# HEAVY METALS ACCUMULATION IN THE TISSUES OF THE COMMON REED (*PHRAGMITES AUSTRALIS*)

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#### Abstract

The aim of the present study was to provide a comparative analysis related to the concentration of heavy metals (Cd, Pb, Ni, Cr) in the tissues (stem, leaves, panicle) of the common reed (Phragmites australis). The reed samples were collected from 10 different sampling stations located in Danube Delta Reservation Biosphere, 2 sampling station from Lake Brateş and 2 sampling station situated on a tributary of the Danube River (Chineja River), Romania. The concentration of heavy metals varied depending on the location, the lowest values were found in plants located on the course of the Chineja River. As well, depending on the plant structure, was observed that the panicle contained the lowest concentrations of the chosen heavy metals for the experiment.

Key words: bioaccumulation, common reed, heavy metals.

## INTRODUCTION

Reed is a perennial plant that can be found in temperate and tropical regions, especially in areas with high humidity. It spreads in all environments with water levels close to the soil surface (irrigated and drained agricultural or non-agricultural areas, swamps, ponds) (Vymazal, 2013).

The plant is dominant mainly in Europe, but also widespread in North America and various regions of South America and Australia (Srivastava et al., 2014; Mykleby et al., 2016).

Common reed (*Phragmites australis* (Cav.) Trin. ex Steud.), has a resistant rhizome system with high propagation capacity and long growth period, increased adaptability to different environmental conditions and strong resistance to pollution (Fraser et al., 2004; Liu et al., 2012).

The reed stem can grow up to 6 m with the ability to survive in high concentrations of toxic contaminants (Bragato et al., 2009).

The way in which reed adapts to environmental conditions (water depth, pH, salinity, pollutants, extreme climate) is through changes in the physiological, morphological, and behavioural characteristics. With diverse patterns in regard to the reproductive and physiological adaptations, *Phragmites australis* can be used successfully in phytoremediation of depredated wetland ecosystems (Qin et al., 2022)

Heavy metals and other pollutants end up in the Danube Delta, after being collected from the entire hydrographic basin, which represents over 800,000 km<sup>2</sup>. The reed is an integral part of the ecology and economy of the Danube Delta by creating an important habitat and by phytoremediation of the Danube waters.

Phytoremediation represents the remediation of contaminated soil, sediments, or water using plants. Compared to other chemical and physical methods of cleaning up, phytoremediation is more environmentally friendly.

Depending on the mechanism of heavy metals removal, phytoremediation can be classified into:

• phytoextraction - heavy metals are stored in the leaves of plants (Lee, 2013);

• phytostabilization - toxic pollutants are transformed into nontoxic or less toxic substances (McSorley et al., 2016); • phyto immobilization - the mobility of heavy metals is reduced by sequestering them within the rhizosphere (Yadav et al., 2018).

The species selected for phytoremediation must have the ability to accumulate a vast range of heavy metals, a high tolerance and a high growth rate (Darajeh et al., 2017; Kushwaha et al., 2018).

*Phragmites australis* is a macrophyte well known for the capacity to uptake, translocate, and accumulate heavy metals in the plant tissues, both from water and sediments (Bonanno & Giudice, 2010; Huang et al., 2018).

Southichak et al., 2016, reported that reed biomass is an effective biosorbent with high sorption performance even if the heavy metals are found in low concentrations.

The current study presents the heavy metals (Cd, Pb, Ni, Cr) concentrations measured in the leaves, stems and panicles of Phragmites australis, sampled from the Danube Delta, Brateş Lake and Chineja River.

## MATERIALS AND METHODS

In order to evaluate the concentration of heavy metals from reed, plant samples were collected from 10 locations in the Danube Delta, 2 locations were in the Brateş Lake, and 2 locations were on the course of a tributary river, Chineja River (near the villages of Tuluceşti and Folteşti), that flows in the Brateş Lake and connects it to the Danube. The coordinates of the sampling points and the codes used to identify them are presented in Table 1. In Figure 1, the sampling points are represented on the map, to present their spatial distribution.

Reed stems, leaves and panicles were collected from the selected sampling points, during the autumn harvest campaign of October 2022.

Plant material was harvested from a 100 m  $\times$  100 m area in each location, and homogenous samples were obtained from randomly selecting stems, leaves, and panicles from each location.

In order to assess the heavy metals quantity in the organs of common reed, the samples digestion is done beforehand. The digestion process is achieved using the digestion oven Topwave Analytikjena Germany. Teflon containers with the samples were subjected to a digestion protocol specific to plants.

Heavy metals were determined from the matrix resulted after the samples digestion. An ANALYTIK JENA ContrAA 700<sup>®</sup> was used, which is an atomic absorption spectral photometer. Samples were analysed in triplicate and the results are express as average  $\pm$  standard deviation.

One-way classification variance analysis and ANOVA tests were used, to highlight statistical differences. When differences were statistically significant ( $p \le 0.05$ ), Tukey's post hoc test was applied to identify which group differs.

Nr.	Coordinates Danube Delta		Code	Coordinates Brateş Lake		Code	Coordinates Chineja River		Code
INI.	Latitude	Longitude	Code	Latitude	Longitude	Code	Latitude	Longitude	Code
1	45°09'32.4"N	29°30'57.6"E	DDa	45°27'45.0"N	28°05'52.5"E	BLa	45°34'19.7"N	28°03'15.2"E	CRa
2	45°09'00.0"N	29°25'55.2"E	DDb	45°30'17.0"N	28°02'30.3"E	BLb	45°43'59.9"N	28°04'27.7"E	CRb
3	45°10'48.0"N	29°25'22.8"E	DDc	-	-	-	-	-	-
4	45°07'55.2"N	29°31'22.8"E	DDd	-	-	-	-	-	-
5	45°10'26.4"N	29°28'22.8"E	DDe	-	-	-	-	-	-
6	45°19'45.7"N	29°24'28.8"E	DDf	-	-	-	-	-	-
7	45°03'06.0"N	29°29'42.4"E	DDg	-	-	-	-	-	-
8	45°06'07.9"N	29°38'31.2"E	DDh	-	-	-	-	-	-
9	44°59'57.9"N	29°24'08.7"E	DDi	-	-	-	-	-	-
10	45°15'14.9"N	29°12'47.9"E	DDj	-	-	-	-	-	-

Table 1. The coordinates of the sampling points and the codes used to identify them

Scientific Papers. Series E. Land Reclamation, Earth Observation & Surveying, Environmental Engineering. Vol. XII, 2023 Print ISSN 2285-6064, CD-ROM ISSN 2285-6072, Online ISSN 2393-5138, ISSN-L 2285-6064



🛇 BLa 🛇 BLb 🛇 CRa 🛇 CRb

Figure 1. Map representation of the sampling points and their spatial distribution

#### **RESULTS AND DISCUSSIONS**

Cadmium was found, generally, in the highest quantity in the reed leaves, followed by stems and panicles. The highest concentration of cadmium was present in the samples from the Brateş Lake (BLa and BLb), with the lowest concentration in the samples from the Chineja River (CRa and CRb).

This trend is observed in all the vegetal parts of the reed. In the Danube Delta, the highest concentrations of cadmium were found in the sampling points closest to the river flow (DDe>DDb>DDc>DDj>DDa>DDd>DDi>DD h>DDf>DDg).

There were no significant differences between the locations from Chineja River, or the locations from Brateş Lake ( $p \ge 0.05$ ).

In the Danube Delta, there were significant differences between all locations for all plant organs ( $p \le 0.05$ ), phenomenon explained by the considerable distance between some of the locations, the flow regime of the river, proximity to human settlements or transportation lanes. Table 2 presents the data obtained following the analysis for cadmium.

Cadmium					
Stems	Leaves	Panicles			
0.70399±0.00018	1.07435±0.00087	0.64935±0.00025			
0.56465±0.00046	1.27301±0.00074	0.74599±0.00030			
0.42208±0.00033	1.12328±0.00038	0.68247±0.00042			
0.78989±0.00051	1.00977±0.00050	0.52130±0.00054			
0.97810±0.00020	1.28461±0.00044	0.75758±0.00023			
0.39432±0.00011	0.84665±0.00055	0.19883±0.00031			
0.48828±0.00037	0.70770±0.00029	0.17904±0.00053			
0.44479±0.00017	0.89962±0.00038	0.30804±0.00025			
0.45650±0.00026	0.95462±0.00011	0.35827±0.00019			
0.78449±0.00040	1.08430±0.00068	0.68019±0.00022			
1.02389±0.00032	1.49960±0.00057	0.86777±0.00049			
1.08836±0.00026	1.52588±0.00033	0.93618±0.00010			
0.29314±0.00037	0.45532±0.00044	0.25523±0.00072			
0.27168±0.00039	0.42615±0.00056	0.21535±0.00081			
	0.70399±0.00018 0.56465±0.00046 0.42208±0.00033 0.78989±0.00051 0.97810±0.00020 0.39432±0.00011 0.48828±0.00037 0.44479±0.00017 0.45650±0.00026 0.78449±0.00040 1.02389±0.00032 1.08836±0.00026 0.29314±0.00037	Stems         Leaves           0.70399±0.00018         1.07435±0.00087           0.56465±0.00046         1.27301±0.00074           0.42208±0.00033         1.12328±0.00038           0.78989±0.00051         1.0077±0.00050           0.78989±0.00020         1.28461±0.00044           0.39432±0.00011         0.84665±0.00025           0.48828±0.00037         0.70770±0.00029           0.44479±0.00017         0.89962±0.00038           0.44479±0.00026         0.95462±0.00018           1.02389±0.00032         1.49960±0.00057           1.08336±0.00026         1.52588±0.00033           0.23914±0.00037         0.45532±0.00044			

Table 2. Cadmium concentrations in the organs of common reed, *Phragmites australis*, present in the sampling locations of the experiment

Data is presented as average  $\pm$  standard deviation

Lead presented a similar trend of phytoaccumulation as cadmium (leaves>stems>panicles), presented in table 3. The highest concentrations were found in the samples from Danube Delta (DDe>DDd>DDc), even though the samples from the Brateş Lake had higher concentrations of lead than some locations from the Danube Delta. The lowest concentrations of lead were found in the samples from the Chineja River, for all the plant organs. Table 3. Lead concentrations in the organs of common reed, *Phragmites australis*, present in the sampling locations of the experiment

	Lead					
μg/g	Stems	Leaves	Panicles			
DDa	2.01140±0.00044	2.98431±0.00076	1.49850±0.00025			
DDb	2.04583±0.00034	3.13357±0.00086	1.77173±0.00029			
DDc	3.46320±0.00055	5.61640±0.00028	2.39464±0.00031			
DDd	4.42338±0.00021	6.35179±0.00064	1.95976±0.00025			
DDe	3.65075±0.00036	6.60013±0.00033	1.61186±0.00019			
DDf	1.57729±0.00030	3.03514±0.00094	0.66278±0.00036			
DDg	1.67411±0.00041	3.94997±0.00061	1.30208±0.00022			
DDh	2.30061±0.00011	4.26136±0.00055	1.13489±0.00030			
DDi	2.64638±0.00027	3.87382±0.00077	1.57638±0.00025			
DDj	2.77918±0.00036	5.95602±0.00024	1.70474±0.00031			
BLa	2.50954±0.00022	4.02576±0.00050	1.65289±0.00035			
BLb	2.55138±0.00029	4.06382±0.00085	1.79135±0.00029			
CRa	1.46570±0.00017	1.95137±0.00066	0.55050±0.00019			
CRb	1.32943±0.00013	1.89554±0.00019	0.48392±0.00017			
CRb		$1.89554 \pm 0.00019$				

Data is presented as average  $\pm$  standard deviation

There were no statistical differences between the samples from Chineja River (CRa and CRb), or the samples from Brateş Lake (BLa and BLb),  $p \ge 0.05$ . There were significant differences between all locations for all plant organs in the Danube Delta ( $p \le 0.05$ ).

Nickel is the metal with the highest concentration, from the analysed 4 heavy metals, measured in the biological samples. The Brateş Lake has the highest concentration of nickel in the tissues of common reed, more than twice the amount measured in the Chineja River. There were no significant differences between the locations from the Chineja River (CRa and CRb,  $p \ge 0.05$ ), and significant differences for nickel found in the reed leaves and panicles from the locations in the Brateş Lake ( $p \le 0.05$ ). In the Danube Delta, the differences varied greatly with statistical significance,  $p \le 0.05$ .

Table 4. Nickel concentrations in the organs of common reed, *Phragmites australis*, present in the sampling locations of the experiment

	Nickel					
μg/g	Stems	Leaves	Panicles			
DDa	3.60375±0.00055	7.33288±0.00059	2.59740±0.00046			
DDb	5.31915±0.00068	7.63807±0.00046	3.82320±0.00022			
DDc	4.87013±0.00081	8.84583±0.00019	3.35249±0.00047			
DDd	4.26540±0.00063	8.30619±0.00031	2.48236±0.00096			
DDe	4.86766±0.00044	9.65873±0.00032	1.77305±0.00055			
DDf	2.99685±0.00071	5.91054±0.00048	1.59067±0.00025			
DDg	2.37165±0.00040	5.43120±0.00017	1.62760±0.00033			
DDh	2.45399±0.00033	5.99747±0.00019	1.29702±0.00043			
DDi	4.96196±0.00040	6.64084±0.00044	2.14961±0.00055			
DDj	4.29510±0.00028	8.70495±0.00088	3.06853±0.00018			
BLa	5.11945±0.00013	9.86312±0.00093	7.85124±0.00010			
BLb	5.18659±0.00027	10.11699±0.00077	8.00593±0.00066			
CRa	2.83369±0.00009	4.25298±0.00006	0.90081±0.00028			
CRb	2.77594±0.00042	4.01019±0.00011	0.84668±0.00031			

Data is presented as average  $\pm$  standard deviation

Chromium was found with the lowest concentration in the samples from the Chineja River (CRa and CRb), with no significant differences between locations, for all reed organs ( $p \ge 0.05$ ). The leaves of *Phragmites australis* accumulated the highest concentration of chromium, followed by stems and panicles. highest value measured The for the concentration of chromium was in the leaves of common reed from the location BLb. The highest concentrations of chromium from stems and panicles were also measured in the same location BLb. Data collected from the Danube Delta presented significant differences between locations, for all the vegetal organs ( $p \le 0.05$ ).

Table 5. Chromium concentrations in the organs of common reed, *Phragmites australis*, present in the sampling locations of the experiment

	Chromium					
μg/g	Stems	Leaves	Panicles			
DDa	2.34663±0.00049	3.49591±0.00082	1.39860±0.00011			
DDb	1.39116±0.00057	3.81904±0.00076	0.65274±0.00011			
DDc	1.29870±0.00066	3.36984±0.00093	0.59866±0.00024			
DDd	2.05371±0.00022	3.42020±0.00021	1.17586±0.00019			
DDe	2.58594±0.00035	3.86349±0.00050	1.12830±0.00005			
DDf	1.89274±0.00041	3.67412±0.00068	0.79533±0.00038			
DDg	1.39509±0.00021	3.29164±0.00034	0.81380±0.00019			
DDh	1.22699±0.00010	2.99874±0.00028	0.64851±0.00016			
DDi	1.32319±0.00061	2.49032±0.00071	0.85985±0.00024			
DDj	1.38959±0.00013	3.35980±0.00015	0.85237±0.00011			
BLa	2.60992±0.00022	3.92512±0.00025	1.85950±0.00029			
BLb	2.75292±0.00010	4.16285±0.00033	1.90047±0.00021			
CRa	1.17256±0.00011	2.40168±0.00048	0.25023±0.00009			
CRb	1.16014±0.00017	2.37629±0.00061	0.24012±0.00009			

Data is presented as average  $\pm$  standard deviation

Heavy metals decreased in the order leaves > stems > panicles, in all the locations selected for the experiment. This trend was also observed by Vymazal and Březinová (2016), Parzych et al. (2016), Rzymski et al. (2014), Ganjali el al. (2014).

The heavy metals uptake by the organs of P. australis were: stems Ni>Pb>Cr>Cd, leaves Ni>Pb>Cr>Cd and panicles Ni>Pb>Cr>Cd.

Bonanno, 2013, found more Pb and Cr than Ni in the stems and the same order in leaves, when comparing the capacity of heavy metals removal by P. australis.

These results are like those obtained by Astel et al. (2014), Jiang et al. (2018), Dan et al. (2017). The strategies used for the uptake of heavy metals are dependent on several factors: pH, metal type, temperature, and depth of the contamination (Ashraf et al., 2017; da Conceição et al., 2016).

Phragmites australis is a valuable plant that can be used successfully for the phytostabilization of trace metals due to the high bioaccumulation capacity, the ability to translocate these metals, and limited mobility. This is because of the different accumulation levels and transport systems, with very specific metal-affinity patterns (Klink, 2017; Pérez-Sirvent et al., 2017).

The capacity of P. australis to bioaccumulate is also dependent on the season, as observed by Grisey et al., 2012.

## CONCLUSIONS

The present study confirms that *P. australis* is a suitable candidate for biomonitoring of heavy metals pollution in aquatic environments. As well, the capacity to bioaccumulate heavy metals in the tissues above the ground and the resistance to metal toxicity, makes this plant a perfect solution for phytoremediation of contaminated environments.

The heavy metals uptake by the organs of *P. australis* were: stems Ni>Pb>Cr>Cd, leaves Ni>Pb>Cr>Cd and panicles Ni>Pb>Cr>Cd.

Further studies should be conducted in an integrated manner, considering different biotic and abiotic components of aquatic environments to obtain a clear view on heavy metals accumulation, transfer and dispersion in water systems.

### ACKNOWLEDGMENTS

The present research was supported by the project An Integrated System for the Complex Environmental Research and Monitoring in the Danube River Area, REXDAN, SMIS code 127065, co-financed by the European Regional Development Fund through the Competitiveness Operational Programme 2014-2020, contract no. 309/10.07.2021.

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Scientific Papers. Series E. Land Reclamation, Earth Observation & Surveying, Environmental Engineering. Vol. XII, 2023 Print ISSN 2285-6064, CD-ROM ISSN 2285-6072, Online ISSN 2393-5138, ISSN-L 2285-6064

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