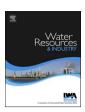
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Effect of substrates on the potential of *Phragmites australis* to accumulate and translocate selected contaminants from landfill leachate

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ABSTRACT

The choice of constructed wetlands substrate is a very important issue, there is still a lack of studies that analyse the effect of different types of substrate on the efficiency of the accumulate and translocate contaminants from landfill leachate. Therefore, the objectives of study were to evaluate the effect of substrate on the potential of *Phragmites australis* to accumulate and translocate selected contaminants from leachate. The lowest translocation was recorded for Fe and Cu, and the highest for K and Ni. Translocation between root and shoots for most parameters (total Kjeldahl nitrogen, total phosphorus, K and Mn) was high (TF > 1.0). However, for Fe, Cu, Cr and Ni translocation was low in most variants (TF < 1.0). Only for Cr there were statistically significant differences between the substrates. The obtained bioaccumulation factor values for Cu, Cr and Ni show that *P.australis* does not have a good ability to phytoaccumulate heavy metals from leachate.

1. Introduction

Many industrial sectors generate wastewater [1–4] which, in excessive quantities, can cause serious environmental damage. Municipal solid waste (MSW) leachate can contain a wide range of contaminants, among them soluble organic compounds, inorganic contaminants, suspended solids, heavy metals and hazardous substances [5,6]. The most commonly reported include ammonium nitrogen, chlorides, sulphates and heavy metals [7–9]. To reduce the negative impact of landfill leachate on the environment, different treatment methods are used: biological, physical and chemical [10].

Particularly noteworthy are solutions based on the use of plants to, among other things, reduce, remove or immobilise pollutants, i. e. phytoremediation in its broadest sense. CW are classified as biological methods using phytoremediation for treatment of, among others, contaminated liquids [11,12]. They are designed and constructed to use natural processes to remove contaminants in a more controlled environment [13]. The main mechanisms of contaminant removal in CW include phytofiltration, phytoextraction, phytostabilisation, phytovolatilisation, phytodesalination and phytodegradation [12,14]. Some of the most important measures of phytoremediation effectiveness include the bioconcentration factor (the ratio of the concentration of a compound in the external environment to the concentration found in plant tissue) and the translocation factor (the ratio of the concentration in the shoots to the

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content in the roots of the plant) [15,16].

CWs have been successfully treating various types of wastewater for decades. Their popularity is due to their low maintenance costs, low energy input, high pollutant removal efficiency and environmental friendliness, among others [17]. Due to their numerous advantages, they have been recognised as a very good solution for wastewater treatment [18].

Plants play an important role in CW, which should have a high capacity to survive in potentially toxic and variable conditions [19]. In recent years, *P. australis* (common reed) is very often used in phytoremediation of contaminated water, soil and sediments [20]. This species is classified as a common emergent macrophyte found on almost all continents and has a high capacity to adapt to changing environmental conditions [21]. Furthermore, it is an inexpensive and undemanding species with a fast growth rate, high biomass production and a deep root system [22,23]. Also, the correct choice of substrate in CW is very important, among other things because it forms basis for the growth of plants and microorganisms. In substrates, filtration, adsorption, sedimentation, flocculation, precipitation and ion exchange take place. They are also an inexpensive and plant-safe way to enhance phytoremediation, including phytostabilisation. Research shows that the right choice of substrates can significantly improve the efficiency of contaminant removal in CW [24]. Both natural materials, such as zeolite or pine bark (which can be a by-product of timber industry), and specially produced materials, such as expanded clay, are used as substrates in CW [25]. Mineral substrates perform well in CW due to their high adsorption capacity, while organic substrates promote plant establishment and growth due to their organic and nutrient content [26,27].

To date, many studies have been conducted on the treatment of different types of wastewater in CW. Phytoremediation has also been addressed many times, e.g. from tannery wastewater, by *Penisetum purpureum*, *Brachiaria decumbens* and *Phragmites australis* in CW gravel beds [28] boron-containing leachate by different plant species (four poplar and willow species) in CW [29] or from mercury-contaminated water by *Typha domingensis* in CW [18]. Phytoremediation from landfill leachate (LL) was studied, evaluating e. g. phytoremediation of partially treated MSW leachate by selected macrophytes in subsurface vertical flow CW systems (VFCW) (K.R.S and [30], phytoremediation of landfill leachate using water hyacinth and water lettuce [31] or removal of organic pollutants by effluent recirculation CW system treating landfill leachate [32].

Although the treatment of different types of wastewater in CW using a variety of plant species has already been the subject of many studies, but there is still a need for research to further understand the treatment mechanisms in CW [29]. The choice of CW substrates is a very important issue, because a properly selected substrate can be a good way to improve the phytoremediation process, there is still a lack of studies that analyse the effect of different types of substrate on the efficiency of the phytoremediation of LL [33].

Enhancement of the phytoremediation process through the role of substrates in CW still requires further research [24].

The few studies that have been conducted so far have analysed the effect of the applied substrate on the potential of plants in CW to accumulate and translocate selected pollutants from landfill leachate. The study of accumulation and translocation in CW is important to better understand and evaluate this treatment process.

Therefore, the authors decided to conduct research. The objectives of this study were: (I) evaluate off the effect of applied substrate on the potential of *P.australis* to accumulate and translocate selected contaminants from LL in CW, and (II) determination of the bioaccumulation and translocation of elements in *P. australis* tissues.

2. Materials and methods

2.1. Landfill leachate used in the study

Two municipal waste landfills with leachate drainage systems located in the Lower Silesia Province, Poland, were selected for the study.

One of the landfills located in Legnica (N 51° 14' 21.317'' E 16° 11' 0.251'') has been in operation since 1977 until now. It has a total capacity of 2.3 million m^3 and the total area is 14.2 ha. The second storage site located in Jawor (N 51° 3' 56.112'' E 16° 12' 38.927'')

 Table 1

 Physicochemical characteristics of the untreated landfill leachate supplied to the CW systems (extracted from the Bioresource Technology [33].

| Parameter | Unit | Raw leachate from landfill | | Landfill le | eachate at 25% concentration | Max. limit values (regulation ME) ^a | | |
|-----------|-------------------------|----------------------------|---------|-------------|------------------------------|--|--|--|
| | | Jawor | Legnica | Jawor | Legnica | | | |
| pН | _ | 8.3 | 9.1 | 8.4 | 8.8 | 6.5–9.0 | | |
| EC | μS/cm | 8070 | 5730 | 2660 | 2010 | - | | |
| TN | mg N/l | 325.3 | 169.27 | 81.99 | 42.59 | 30 | | |
| TKN | mg N/l | 323.53 | 167.16 | 79.69 | 40.14 | - | | |
| AN | mg N-NH ₄ /l | 320.35 | 127.73 | 75.56 | 29 | 10 | | |
| TP | mg P/l | 17.61 | 10.49 | 1.6 | 0.93 | 2 | | |
| K | mg/l | 561.8 | 407.5 | 138.3 | 94.1 | 80 | | |
| Fe | mg/l | 6.11 | 0.91 | 1.67 | 0.28 | 10 | | |
| Mn | mg/l | 0.75 | 0.13 | 0.18 | 0.03 | - | | |
| Ni | mg/l | 95 | 133.8 | 67.9 | 75.6 | 0.5 | | |
| Cr | mg/l | 78.5 | 108.4 | 66.6 | 59.3 | 0.1 | | |
| Cu | mg/l | 25.5 | 62.3 | 56.3 | 54.7 | 0.5 | | |
| Pb | mg/l | 0.005 | 0.005 | 0.005 | 0.005 | 0.5 | | |
| Cd | mg/l | 0.005 | 0.005 | 0.005 | 0.005 | 0.2 | | |

^a Max. limit values in accordance with Regulation of ME of 12 July 2019.

has been in operation since 1977 and was closed at the end of 2020. Its total capacity is 231,000 m³, while the total area is 3.37 ha. The leachate from these landfills was not highly contaminated, but in the case of most parameters even after dilution (25% landfill leachate - LL) it did not meet the requirements imposed by the Polish law (Regulation of ME of 12 July 2019), which would allow its direct discharge into water or soil. Therefore, before discharge into the environment it was necessary to treat them.

The prepared models of CW were dosed with raw leachates from both landfills described above (Legnica and Jawor) and leachate at 25% concentration (diluted with tap water) also from both landfills. The final choice of dilution (25%) was based on previous toxicity analyzes and studies on the physicochemical composition and leachate treatment in CW systems.

Previously, the following concentrations of leachate samples were carried out in studies: 6.25, 12.5, 25, 50 and 100%. A similar range of leachate concentrations was also selected in several publications [34,35].

Table 1 extracted from the research conducted by Ref. [33] presents selected physicochemical parameters of the leachates used in this study, as well as the legal requirements to be met when discharging the leachate to water or to the ground (Regulation of ME of 12 July 2019).

2.2. Experimental procedure

Twelve models of VFCW systems were prepared for the study. CW models were fed with leachate from two municipal landfills (Jawor and Legnica), in concentrations of 25% (diluted with tap water) and 100%. Three types of fillings have been prepared for each variant (zeolite, LECA or pine bark). Each experimental system prepared consisted of two PVC containers with a capacity of 30 l. In the upper container, the substrate consisted of coarse gravel, fine gravel, a replacement layer depending on the variant (zeolite, LECA or pine bark) and sand (Fig. 1). In the lower tanks leachate was placed, which was dosed by means of pumps with a capacity of 100 l/day, onto the bed for 24 h/day for the entire duration of the experiment.

The upper tank was retrofitted with an outlet pipe mounted at the base through which the solution returned to the lower tank to achieve a continuous recirculation system (Fig. 1). The VFCW was dosed with leachate from two landfills (Legnica and Jawor) at 100% and 25% concentration (diluted with tap water). In each system 5 *Phragmites australis* seedlings were placed. *P. australis* seedlings with a root surface of approximately 3.5 cm were used in the study.

For plant acclimatisation, biofilm development, and additional stabilisation of the wetland floor, the leachate was applied for 38 days [36,37] while the actual experiment lasted 21 days [38].

The experiment was conducted from spring to autumn. During the conduct of the experiment, the average temperature was maintained at $20.7\,^{\circ}$ C, the minimum temperature was $12.3\,^{\circ}$ C and the maximum temperature was $31.3\,^{\circ}$ C. The publications show that the temperature in the range from $0\,^{\circ}$ C to $35\,^{\circ}$ C does not have a significant effect on the effectiveness of the removal of contaminants from the leachate in CW [33,39,40].

2.3. The scope of physicochemical analyses

1) Analysis of the selected components content in plant material

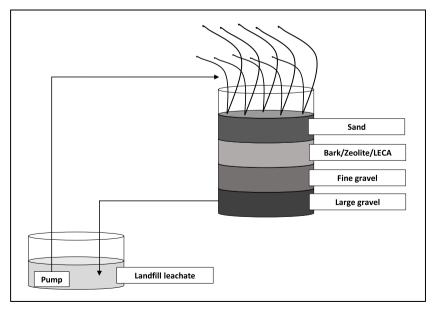


Fig. 1. Diagram of sample CW.

At the end of the experiment (i.e. after 59 days), the plants were removed from the CW and separated into an above-ground part (stem, leaves) and an underground part (roots).

The collected plants were dried at room temperature. The dried biomass was ground in a laboratory grinder and subjected to chemical analyses. Determination of selected components in plant material from all prepared models was performed after wet digestion, in a mixture of concentrated perchloric, sulfuric and nitric acid (ratio 10 HNO₃:1H₂SO₄:4 HClO₄) [41]. All analyses were performed in triplicate manner.

The samples were analysed for total phosphorus (TP), total Kjeldahl nitrogen (TKN) and manganese (Mn), nickel (Ni), potassium (K), iron (Fe), copper (Cu), chromium (Cr), lead (Pb) and cadmium (Cd). The limit of detection for Cd is 0.0001 mg/L, Cr is 0.0001 mg/L, Cu is 0.0001 mg/L, Ni is 0.0001 mg/L, Mn is 0.0001 mg/L, K is 0.001 mg/L and Fe 0.0001 mg/L. To determine the content of heavy metals was used Atomic Absorption Spectrometry (AAS), accordance standards - ISO 15586:2003.

2) BCF bioconcentration and TF translocation factors

Bioconcentration factors (BCF) and translocation factors (TF) were calculated to determine the ability of a plant to accumulate elements from the substrate or external solution. BCF is the ratio of the concentration of contaminants in the above/below ground organs of a plant (mg kg $^{-1}$) to the concentration in the external solution (mg L $^{-1}$). The BCF was calculated as follows [42,43]:

$$BCF = \frac{C_p}{C_w} \tag{1}$$

where.

 C_p – pollutant concentration in the plant (mg/kg),

C_w – concentration in the external solution (mg/dm³).

A higher BCF value indicates better phytoaccumulation potential.

TF, on the other hand, indicates the ability to transfer contaminants from the underground to the aboveground parts of the plant. It is calculated as the ratio of the concentration of contaminants accumulated in the aboveground parts of the plant to its underground parts:

$$TF = C_a/C_u \tag{2}$$

where.

C_a – pollutant concentration in aboveground tissues (mg/kg, mg/g),

C₁₁ - pollutant concentration in underground tissues (mg/kg, mg/g).

BCF and TF values above 1 characterise good bioaccumulators and indicate that the plant has the ability to translocate contaminants [16,44].

2.4. Substrates used in the CW

The choice of a substrate in the systems (Fig. 1) was dictated by the widespread availability of materials in Poland and worldwide. Additionally, the cost of the materials and lack of need for pre-treatment were taken into account. Depending on the variant, the replacement layer consisted of a mineral substrate zeolite (further in the article marked with the letter Z) or expanded clay: Lightweight Expanded Clay Aggregate – LECA (further in the article marked with the letter L) or an organic substrate - pine bark (further in the article marked with the letter B).

Bark 150–200 mm thick was used, the C:N ratio was 104.93 (C = 51.326%, N = 0.489%). However, the content of other components was as follows: potassium 1305.5 mg/kg, magnesium 635 mg/kg and phosphorus 756 mg/kg. Medium-sized bark, between 20 and 60 mm, was used as infill.

The total exchange capacity with respect to ammonium ion of the zeolite used according to the manufacturer's information is 0.7 val/l. The particle size of the zeolite used as the CW filling is 2.5–5 mm. The particle size of the expanded clay used as the CW filling is 8–16 mm.

In all CW systems, a layer of fine and coarse gravel and sand was used in addition to the replacement layer, each layer being 50–100 mm thick.

2.5. Data treatment and statistical analysis

The results of the analyses were analysed using Statistica 13.1 software (StatSoft Polska, StatSoft, Inc., Tulsa, OK, USA). Analysis of variance was used to evaluate differences between the experimental variants.

3. Results and discussion

The physicochemical composition of leachates from two landfills after treatment in CW systems, at concentrations of 25% and 100% was analysed. The 3 types of fillings used (Z - zeolite, B - barge, L - LECA) and the maximum permissible values allowing their discharge to water or land imposed by Polish regulations (Regulation of ME of 12 July 2019) were taken into account. The post-treatment leachate, irrespective of the concentration used (i.e. 25% and 100%) and the CW filling variant (bark, zeolite, expanded clay), did not meet the legal requirements for discharge to water or to the ground. A detailed description of the results of the physicochemical properties of the leachate before and after treatment in CW was presented in Ref. [33].

3.1. Translocation of selected contaminants by P.australis

TF represents the translocation of elements from underground to above-ground parts of plants because it is the ratio of metal concentrations in the upper parts of plants to metal concentrations in the roots [45]. Table 2 presents the translocation of selected contaminants from CW with different substrates (bark, zeolite, LECA) into which leachates from two landfills (in Jawor and Legnica) were dosed at 25 and 100% concentration.

Translocation within plant tissues is dependent on plant species, element and a range of environmental conditions such as pH, reduction potential, temperature or salinity. Also, the parameters of the substrate used, such as particle size, organic matter content, nutrients and the presence of other ions, can affect the uptake and translocation of contaminants [46].

When analysing the mobility of elements from roots to stems, it was observed that the translocation factor (TF) varied between the types of substrate in the CW and the landfills from which the leachate originated [47]. suggested that a substrate may affect metal translocation. For 100% LL in total for all parameters analysed, the highest TF was obtained in the CW with bark substrate and the lowest with zeolite substrate, which could be due to the properties of the bark (Section 2.3), i.e. high organic matter content, nutrients and the presence of other ions, which could also influence the high TF [46]. Analysis of variance showed statistically significant differences between TF values for the zeolite - bark, bark - LECA substrate pairs (Fig. 2). On the other hand, for 25% LL in total, for all parameters, the highest TF was observed in CW with zeolite and the lowest with bark substrate. The differences between the values for the analysed substrates were not statistically significant.

Analysing the translocation of the selected parameters together (without differentiating by the applied substrates, origin and doses of dosed leachates), it can be observed that the lowest translocation was shown by Fe and Cu, and the highest by K and Ni, which confirms that TF varies depending on the type of contamination [48].

The low Fe translocation factor values indicate that *P. australis* accumulates Fe mainly in underground tissues and inhibits its entry into important cellular organs such as mitochondria and cell nuclei, which is consistent with the results obtained by other researchers who also observed low TF values for Fe [49,50].

Low translocation rates were also observed for Cu, confirming that Cu accumulates mainly in plant roots and shows low mobility from roots to aboveground organs. This was also observed by Stoltz et al. and Bonnano et al. [51,52] indicating that underground organs are the main areas of accumulation of metals, including Cu.

The highest translocation rate was recorded for K, which has high mobility, making it not readily available to plants [53]. Also for Ni, high TF values were obtained, confirming that Ni is highly mobile and is therefore highly transported both from the substrate to the roots and to other plant tissues [54].

Higher TF values were mostly obtained in CWs dosed with 100% LL than with 25% LL, but these were not large differences. The exceptions were three parameters, i.e. TKN, K and Ni, where higher TF was recorded in CW with 25% LL. In most cases the differences

Table 2
Translocation factor (TF) in *Phragmites australis* tissue in CW (25% LL and 100%LL) with different substrates (Z-zeolite, B-bark and L-LECA).

| Parameter | Landfill | Translocation factor | | | | | | | |
|-----------|----------|----------------------|---------------|------|--------------------------|------|------|--|--|
| | | 25% concen | tration of LL | | 100% concentration of LL | | | | |
| | | Z | В | L | Z | В | L | | |
| TKN | Legnica | 1.48 | 1.91 | 1.94 | 1.34 | 1.97 | 1.75 | | |
| | Jawor | 1.56 | 1.34 | 1.64 | 0.14 | 0.85 | 1.29 | | |
| TP | Legnica | 1.18 | 1.23 | 0.94 | 0.59 | 2.21 | 1.97 | | |
| | Jawor | 3.40 | 0.95 | 1.12 | 1.12 | 0.67 | 2.71 | | |
| K | Legnica | 0.90 | 1.49 | 4.08 | 2.35 | 2.30 | 2.92 | | |
| | Jawor | 2.07 | 1.68 | 2.05 | 1.01 | 0.55 | 2.48 | | |
| Fe | Legnica | 0.35 | 0.21 | 0.06 | 0.23 | 0.28 | 0.08 | | |
| | Jawor | 0.24 | 0.16 | 1.15 | 0.07 | 6.06 | 0.64 | | |
| Mn | Legnica | 1.22 | 1.55 | 0.55 | 1.41 | 0.69 | 1.18 | | |
| | Jawor | 1.72 | 1.15 | 0.93 | 2.31 | 3.39 | 0.89 | | |
| Cu | Legnica | 0.78 | 1.09 | 0.74 | 1.15 | 0.87 | 0.97 | | |
| | Jawor | 0.91 | 1.01 | 0.87 | 1.11 | 1.44 | 0.81 | | |
| Cr | Legnica | 2.89 | 1.04 | 0.28 | 0.63 | 1.77 | 0.44 | | |
| | Jawor | 0.70 | 1.22 | 0.76 | 0.74 | 2.61 | 0.85 | | |
| Ni | Legnica | 1.84 | 0.77 | 0.27 | 0.14 | 0.03 | 0.22 | | |
| | Jawor | 3.57 | 2.03 | 3.89 | 0.71 | 3.58 | 1.07 | | |

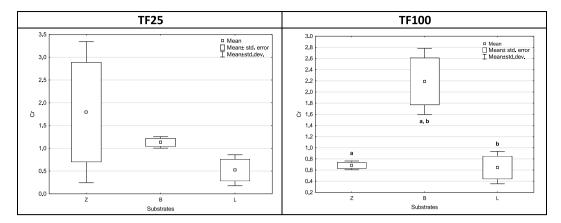


Fig. 2. Comparison of characteristic values (mean, mean \pm standard error, mean \pm standard deviation) of TF chromium translocation factor at 25% and 100% leachate concentration, divided into substrates (Z - zeolite, B - bark, L - LECA). Pairs of letters: a-a, b-b marked the occurrence of statistically significant differences (marked differences are significant with p < 0.05).

in TF values were not large and ranged from 0.7 to 0.9.

Root/stem translocation in *P.australis* for most parameters (TKN, TP, K and Mn), in both CW with 25%LL and 100% LL, was high, i. e. TF > 1.0. Mn showed high mobility and in most cases TF was >1.0, with the exception of 25% LL (Legnica, Jawor) and 100% LL (Jawor, LECA and Legnica, bark), where TF was <1.0. The high values of manganese translocation factor can be explained by the fact that it plays an important metabolic role [55]. In contrast, root/stem translocation for: Fe, Cu, Cr and Ni, in both CW with 25%LL and 100% LL, was mostly low and <1.0.

3.2. Bioaccumulation of selected pollutants by P.australis

Table 3 presents the bioconcentration of selected pollutants in CW with different substrates (bark, zeolite, LECA) into which leachates from two landfills (in Jawor and Legnica) were dosed at 25% and 100% concentration.

The bioconcentration factor (BCF) measures the ability of a plant to accumulate elements from the substrate [56]. Some researchers consider plants with a BCF of >1.0 to be a good accumulator [44]. However, others consider that a good metal accumulator should have a much higher BCF, i.e. >1000 [57,58].

For most parameters (TKN, TP, K, Fe, Mn) in all variants, irrespective of the origin and selected concentration of leachate and the applied substrate (zeolite, bark and LECA), BCF values were >1.0. Only in a few cases BCF values above 1000 were recorded, i.e. for Fe and Mn parameters, in 100% LL. Analysing the total BCF values obtained (without differentiating between substrate types and landfills), it was observed that BCF values were significantly higher in underground tissues than in aboveground tissues and in 100% LL than in 25% LL.

The higher BCF values in underground tissues may be due to a defence mechanism of the plant. Thanks to the strategy of preventing the movement of toxic substances from the roots to the aboveground organs, the plant protects itself from their harmful effects, among others on photosynthesis processes [59]. In most cases, the highest BCFs in total (for both landfills and all parameters) were recorded on bark substrates, which may be due to its properties (Section 2.3). As it is known, the high content of organic matter and nutrients can affect the uptake and transport of pollutants [46]. Only for underground tissues at 25% concentration the highest BCF was achieved on LECA substrate, which was due to the high accumulation of Fe on this substrate (BCF for Fe - 381.46).

It can be observed from this study that nitrogen accumulates mainly in aboveground tissues. Irrespective of the applied LL dilution, higher total BCF values for TKN were recorded in aboveground tissues in 100% LL (i.e. BCF total 410.42 - aboveground tissues and 318.4 - underground tissues) than in 25% LL (where BCF total 39.29 - aboveground tissues and 23.89 - underground tissues). Higher nitrogen accumulation in aboveground tissues than in underground tissues was also noted by others in their studies [60]. Comparing the substrates used, in the CW to which 100% LL was dosed, the highest total BCF was recorded for TKN on bark substrates, which could be due to the influence of the high organic matter and nutrient content of the substrate used [46]. The differences found were not statistically significant (Fig. 3). On the other hand, in CW where 25% LL was dosed, the highest total BCF occurred on zeolite substrates. Analysis of variance showed that the BCF values for zeolite substrates for the aboveground parts of the plants were significantly different from the values found for the underground parts and bark and LECA substrates (Fig. 3).

Phosphorus, like nitrogen, accumulates mainly in aboveground tissues. Regardless of the LL dilution used, higher total BCF for TP was recorded in aboveground tissues in 100% LL (i.e. BCF total 229.82 - aboveground tissues and 171.55 - belowground tissues) than in 25% LL (where BCF total 29.16 - aboveground tissues and 23.82 - belowground tissues). In both 25% LL and 100% LL, comparing the substrates used, the highest BCF for TP was recorded on the bark substrates, which was most likely due to the presence of phosphorus in the substrate (756 mg/kg). During the study, it was observed that the accumulation capacity of phosphorus in *P. australis* tissues was much lower than that of nitrogen. The total BCF for TKN was 792, while that for TP was 454.3, confirming that macrophytes are better at accumulating nitrogen. This was also observed during nitrogen and phosphorus accumulation studies in CW with horizontal

Table 3
Bioconcentration (BCF) in *Phragmites australis* tissue in CW (25% LL and 100%LL) with different substrates (Z-zeolite, B-bark and L-LECA).

| Parameter | Landfill | Biocencentration factor | | | | | | | | | | | |
|-----------|----------|-------------------------|--------|-------|-----------------------------|--------|--------|--------------------------|--------------|--------|---------------------|--------|---------|
| | | 25% concentration of LL | | | | | | 100% concentration of LL | | | | | |
| | | Aboveground tissues | | | Underground tissues Abovegr | | | Abovegroui | ound tissues | | Underground tissues | | |
| | | Z | В | L | Z | В | L | Z | В | L | Z | В | L |
| TKN | Legnica | 8.73 | 7.18 | 7.85 | 5.92 | 3.77 | 4.05 | 107.73 | 123.38 | 93.15 | 80.2 | 62.63 | 53.19 |
| | Jawor | 7.43 | 3.25 | 4.85 | 4.77 | 2.43 | 2.95 | 6.83 | 32.48 | 46.85 | 47.72 | 38.27 | 36.39 |
| TP | Legnica | 6.27 | 8.65 | 3.74 | 5.29 | 7.01 | 3.98 | 21.69 | 71.02 | 35.75 | 36.94 | 32.17 | 18.11 |
| | Jawor | 3.96 | 3.35 | 3.19 | 1.16 | 3.53 | 2.85 | 34.64 | 25.55 | 41.17 | 30.81 | 38.33 | 15.19 |
| K | Legnica | 2.86 | 4.57 | 4.43 | 3.18 | 3.07 | 1.09 | 56.19 | 42.33 | 52.89 | 23.95 | 18.39 | 18.1 |
| | Jawor | 3.45 | 2.85 | 3.35 | 1.66 | 1.7 | 1.63 | 36.54 | 15.79 | 44.33 | 36.25 | 28.94 | 17.89 |
| Fe | Legnica | 32.27 | 19.64 | 23.87 | 91.44 | 94.87 | 381.46 | 304.62 | 188.57 | 133.63 | 1308.1 | 670.66 | 1734.84 |
| | Jawor | 2.02 | 3.88 | 4.29 | 8.36 | 23.6 | 3.74 | 23.04 | 232.37 | 111.87 | 324.42 | 38.36 | 174.68 |
| Mn | Legnica | 61.85 | 179.23 | 36.31 | 50.62 | 115.46 | 66.15 | 1330 | 1559.2 | 664.62 | 943.08 | 2270 | 561.54 |
| | Jawor | 9.71 | 31.85 | 6.77 | 5.64 | 27.8 | 7.32 | 207.6 | 376.27 | 121.73 | 89.87 | 111.07 | 136.53 |
| Cu | Legnica | 0.07 | 0.04 | 0.04 | 0.08 | 0.04 | 0.05 | 0.68 | 0.5 | 0.6 | 0.59 | 0.57 | 0.61 |
| | Jawor | 0.15 | 0.15 | 0.11 | 0.16 | 0.15 | 0.13 | 1.22 | 1.18 | 1.27 | 1.09 | 0.82 | 1.57 |
| Cr | Legnica | 0.07 | 0.02 | 0.03 | 0.02 | 0.02 | 0.1 | 0.24 | 0.15 | 0.23 | 0.38 | 0.09 | 0.53 |
| | Jawor | 0.01 | 0.04 | 0.03 | 0.02 | 0.04 | 0.03 | 0.3 | 0.51 | 0.45 | 0.41 | 0.19 | 0.53 |
| Ni | Legnica | 0.02 | 0.01 | 0.01 | 0.01 | 0.01 | 0.02 | 0.01 | 0 | 0.03 | 0.05 | 0.03 | 0.15 |
| | Jawor | 0.01 | 0.02 | 0.01 | 0 | 0.01 | 0 | 0.05 | 0.12 | 0.08 | 0.08 | 0.03 | 0.08 |

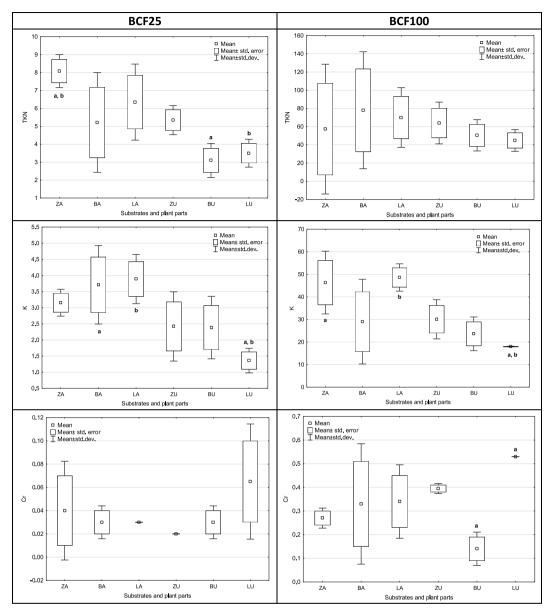


Fig. 3. Comparison of characteristic values (mean, mean \pm standard error, mean \pm standard deviation) of the BCF bioconcentration factor for TKN, K and Cr at 25% and 100% leachate concentration, divided into substrates and plant parts (A - aboveground parts, U - underground parts, Z - zeolite, B - bark, L - LECA). Pairs of letters: a-a, b-b marked the occurrence of statistically significant differences (marked differences are significant with p < 0.05).

subsurface flow [60].

Potassium (K) is an essential nutrient for plant growth, right after nitrogen (N) and phosphorus (P). Due to the fact that it moves easily in the environment, it is not readily available for uptake by plants, leading to economic losses [53]. The bioaccumulation factor of K varied quite strongly between the dilutions and substrates used. Significantly higher BCF values for K were obtained in 100% LL than in 25% LL and in underground tissues than in aboveground tissues. The higher BCF values in underground tissues may be due to the fact that K is mainly taken up by the roots. In CW, where 100% LL was dosed, quite large differences were observed between the applied fills. The highest BCF value was achieved on zeolite substrates, followed by LECA and bark. Statistically significant differences in BCF values for K occurred for the aboveground parts of plants growing on zeolite and LECA media versus the underground parts of plants on LECA media (Fig. 3). The high BCF for K in the case of zeolite substrates can be explained by the fact that zeolites are very efficient ion exchangers and perform well in retaining K [53].

BCF of Fe in underground tissues was significantly higher than BCF in aboveground tissues regardless of the substrate used, origin and LL concentration. Also other researchers [49] have observed a significantly higher accumulation of Fe in roots than in

aboveground tissues. By concentrating Fe in the roots, the plant increases its tolerance to unfavourable environmental conditions [49]. High bioaccumulation rates were recorded for Mn, which may be due to the fact that *P. australis* is very tolerant to high concentrations of Mn and can accumulate it in large quantities in its tissues [61].

Irrespective of LL concentration, higher Mn contents were recorded in aboveground tissues compared to underground tissues. In contrast, others during the study observed higher Mn contents in roots [61].

In several studies [47,62] have shown that P.australis is a good accumulator of heavy metals. However, in this study it was observed that regardless of the selected leachate concentration or the type of substrate, for most metals (Cu, Cr and Ni) the BCF was low (i.e. BCF<1.0). Only in the case of Cu in 100% LL from Jawor, the BCF was slightly above 1.0. The results obtained show that this species does not have a good ability to phytoaccumulate heavy metals (Cu, Cr and Ni) from landfill leachate in CW. This coincides with the results of other authors who also found that P.australis does not perform well as a metal accumulator [47,50]. In addition, the results obtained confirm that plant uptake plays a minor role in the removal of heavy metals from leachate in CW [63]. Statistically significant differences between the BCF values occurred for Cr, related to the underground parts of the plants and the bark and LECA substrate (Fig. 3).

4. Conclusions

After treatment of leachate in CW, there were differences in many cases that made it difficult to clearly assess the effectiveness of the reduction of selected parameters on the substrates used.

- 1. Translocation of selected elements in P.australis tissues differed between the parameters analysed. In most cases, higher TF values were recorded in CW dosed with 100% LL. The lowest translocation was recorded for Fe and Cu, and the highest for K and Ni. Translocation between root and shoots for most parameters (TKN, TP, K and Mn) was high (TF > 1.0). However, for Fe, Cu, Cr and Ni translocation was low in some variants (TF < 1.0). Only for Cr there were statistically significant differences between the substrates.
- 2. In all cases, higher bioaccumulation was observed in underground tissues than in aboveground tissues. In most cases, higher BCF values were recorded at 100% LL than at 25% LL. The substrate of pine bark had the greatest effect on the accumulation of selected contaminants in *P. australis*, and in most cases the highest total BCF values were recorded there. The BCF values found only in a few cases (TKN, K and Cr) showed significant differences between the tested fills and plant parts.
- 3. The obtained BCF values for Cu, Cr and Ni show that *P. australis* does not have a good ability to phytoaccumulate heavy metals from landfill leachate.

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Author contributions

Conceptualization, A.W. and A. S.-P.; methodology, A.W. and A. S.-P.; software, A. S.-P.; validation, A. S.-P.; formal analysis, A.W. and A. S.-P; investigation, A.W.; resources, A.W. and A.S.-P.; data curation, A.W.; writing—original draft preparation, A.W.; writing—review and editing, A.S.-P..; visualization, A.W. and A. S.-P; supervision, A. S.-P; funding acquisition, A.W.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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