoot extension. It was found that both heavy metals affected (significantly) the germination process, especially chromium. Germination percentages lower than 15% and 25% for lead were also obtained. Concerning root architecture, the number of roots and total length from heavy metal treatments were not significantly different from control. However, the total area treatment with chromium showed significant differences with respect to the control.

**Keywords:** Plant physiology, heavy metals, aquatic plants, rhizotron, Yahuarcocha, *Typha latifolia*, Ecuador.

### Resumen

El presente trabajo evalúa el efecto de diferentes concentraciones de plomo y cromo sobre la arquitectura de raíz y porcentaje de germinación de *Typha latifolia*. Para ello, se recolectaron semillas viables de dos sitios en la Laguna de Yahuarcocha (Ibarra-Ecuador). Con relación al porcentaje de germinación se trataron las semillas con procesos pre-germinativos; posteriormente fueron expuestas a concentraciones de 50, 100 y 200 ppm de plomo y 10, 20, 40 ppm de cromo. Para el análisis de la arquitectura de la raíz se cultivaron plántulas de *T. latifolia* por cuatro semanas en solución Hoagland al 50 %, luego fueron trasladadas a rizotrones en los que se añadieron los metales pesados en concentraciones similares al ensayo de germinación. Las plántulas se mantuvieron en dichas condiciones por 15 días en los que se registraron variables como el número de raíces, área total, y longitud total. Se realizaron registros fotográficos de los parámetros en estudio y fueron analizados en el programa ImageJ y su extensión SmartRoot. En el porcentaje de germinación se encontró que los dos metales afectan al proceso de germinación, especialmente el cromo, con el que se obtuvo porcentajes de germinación menores al 15 % y con plomo con un 25 %. Con respecto a la arquitectura de raíz el número de raíces y la longitud total no varió entre el control y los tratamientos con metales. Sin embargo, el tratamiento referente al área total de la raíz tratada con cromo presentó diferencias significativas con respecto al control.

**Palabras clave:** Fisiología vegetal, metales pesados, plantas acuáticas, rizotrón, Yahuarcocha, *Typha latifolia*, Ecuador.

---

Suggested citation: Oquendo, R., Fernández, L., Pabón-Garcés, G. and Vásquez-Hernandez, L. (2026). Effects of lead and chromium on germination and root architecture of *Typha latifolia* (Typhaceae) seedlings . La Granja: Revista de Ciencias de la Vida. Vol. 43(1):145-159. <https://doi.org/10.17163/lgr.n43.2026.08>.

---

#### Orcid IDs:

Renato Oquendo: <https://orcid.org/0000-0001-6916-3834>

Loiret Fernández: <https://orcid.org/0000-0002-2058-781X>

Galo Pabón-Garcés: <https://orcid.org/0000-0001-6044-4429>

Lucía Vásquez-Hernandez: <https://orcid.org/0000-0002-1876-0759>

## 1 Introduction

Environmental pollution by heavy metals has increased significantly due to anthropogenic activities, particularly those derived from industrial practices. Since these contaminants are non-biodegradable substances, they exert lethal effects on plants, such as the reduction of transpiration rate and cell division (Sharma et al., 2022). In response to their exposure, plants exhibit decreased seed germination rates, inactivation of essential enzymes, and impairment of photosynthesis and water regulation (Zaidi et al., 2012). Moreover, heavy metals can alter plant morphology by reducing biomass, stem diameter, length, surface area, and soil distribution, as well as by decreasing the formation of lateral and adventitious roots (Baligar et al., 1998).

The root system of plant species plays a vital role in their growth and development, as it enables the absorption of water, nutrients, and minerals from the soil. For this reason, root architecture exhibits plasticity that depends on soil type and environmental conditions (Armengaud et al., 2009; Franco et al., 2011). The roots of plants tolerant to heavy metals and abiotic stress can respond not only to nutrient deficiencies but also to metal excess in the soil. This condition often leads to reduced growth parameters such as root elongation, biomass production, and root hair development, due to toxicity and the disruption of physiological processes (Barceló and Poschenrieder, 1992; Kahle, 1993).

Despite the detrimental effects of heavy metals, certain plants are known for their tolerance to these contaminants. Some are even capable of incorporating metals through absorption processes and storing them in the vacuole, thus avoiding the aforementioned physiological damages (Pal Singh et al., 2013). One such example is *Typha latifolia*, an aquatic species distributed throughout the Northern Hemisphere and introduced and naturalized in South America, East Africa, Australia, and Hawaii (Smith, 2018). Due to its high tolerance threshold, this species is considered a hyperaccumulator of heavy metals in its roots, with concentrations that may exceed those of the surrounding substrate (Branković et al., 2011). However, despite this tolerance, *T. latifolia* may still exhibit changes in root architecture and a gradual decrease in root growth depending on the concentration of metals to which it is exposed.

Reported growth inhibition thresholds are in the range of 200–500 ppm for lead and 10–60 ppm for chromium (Sasmaz et al., 2008; Chen et al., 2014; Han et al., 2015).

Several techniques have been used to quantify these changes, among which root architecture analysis stands out. This method involves the systematic observation of measurable variables using instruments known as rhizotrons, which allow the manipulation of nutrient concentrations and other external factors (Busch et al., 2006). The description of the root system in plants grown in rhizotrons enables documentation of root growth and form, providing insight into their morphology, topology, and root–root interactions (Lobet et al., 2011). Furthermore, studying the seed germination of wetland plants tolerant to heavy metals, such as *Typha latifolia*, allows for determining the degree of seed resistance to contaminant exposure (Zhang, 2015), which is important in ecological restoration and environmental remediation projects.

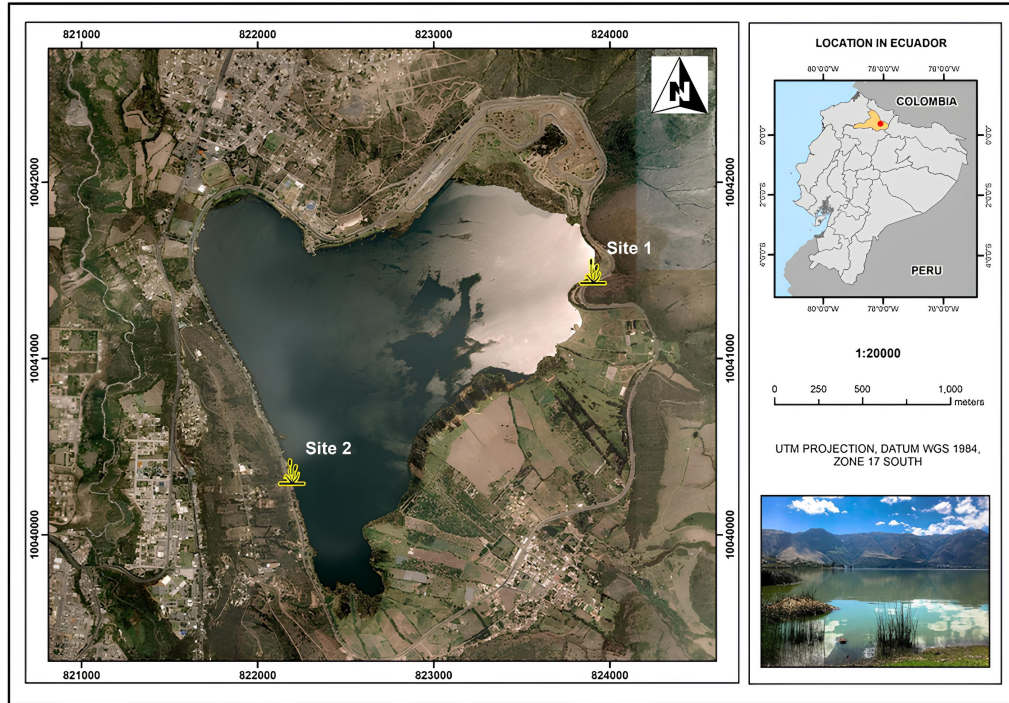
Because of the latter, the objective of this study is to quantify the effect of different concentrations of lead and chromium on the seed germination and root architecture of *Typha latifolia* seedlings under laboratory conditions.

## 2 Materials and Methods

### 2.1 Seed Collection

Seeds of *Typha latifolia* were collected from spontaneously growing populations in Yahuarcocha Lagoon (2,200 m.a.s.l.), located in the province of Imbabura, Ecuador (Figure 1). Two collection sites were selected: the first, located on the northeastern shore of the lagoon (Site 1), locally known as “La vuelta de la paloma,” and the second on the southwestern shore (Site 2), in the area referred to as “La recta de ingreso al poblado de San Miguel de Yahuarcocha.”

From each plant, one mature inflorescence (dark brown in color, according to Smith (2018)) was collected, for a total of 20 inflorescences (ten per sampling site). The samples were stored under refrigeration at 3 °C for seven days. Seed collection was conducted during the rainy season between April and May 2021.



**Figure 1.** Seed collection sites of *T. latifolia* in Yahuarcocha Lagoon. Site 1: northeastern shore of the lagoon; Site 2: southwestern shore. Collection period: April–May 2021 (rainy season).

**Table 1.** Experimental conditions of the treatments in the germination test of *Typha latifolia* seeds collected from two sampling sites in Yahuarcocha Lagoon.

Treatment	Replicates	Description*
Site 1	3	100 seeds from Site 1, hydrated in saline water (1 % NaCl) for 24 h, then hydrated in distilled water for 20 days.
Site 2	3	100 seeds from Site 2, hydrated in saline water (1 % NaCl) for 24 h, then hydrated in distilled water for 20 days.
Control 1	3	100 seeds from Site 1, hydrated in distilled water for 20 days.
Control 2	3	100 seeds from Site 2, hydrated in distilled water for 20 days.

\*General experimental conditions: pH between 6.5 and 7.5; photoperiod of 12 h light / 12 h dark; average temperature of 21 °C; hydration depth of 0.5 cm.

## 2.2 Seed Germination

The seeds, which range from 20,000 to 700,000 per spike (Smith, 2018), were disinfected using 2.5 % sodium hypochlorite (USDA, 2016). They were then placed in Petri dishes containing deionized water to separate viable seeds (those that sink) from non-viable ones (those that float) (Bourgeois et al., 2012).

A total of 600 viable seeds (300 from each site) were selected and placed in three Petri dishes (100 per dish). Each group of seeds was immersed in slightly saline water (1 % NaCl) with a pH between 6.5 and 7.5 for 24 h, following the recommendations of Semenova (2014) for this species.

Subsequently, the seeds were submerged to a depth of 0.5 cm in distilled water until germination,

under a photoperiod of 12 h light / 12 h dark and an average temperature of 21 °C. No scarification treatment was applied, as reported by the Royal Botanical Gardens, Kew (2016), since this procedure reduces germination rates to below 58 % in *Typha latifolia*.

The control treatments consisted of 100 seeds from Site 1 (Control 1) and Site 2 (Control 2), hydrated in distilled water for 20 days, with three replicates each (Table 1).

The effect of lead on seed germination was evaluated by adding the following concentrations to each Petri dish: 50 ppm, 100 ppm, and 200 ppm, in the form of  $Pb(NO_3)_2$ , with three replicates (Table 2).

**Table 2.** Experimental conditions of treatments in the test assessing the effects of different lead (Pb) concentrations on the germination of *Typha latifolia* seeds collected from Yahuarcocha Lagoon.

Treatment	Replicates	Description*
0 ppm (control)	3	100 seeds hydrated in distilled water without lead for 20 days.
50 ppm	3	100 seeds hydrated in distilled water with 50 ppm of lead for 20 days.
100 ppm	3	100 seeds hydrated in distilled water with 100 ppm of lead for 20 days.
200 ppm	3	100 seeds hydrated in distilled water with 200 ppm of lead for 20 days.

\*General experimental conditions: seeds hydrated in saline water (1 % NaCl) for 24 h prior to the addition of different lead concentrations; pH between 6.5 and 7.5; photoperiod of 12 h light / 12 h dark; average temperature of 21 °C; hydration depth of 0.5 cm.

To evaluate the effect of chromium on seed germination, concentrations of 10 ppm, 20 ppm, and 40 ppm were used in the form of  $K_2Cr_2O_7$ , with three replicates for each concentration (Table 3). In both

experimental sets, the control treatments consisted of seeds hydrated in saline water (1 % NaCl) for 24 h as a pregerminative treatment, followed by hydration with distilled water.

**Table 3.** Experimental conditions of treatments in the test assessing the effects of different chromium (Cr) concentrations on the germination of *Typha latifolia* seeds collected from Yahuarcocha Lagoon.

Treatment	Replicates	Description*
0 ppm (control)	3	100 seeds hydrated in distilled water without chromium for 20 days
10 ppm	3	100 seeds hydrated in distilled water with 10 ppm of chromium for 20 days
20 ppm	3	100 seeds hydrated in distilled water with 20 ppm of chromium for 20 days
40 ppm	3	100 seeds hydrated in distilled water with 40 ppm of chromium for 20 days

\* General experimental conditions: seeds hydrated in saline water (1 % NaCl) for 24 h prior to the addition of different chromium concentrations; pH between 6.5 and 7.5; photoperiod of 12 h light / 12 h dark; average temperature of 21 °C; hydration depth of 0.5 cm.

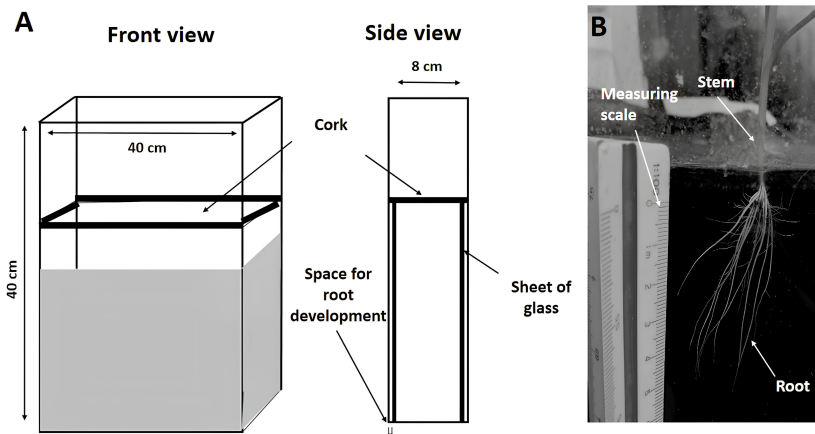
In the first experiment, the variable evaluated was the germination percentage of seeds from Sites 1 and 2 compared to the germination percentages of their respective control treatments (Control 1 and Control 2). In the second and third experiments, the germination percentage was assessed in treatments with seeds exposed to different concentrations of lead and chromium, respectively, as described above.

### 2.3 Root Architecture

To study root growth, a rhizotron was constructed with the following dimensions: 40 cm in front, 8 cm in width (lateral), and 40 cm in depth (in the direction of gravity) (Figure 2). The rhizotron was

equipped with a double 5 mm thick glass front, allowing for temporal monitoring of root growth through photographic recording. On the surface of the growth medium (50% standard Hoagland solution), a 4 mm cork sheet was placed, on which seedlings obtained from the control treatment of the germination experiment were positioned.

A total of three seedlings were transplanted into each rhizotron (45 days after germination), resulting in a total of 21 seedlings. Photographic records were taken at 0 (zero), 5, 10, and 15 days after transplantation (DAT). The rhizotrons were placed outdoors under sunlight at an approximate altitude of 2,220 m.a.s.l. and an average temperature of 20 °C.



**Figure 2.** Characteristics of the constructed rhizotron. A. Dimensions of the container from the front and side views. B. Front view of a *Typha* seedling growing in the rhizotron.

### 2.4 Growth Conditions

Each rhizotron was filled with 8 L of half-strength Hoagland solution to simulate the natural conditions of the collection site (Hoagland and Arnon, 1938). Solutions were then added to achieve concentrations of 50, 100, and 200 ppm of lead and 10, 20, and 40 ppm of chromium. The concentrations of metal solutions were selected based on the species' tolerance reported in the literature and on the ranges of these metals found in previous studies characterizing the water, soil, sediment, and roots of *Typha latifolia* in Yahuarcocha Lagoon (Oquendo et al., 2020).

Control treatments consisted of seedlings grown exclusively in 50% standard Hoagland solution. The germination and root architecture experiments of *Typha latifolia* were conducted at the Environmental Research Laboratory (LABINAM) of Universidad Técnica del Norte.

### 2.5 Image Analysis

Sequential photographs of all roots taken at the specified recording times (0, 5, 10, and 15 days after transplanting) were processed using the software ImageJ and its SmartRoot extension. The variables evaluated included: number of roots, total root area (cm<sup>2</sup>), and total root length (cm) of the seedlings.

## 2.6 Statistical Analysis

Based on the data obtained for *Typha latifolia* seed germination percentage, the effects of lead and chromium on germination, and the effects of these heavy metals on root growth (in the rhizotron), three one-way analyses of variance (ANOVA) were performed. The first ANOVA compared the germination of seeds hydrated in distilled water (Controls 1 and 2) with that of seeds subjected to a pregerminative treatment (hydrated in 1% NaCl for 24 h), for both Site 1 and Site 2. The second and third ANOVAs determined the effect of different heavy metal concentrations (lead and chromium) on the variables: germination percentage (in the second ANOVA) and number of roots, total area, and total root length (in the third ANOVA).

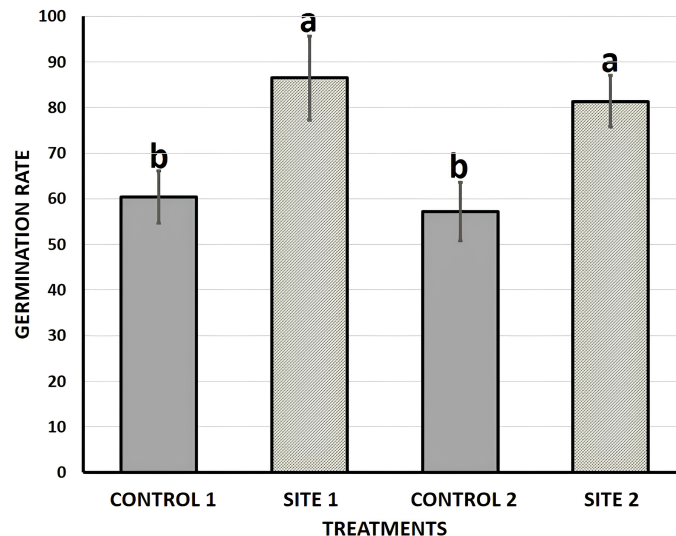
Subsequently, Tukey's multiple mean comparison test was applied for each ANOVA. The assumptions of normality and homogeneity of variances were verified using the Shapiro–Wilk and Levene

tests, respectively. The level of significance used was 0,05, and the statistical software employed was Statistica version 10.

## 3 Results and Discussion

### 3.1 Germination

*Typha latifolia* seeds subjected to pregerminative treatment—hydrated in saline water (1% NaCl) for 24 h and subsequently in distilled water for 20 days—showed a germination rate of  $86,5\% \pm 9,2\%$  (standard deviation) for seeds from Site 1, while those from Site 2 exhibited a similar germination rate of  $81,4\% \pm 5,9\%$ . In contrast, seeds hydrated only in distilled water for 20 days showed lower germination percentages, decreasing to  $60,4\% (\pm 5,7\%)$  for Control 1 and  $57,2\% (\pm 5,5\%)$  for Control 2 (Figure 3).



**Figure 3.** Germination percentage of *Typha latifolia* seeds from Yahuarcocha Lagoon. Control seeds (1 and 2, columns marked with b) were hydrated for 20 days, whereas seeds from Sites 1 and 2 (marked with a) were additionally subjected to a 24 h prehydration in saline water (1% NaCl) as a pregerminative treatment. Different letters represent significant differences among treatments ( $p < 0,05$ ).

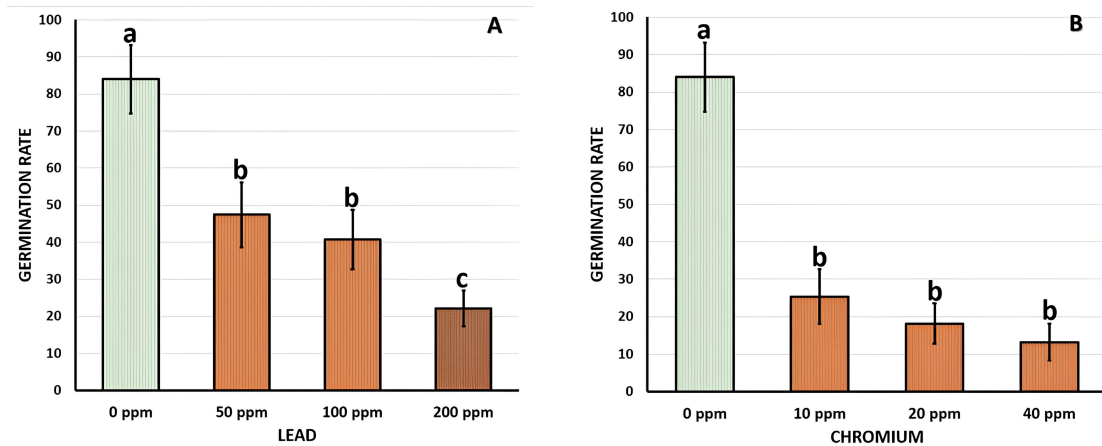
No significant differences were observed between the germination percentages of seeds from the two collection sites; however, significant differences were found between the pregerminative treatments from both sites (columns a) and their

respective controls (1 and 2), whose seeds did not receive pregerminative treatment (columns b).

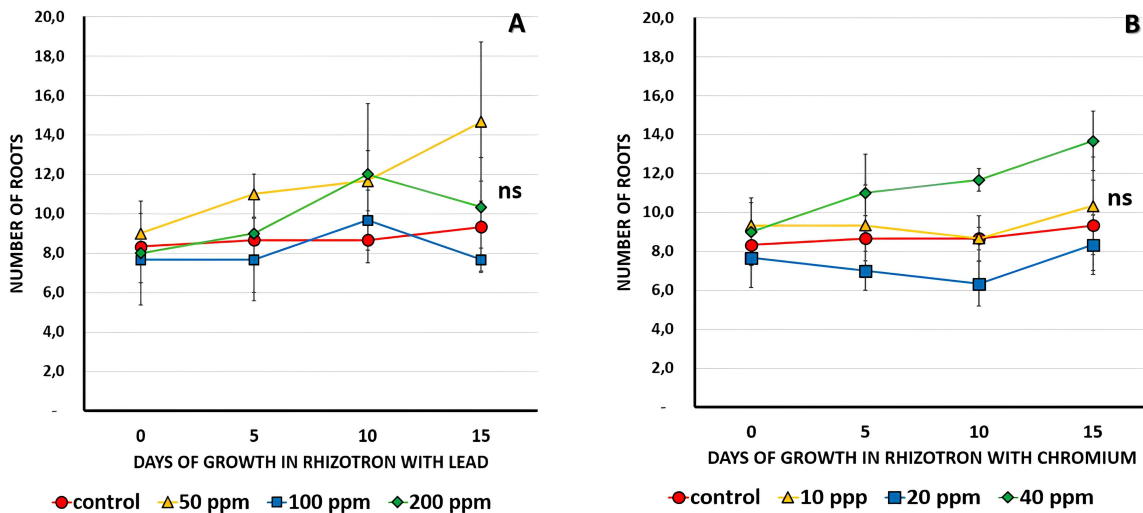
Regarding the germination percentages under different concentrations of heavy metals during the

hydration phase (Figure 4A), the results (presented with their respective standard deviations) for lead addition were  $47,4 \pm 8,7\%$  germination at 50 ppm,  $40,7 \pm 8,0\%$  at 100 ppm, and decreased to  $22,1 \pm 4,8\%$  at 200 ppm of lead. The germination percentage of the control treatment (pregerminative + hydration) was  $84,0 \pm 9,2\%$ . Statistically significant differences were observed between the control (a), the 50 and 100 ppm concentrations (b), and the 200 ppm lead concentration (c) (Figure 4A).

For chromium, germination at 10 ppm was low, and it reached  $25,4 \pm 7,3\%$ . At 20 ppm, germination decreased to  $18,2 \pm 5,3\%$ , and at 40 ppm it further declined to  $13,2 \pm 4,9\%$ . The germination percentage of the control treatment (pregerminative + hydration) was again  $84,0 \pm 9,2\%$  (Figure 4B). In this case, significant differences were found between the control treatment (a) and all three chromium concentrations (b): 10, 20, and 40 ppm.



**Figure 4.** Percentage of *T. latifolia* seeds germinated under treatments: A) lead, and B) chromium. Mean values and standard deviations are shown. Different letters indicate significant differences according to Tukey's test ( $p < 0,05$ ). The experiment was replicated three times ( $n = 3$ ).



**Figure 5.** Variation in the number of *T. latifolia* roots in the presence of different concentrations of lead (A) and chromium (B) in the growth medium. Mean values and standard deviations are shown. No significant differences were found according to Tukey's test at  $p < 0,05$ ;  $n = 3$ .

### 3.2 Root Architecture

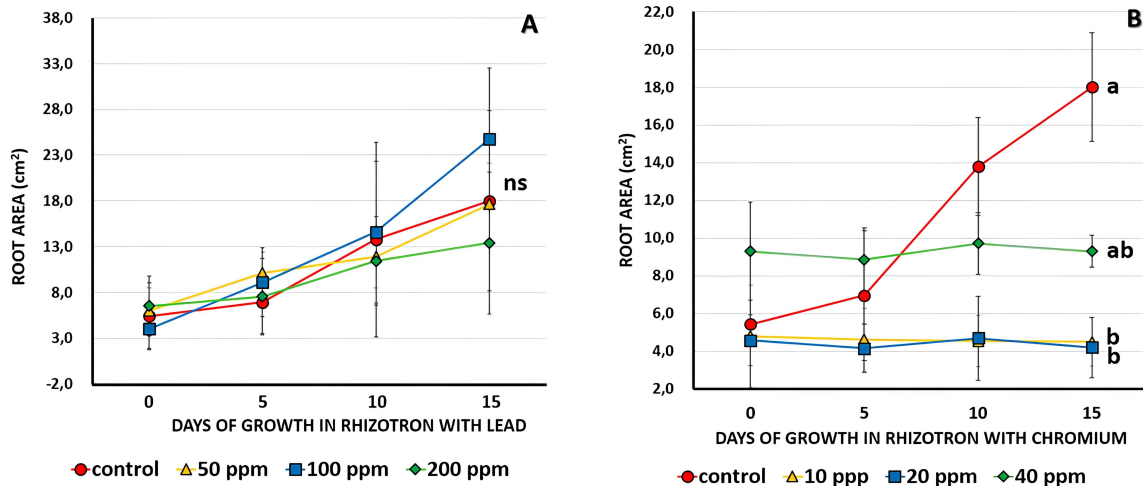
When evaluating the effect of lead on the number of roots of seedlings grown in the rhizotron, no statistically significant differences (ns) were detected, although the absolute number of roots increased from the day of transplanting (0 DAT) to 15 DAT, when the evaluation concluded (Figure 5A). The results (all reported with their respective standard deviations) showed that the control treatment slightly increased the number of roots from  $8,3 \pm 0,6$  to  $9,3 \pm 2,3$  at 15 DAT. The highest increase occurred at 50 ppm of lead, from  $9,0 \pm 1,0$  roots (at 0 DAT) to  $14,7 \pm 4,0$  roots at 15 DAT. At 100 ppm of lead, the average number of roots remained constant ( $7,7 \pm 0,9$ ) throughout the experiment. No significant differences were found among treatments (50, 100, and 200 ppm) or between these and the control (Figure 5A).

Similarly, a slight increase in root number was observed in the presence of chromium in the growth medium. However, unlike lead, the greatest increase occurred at the highest chromium concentration (40 ppm), where the number of roots increased slightly, from  $9,0 \pm 1,7$  to  $13,7 \pm 1,5$  by the end of

the experiment (15 DAT), without significant differences. No statistically significant differences were found among chromium treatments (10, 20, and 40 ppm) or between them and the control (Figure 5B).

The average total root area ( $\text{cm}^2$ ) increased by approximately three to six times over the 15-day evaluation period in all treatments—control and lead concentrations of 50, 100, and 200 ppm. The highest increase in total root area occurred at 100 ppm of lead, where values increased from  $4,00 \pm 2,1 \text{ cm}^2$  to  $24,8 \pm 7,8 \text{ cm}^2$ . Conversely, the smallest increase occurred at 200 ppm of lead, from  $6,5 \pm 3,3 \text{ cm}^2$  to  $13,4 \pm 7,7 \text{ cm}^2$ . However, no significant differences were detected among the four treatments (Figure 6A).

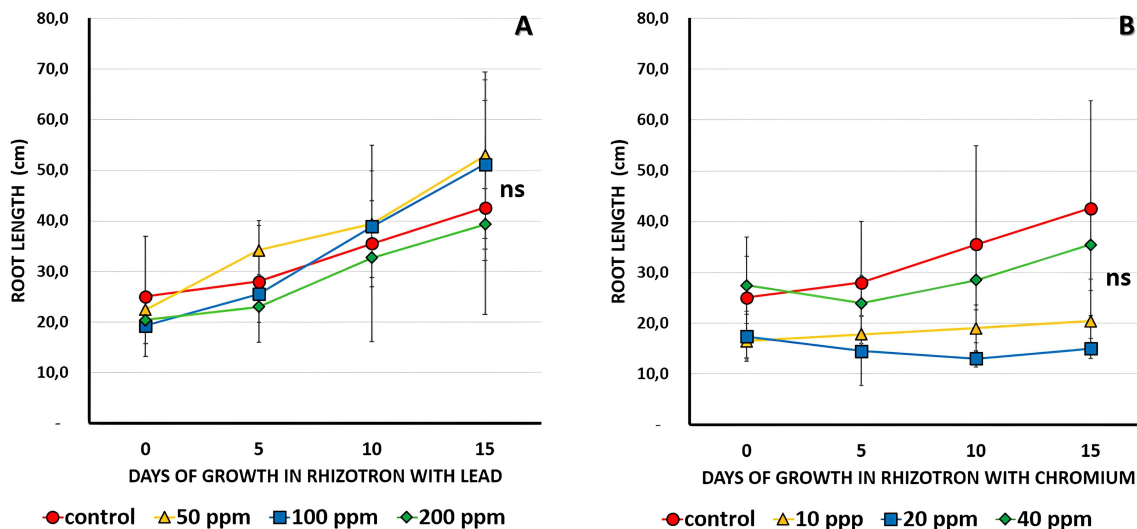
In the presence of chromium, no increase in total root area was recorded at any of the concentrations tested (10, 20, and 40 ppm), as the values remained constant (Figure 6B). Only the control treatment showed an increase in total root area, from  $5,4 \pm 3,7 \text{ cm}^2$  to  $18,0 \pm 2,9 \text{ cm}^2$ , resulting in significant differences between the control (a) and all three experimental treatments (b).



**Figure 6.** Variation in *T. latifolia* root area in the presence of different concentrations of lead (A) and chromium (B). Mean values and standard deviations are shown. Different letters indicate significant differences according to Tukey's test at  $p < 0,05$ ;  $n = 3$ .

Total root length (cm) under different lead concentrations in the growth medium showed a steady increase throughout the experiment across all treatments. This increase was slightly higher at 50 and 100 ppm of lead, with values rising from  $22,4 \pm 2,2$  cm at the beginning to  $52,9 \pm 16,4$  cm

at the end for 50 ppm, and from  $19,2 \pm 3,5$  cm to  $51,1 \pm 16,7$  cm for 100 ppm, respectively. No significant differences were observed among treatments (Figure 7A).



**Figure 7.** Variation in *T. latifolia* root length under different concentrations of lead (A) and chromium (B). Mean values and standard deviations are shown. No significant differences were detected according to Tukey's test at  $\alpha = 0,05$ ;  $n = 3$ .

In contrast, the addition of chromium to the Hoagland solution caused an increase in total root length at 40 ppm (from  $27,4 \pm 5,7$  cm to  $35,4 \pm 6,8$  cm) and a slight increase at 10 ppm (from  $16,5 \pm 3,6$  cm to  $20,4 \pm 6,1$  cm). Conversely, at 20 ppm of chromium, total root length decreased from  $17,4 \pm 4,9$  cm to  $15,0 \pm 1,9$  cm during the 15-day evaluation period, likely due to the death of previously counted lateral roots, which reduced the final mean length. The greatest increase was observed in the control treatment, where total root length grew from  $25,0 \pm 11,9$  cm to  $42,6 \pm 21,2$  cm over the 15-day growth period (Figure 7B).

### 3.3 Discussion

Multiple factors can affect the mother plant during seed development and induce changes in germination traits (Baskin and Baskin, 2014). In this context, phenotypic plasticity is considered one of the principal mechanisms by which seeds adapt to changing environmental conditions (Gratani, 2014).

For instance, traditional germination of mature *Typha latifolia* seeds has been carried out by simple hydration in distilled water; however, a pregerminative hydration in 1% NaCl solution for 24 h significantly increased germination relative to the traditional approach by an average of 25.2%, consistent with the improvements reported by Semenova (2014). This solution may act by neutralizing potential germination inhibitors and delaying the growth of microorganisms that negatively affect germination (Lorenzen et al., 2000). According to the USDA (2016), *T. latifolia* seeds attain high germination percentages when proper procedures are followed, and mechanical scarification is not recommended: as noted by the Royal Botanical Gardens, Kew (2016), scarification does not enhance germination because cattail seeds are sufficiently water-permeable to trigger the process. The high germination percentage may be explained by the absence of dormancy in this species (Stewart et al., 1997), although Nishihiro et al. (2004) report physiologi-

cal dormancy mechanisms in *Typha angustifolia*, a species closely related to *T. latifolia*, which could be common across the genus *Typha* (Baskin and Baskin, 2014).

No significant differences in germination were observed between the two sampling sites, likely because Yahuarcocha Lagoon is non-stratified: its waters mix throughout the year due to wind influence and inflowing streams, as indicated in the Lagoon Management Plan (IMI-UTN, 2012). Accordingly, based on *T. latifolia* germination, the two sites correspond to a single ecological population within the lagoon.

As for the effects of heavy metals on germination, the results (Figures 4A and 4B) show that both lead and chromium at the concentrations tested significantly reduced germination. Heavy metals often exert inhibitory effects on this process, as demonstrated by Oleszczuk (2008) and Pandey et al. (2008).

Lead, for example, reduced germination by 43 % at 50 ppm, and by 74 % at the highest concentration tested (200 ppm). This marked reduction may be attributable to lead (Pb), a non-essential metal that causes prolonged toxicity in plants, inducing adverse changes in seed morphology and germination as well as in enzymatic activities (Nas and Ali, 2018). These effects become more pronounced with increasing concentration and longer exposure. By reacting with sulfhydryl groups, lead inhibits enzyme activities and promotes the production of reactive oxygen species (ROS), causing oxidative bursts (Zulfiqar et al., 2019). In *Oryza sativa*, Gautam et al. (2010) found that reduced germination resulted from inhibition of  $\alpha$ -amylase activity, which in turn impaired starch degradation; similar effects may occur in grasses and graminoids (Sharma et al., 2022), including members of Typhaceae.

The reduction in germination was even greater in the presence of chromium: at only 10 ppm, germination decreased by 70 %, and at 40 ppm it declined by 85 %. Chromium's toxic effects compromise the physiology of seed germination (Zaranyika and Nyati, 2017). Nevertheless, Rout et al. (2000) showed that despite an initial reduction, seeds may develop tolerance and acclimation to chromium—particularly when mother plants originate from contaminated sites compared with uncontamina-

ted ones—traits characteristic of hyperaccumulator species.

Heavy-metal tolerance also depends on plant genetics. Studies by Cheng et al. (2008) on 12 *Oryza sativa* genotypes and by Zhang (2015) on aquatic and marsh species such as *Phragmites australis*, *Arundo donax*, *Cyperus alternifolius*, and *Alisma plantago-aquatica* reported broad variability in adaptive responses, although the germination index generally decreased significantly as chromium concentrations increased. Pan et al. (2008) note that responses lacking predictable patterns at higher levels of cadmium, chromium, and mercury are genotype dependent.

Regarding root architecture, rhizotron experiments revealed structural changes in the *T. latifolia* root system in response to different concentrations of lead and chromium. The Hoagland solution used as the growth medium allowed assessment of root architecture while approximating the physicochemical and nutritional conditions of Yahuarcocha Lagoon.

Post-embryonic roots formed within five days after transplanting and continued to grow at 15 and 20 days post-transplanting, establishing a highly branched and functional root system.

For seedlings exposed to different lead concentrations, the average number of roots did not decrease—even at 200 ppm (Figure 5A)—indicating strong tolerance to this metal. Similar findings were reported by Buta et al. (2011) in *Eichhornia crassipes* exposed to lead, where root development did not decline, supporting its status as a tolerant aquatic species utilized in wastewater treatment.

In contrast, studies in the aquatic species *Hydrocotyle umbellata* reported that exposure to chromium and lead at varying concentrations reduced total biomass productivity; at high chromium concentrations, roots turned dark brown and leaf necrosis occurred (Yongpisanphop et al., 2005). Likewise, in *Fagus sylvatica*, root elongation and biomass were significantly reduced in soils containing 44 ppm lead, and root architecture exhibited abnormal morphology with strongly inhibited root hair development (Breckle and Kahle, 1992; Kumar Yadav et al., 2021).

In *T. latifolia* seedlings exposed to chromium, the average number of roots did not decrease, again indicating tolerance to this heavy metal. However, by the end of the experiment, many seedlings displayed abnormal (stunted) root growth and pronounced leaf yellowing. In a comparable study in which *Arabidopsis thaliana* seedlings were grown under different chromium concentrations, plants were more tolerant at lower concentrations; however, gradual increases in chromium led to growth inhibition (Martínez-Trujillo et al., 2013). Similarly, Ortiz-Castro et al. (2007) reported that chromium in the root system and architecture of *A. thaliana* seedlings at concentrations above 200  $\mu\text{M}$  ( $\approx$  60 ppm—higher than in the present study) halted root growth and induced leaf chlorosis, an effect consistent with our observations.

Chromium is recognized as highly toxic to seedlings, impairing the absorption and transport of metals and nutrients from the soil, as extensively documented. For example, in *Pistia stratiotes*, chromium exposure produced toxic inhibition of growth, elongation, and normal root structure (Mufarrege et al., 2010); in *Cicer arietinum*, root length decreased progressively as chromium increased from 20 to 100 ppm (Medda and Kumar Mondal, 2017).

Root area (Figure 6A) and total root length (Figure 7A) increased slightly in the presence of lead, with no significant differences among treatments (including the control), corroborating the high tolerance of *T. latifolia* to lead and supporting its ecological use in removing nutrients and contaminants in natural or engineered water-treatment systems.

By contrast, chromium exposure halted seedling growth in the rhizotron throughout the 15-day evaluation, causing both total root area (Figure 6B) and total root length (Figure 7B) to remain constant. Meanwhile, the control showed a significant increase in root area, indicating normal growth when chromium is absent from the nutrient solution.

Overall, we found that both seed germination and root-system growth in *T. latifolia* exposed to lead persisted, indicating tolerance—at least for these two traits—and enabling survival in environments contaminated by this heavy metal (Branković et al., 2011). Seeds and seedlings exposed to chromium were less tolerant (in germination and root growth); how-

ever, tolerance may increase as plants develop under higher chromium concentrations (acclimation-based tolerance). These characteristics endow *T. latifolia* with attributes suitable for application in environmental remediation systems for contaminated waters.

## 4 Conclusions

Applying certain pregerminative treatments significantly increases germination percentage compared with seeds placed solely in distilled water. The *Typha latifolia* populations assessed likely belong to a single ecological population, as inferred from their germination performance under the applied treatments. Likewise, exposure to lead and chromium reduces germination percentage, particularly the latter.

Systematic use of rhizotrons enabled observation of sequential root development outside the natural habitat and control of key analytical variables. The metal concentrations tested did not alter root number or length; however, chromium exposure did affect total root area relative to the control, leading to the death of several seedlings—likely because reduced root area impaired processes such as water and nutrient uptake.

## Acknowledgments

We thank the staff of the Plant Physiology Laboratory at the University of Havana and Universidad Técnica del Norte.

## Author's contribution

**M.A.G.:** Conceptualization, Formal analysis, Investigation, Methodology, Visualization, Writing – original draft, Writing – review & editing. **A.V.S.:** Conceptualization, Formal analysis, Investigation, Methodology, Visualization, Writing – original draft, Writing – review & editing. **V.P.V.:** Conceptualization, Formal analysis, Methodology, Visualization, Writing – original draft, Writing – review & editing.

## References

- Armengaud, P., Zambaux, K., Hills, A., Sulpice, R., Pattison, R. J., Blatt, M. R., and Amtmann, A. (2009). Ez-rhizo: Integrated software for the fast and accurate measurement of root system architecture. *Plant Journal*, 57(5), 945–956. Online: <https://h7.cl/1nP4q>
- Baligar, V. C., Fageria, N. K., and Elrashidi, M. A. (1998). Toxicity and nutrient constraints on root growth. *HortScience*, 33(6), 960–965. Online: <https://h7.cl/1nP4t>
- Barceló, J., and Poschenrieder, C. (1992, January). Respuestas de las plantas a los contaminantes: “la fitorremediación”. *Suelo y Planta*, 2, 345–361. Online: <https://h7.cl/1nP4a>
- Baskin, C. C., and Baskin, J. M. (2014). *Seeds: Ecology, biogeography, and evolution of dormancy and germination*. Academic Press.
- Bourgeois, B., Hugron, S., and Poulin, M. (2012). Establishing a moss cover inhibits the germination of *Typha latifolia*, an invasive species, in restored peatlands. *Aquatic Botany*, 100, 76–79. Online: <https://h7.cl/1iOuv>
- Branković, S., Pavlović-Muratspahić, D., Topuzović, M., Glišić, R., Banković, D., and Stanković, M. (2011). Environmental study of some metals on several aquatic macrophytes. *African Journal of Biotechnology*, 10(56), 11956–11965. Online: <https://h7.cl/1iOv0>
- Breckle, S.-W., and Kahle, H. (1992). Effects of toxic heavy metals (cd, pb) on growth and mineral nutrition of beech (*Fagus sylvatica* L.). *Vegetatio*, 101, 43–53. Online: <https://h7.cl/1iOv6>
- Busch, J., Mendelssohn, I. A., Lorenzen, B., Brix, H., and Miao, S. (2006). A rhizotron to study root growth under flooded conditions tested with two wetland cyperaceae. *Flora: Morphology, Distribution, Functional Ecology of Plants*, 201(6), 429–439. Online: <https://h7.cl/1iOv7>
- Buta, E., Paulette, L., Mihăiescu, T., Buta, M., and Cantor, M. (2011). The influence of heavy metals on growth and development of *Eichhornia crassipes* species, cultivated in contaminated water. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, 39(2), 135–141. Online: <https://h7.cl/1nP4O>
- Chen, Y.-L., Hong, X.-Q., He, H., Luo, H.-W., Qian, T.-T., Li, R.-Z., ... Yu, H.-Q. (2014). Biosorption of cr (vi) by *Typha angustifolia*: Mechanism and responses to heavy metal stress. *Bioresource Technology*, 160, 89–92. Online: <https://h7.cl/1nP4S>
- Cheng, W., Zhang, G., Yao, H., and Zhang, H. (2008). Genotypic difference of germination and early seedling growth in response to cd stress and its relation to cd accumulation. *Journal of Plant Nutrition*, 31(4), 702–715. Online: <https://h7.cl/1iOvh>
- Franco, J. A., Bañón, S., Vicente, M. J., Miralles, J., and Martínez-Sánchez, J. J. (2011). Root development in horticultural plants grown under abiotic stress conditions - a review. *Journal of Horticultural Science and Biotechnology*, 86(6), 543–556. Online: <https://h7.cl/1iOvm>
- Gautam, M., Sengar, R. S., Chaudhary, R., Sengar, K., and Garg, S. (2010). Possible cause of inhibition of seed germination in two rice cultivars by heavy metals pb<sup>2+</sup> and hg<sup>2+</sup>. *Toxicological and Environmental Chemistry*, 92(6), 1111–1119. Online: <https://h7.cl/1nP50>
- Gratani, L. (2014). Plant phenotypic plasticity in response to environmental factors. *Advances in Botany, 2014*, 1–17. Online: <https://h7.cl/1iOvt>
- Han, J., Chen, F., Zhou, Y., and Wang, C. (2015). High pb concentration stress on *Typha latifolia* growth and pb removal in microcosm wetlands. *Water Science and Technology*, 71(11), 1734–1741. Online: <https://h7.cl/1iOvw>
- Hoagland, D. R., and Arnon, D. I. (1938). Preparing the nutrient solution. *The Water-Culture Method for Growing Plants Without Soil*, 347, 29–31.
- IMI-UTN. (2012). *Plan de manejo de la laguna de yahuarcocha* (Inf. Téc.). Dirección de Turismo del Ilustre Municipio de Ibarra – Facultad de Ingeniería en Ciencias Agropecuarias y Ambientales, Universidad Técnica del Norte.
- Kahle, H. (1993). Response of roots of trees to heavy metals. *Environmental and Experimental Botany*, 33(1), 99–119. Online: <https://h7.cl/1iOvE>
- Kumar Yadav, V., Santos-González, J., and Köhler, C. (2021). Int-hi-c reveals distinct chromatin architecture in endosperm and leaf tissues of *Ara-bidopsis*. *Nucleic Acids Research*, 49(8), 4371–4385. Online: <https://h7.cl/1nP5k>

- Lobet, G., Pagès, L., and Draye, X. (2011). A novel image-analysis toolbox enabling quantitative analysis of root system architecture. *Plant Physiology*, 157(1), 29–39. Online: <https://h7.cl/iOvM>
- Lorenzen, B., Tinerella, P. P., Newman, S., and Steward, K. K. (2000). Seed germination of two everglades species, *Cladium jamaicense* and *Typha domingensis*. *Aquatic Botany*, 66(3), 169–180. Online: <https://h7.cl/iOvU>
- Martínez-Trujillo, M., Cárdenas-González, J. F., González-Morales, S., and Flores-Hernández, A. (2013). Respuestas morfogénicas de las raíces de *Arabidopsis thaliana* al estrés de cr(vi). *Revista Chilena de Historia Natural*, 86, 207–219. Online: <https://h7.cl/iOvZ>
- Medda, S., and Kumar Mondal, N. (2017). Chromium toxicity and ultrastructural deformation of *Cicer arietinum* with special reference of root elongation and coleoptile growth. *Annals of Agrarian Science*, 15(3), 396–401. Online: <https://h7.cl/iOw2>
- Mufarrege, M. M., Hadad, H. R., and Maine, M. A. (2010). Response of *Pistia stratiotes* to heavy metals (cr, ni, and zn) and phosphorous. *Archives of Environmental Contamination and Toxicology*, 58(1), 53–61. Online: <https://h7.cl/iOwh>
- Nas, F., and Ali, M. (2018). The effect of lead on plants in terms of growing and biochemical parameters: A review. *MOJ Ecology & Environmental Sciences*, 3(4), 265–268. Online: <https://h7.cl/1nP62>
- Nishihiro, J., Teramura, A., Hosomi, M., and Nakagawa, M. (2004). Germination characteristics of lakeshore plants under an artificially stabilized water regime. *Aquatic Botany*, 79(4), 333–343. Online: <https://h7.cl/iOwI>
- Oleszczuk, P. (2008). Phytotoxicity of municipal sewage sludge composts related to physicochemical properties, pahs and heavy metals. *Ecotoxicology and Environmental Safety*, 69(3), 496–505. Online: <https://h7.cl/iOwO>
- Oquendo, R., Fernández, L., and Pérez, L. (2020). Concentración de plomo y cromo en poblaciones de *Typha latifolia* L. (typhaceae) en el lago yahuarcocha; imbabura. *Revista Cubana de Ciencias Biológicas*, 8, 1–12. Online: <https://h7.cl/iOwR>
- Ortiz-Castro, R., Contreras-Cornejo, H. A., Macías-Rodríguez, L., and López-Bucio, J. (2007). Effects of dichromate on growth and root system architecture of *Arabidopsis thaliana* seedlings. *Plant Science*, 172(4), 684–691. Online: <https://h7.cl/1nP6B>
- Pal Singh, H., Du, J., and Verma, S. K. (2013). Chromium toxicity and tolerance in plants. *Environmental Chemistry Letters*, 11(3), 229–254. Online: <https://h7.cl/iOwZ>
- Pan, X. B., Zhu, C., and Cheng, C. (2008). Assessment of techniques for screening alfalfa cultivars for aluminum tolerance. *Euphytica*, 164(2), 541–549. Online: <https://h7.cl/iOx0>
- Pandey, S. N., Nautiyal, B. D., and Sharma, C. P. (2008). Pollution level in distillery effluent and its phytotoxic effect on seed germination and early growth of maize and rice. *Journal of Environmental Biology*, 29(2), 267–270. Online: <https://h7.cl/iOx5>
- Rout, G. R., Samantaray, S., and Das, P. (2000). Effects of chromium and nickel on germination and growth in tolerant and non-tolerant populations of *Echinochloa*. *Chemosphere*, 40, 855–859. Online: <https://h7.cl/1nP6N>
- Royal Botanical Gardens, Kew. (2016). *Seed information database*. Online: <http://data.kew.org/sid/>
- Sasmaz, A., Obek, E., and Hasar, H. (2008). The accumulation of heavy metals in *Typha latifolia* grown in a stream carrying secondary effluent. *Ecological Engineering*, 33(3-4), 278–284. Online: <https://h7.cl/1nP6T>
- Semenova, A. (2014). *Method for producing aquatic plants for the purification of contaminated water* (EP: PCT n.º WO131930 A1).
- Sharma, A., Singh, H., Kaur, N., Singh, N., Singh, P., and Bhardwaj, R. (2022). Heavy metal induced regulation of plant biology: Recent insights. *Physiologia Plantarum*, 174(e13688). Online: <https://h7.cl/1nP6Z>
- Smith, P. P. (2018). *The book of seeds: A life-size guide to six hundred species from around the world*. The University of Chicago Press.

- Stewart, H., Miao, S. L., Colbert, M., and Carraher, C. E. (1997). Seed germination of two cattail (*Typha*) species as a function of everglades nutrient levels. *Wetlands*, 17, 116–122. Online: <https://h7.cl/1nP72>
- USDA. (2016). *Plants database | usda plants guide*. Online: <http://plants.usda.gov/java/>
- Yongpisanphop, J., Kruatrachue, M., and Pokethitiyook, P. (2005). Toxicity and accumulation of lead and chromium in *Hydrocotyle umbellata*. *Journal of Environmental Biology*, 26(1), 79–89. Online: <https://h7.cl/1iOxs>
- Zaidi, A., Ahmad-Wani, P., and Saghir-Khan, M. (2012). *Toxicity of heavy metals to legumes and bioremediation*. Springer.
- Zaranyika, M. F., and Nyati, W. (2017). Uptake of heavy metals by *Typha capensis* from wetland sites polluted by effluent from mineral processing plants: implications of metal–metal interactions. 3 *Biotech*, 7(5), 1–11. Online: <https://h7.cl/1nP7a>
- Zhang, G. (2015). Influence of heavy metal wastewater on seed germination of different wetland plants. *Journal of China Agricultural University*, 20, 167–178. Online: <https://h7.cl/1nP7c>
- Zulfiqar, U., Farooq, M., Hussain, S., Maqsood, M., Hussain, M., Ishfaq, M., ... Anjum, M. Z. (2019). Lead toxicity in plants: impacts and remediation. *Journal of Environmental Management*, 250, 109557. Online: <https://h7.cl/1nP7j>