



Phytoremediation of metal-contaminated bottom sediments by the common ice plant (*Mesembryanthemum crystallinum* L.) in Poland

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Abstract

Purpose The aim of the study was to propose a phytoremediation-based approach toward the proper utilization of post-industrial, metal-contaminated bottom sediments. The common ice plant, *Mesembryanthemum crystallinum* L. (Aizoaceae), an abiotic-stress tolerant, C3/CAM intermediate halophyte, was tested for growth in substrates containing bottom sediments and for biological removal of metal pollutants. In variant tests, the sediments were admixed with non-toxic components to reduce the ecotoxicity hazards and improve growth conditions.

Materials and methods Bottom sediment samples were collected from Lake Chechło in the industrial area of Poland. They were amended with universal soil and other materials (sand, lime, plant ash) and then used as growth substrates. After 30-day growth the plant biomass and rhizospheric microbiota population were assessed. The elemental content was determined in the substrate as well as in plant organs with inductively coupled plasma–optical emission spectrometry (ICP-OES). Bioaccumulation factors (BAFs, indicating phytoextraction processes) and root-to-shoot translocation factors (TFs) were calculated for all the metals to trace their behavior upon phytoremediation. Ecotoxicity assessments were performed by using a set of biotests (Phytotoxkit, Ostracodtoxkit F, and Microtox).

Results *M. crystallinum* proved its ability to grow under harsh conditions of toxic and poor-quality substrates, while allowing for proliferation of rhizosphere bacteria. The plant growth was accompanied by the accumulation of Na and several other metals which were partially removed from the bottom sediment-containing soils. Depending on the experimental variant, the maximum removal achieved upon the 30-day test was: for Cd, 18.1%, Cu, 47.6%, Cr, 32.7%, Pb, 36.6%, and Zn, 24.1%. *M. crystallinum* hyperaccumulated Zn and accumulated (either in roots or shoots) Cd, Cu, Cr, and Ni. The maximum BAF values (> 1.0) were obtained for the following metals: Cd, Cr, Ni, Cu (roots) and Cd, Cr, Ni, Zn (shoots). The highest values of TF (> 1), confirming high phytoremediation potential, were calculated for Na (33.33), Cd (1.47), Cu (1.77), Cr (7.85), and Zn (4.02). Bottom sediments revealed class III toxicity (acute), which was decreased by admixing with other materials. Surprisingly, the treatment with *M. crystallinum* led to an increase of toxicity levels, possibly by mobilizing potentially toxic elements during plant growth and microbial population development. However, mixing the sediments with universal soil and lime enabled us to maintain class I (no acute toxicity).

Conclusion The common ice plant reveals strong application potential for use in reclamation of soils or revitalization of industrially degraded areas containing bottom sediments.

Keywords Remediation of sediments · Soil reclamation · Anthropogenic pollutants · Potentially toxic elements · Ecotoxicity assessment

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1 Introduction

Bottom sediments, especially those found in reservoirs of industrial areas, are considered ecologically important parts of the aquatic environment. They perform many important ecological or geochemical functions, such as providing

organisms with sources of carbon and minerals, participating in elemental cycles or acting as geosorbents for pollutants (Förstner and Salomons 2010; Baran et al. 2011; Herschy 2012a). They also contain suspended materials from rivers, various precipitates, anthropogenic contaminants, as well as products of other biological activities. In many cases, bottom sediments are final targets for depositions of a variety of substances including toxic pollutants. The anthropogenic pollution sources may lead to discharge of pesticides, metals, and other toxic elements to surface waters, which then cause ecological problems in lake systems. The environmental risk of numerous recalcitrant contaminants occurring in the aquatic environment, especially metals, is strongly affected by humic substances present in the sediments. These substances may act as complexing agents influencing the element mobilities and thus their bioavailabilities and toxicities (Chen et al. 2004; Herschy 2012a; Ali et al. 2013; Mwamburi 2018; Śliwa-Cebula et al. 2020).

In order to reduce the hazards caused by the presence of toxic compounds in aquatic systems, the contaminated bottom sediments should be properly managed or utilized. Usually sediments are removed by dredging (Herschy 2012b; Liu et al. 2015; Chen et al. 2019). The removed material can be classified into the following categories: (1) material suitable for direct reuse in environmental applications, (2) limited use material that can be applied after appropriate processing or (3) waste material to be landfilled. Among the most serious risks associated with bottom sediment management are physico-chemical processes that lead to the release of metals into the environment. A particular caution is that the mobility of elements occurring in sediments is highly changeable and depends on a number of conditions (Salomons et al. 1987; Baran et al. 2015; Fathollahzadeh et al. 2015).

Recently, bottom sediments have attracted considerable interest as potential components of growth substrates used in agricultural practice. These sediments can also serve as suitable materials for reclamation and revitalization of anthropogenically degraded areas. Therefore, establishing the metal content, concentration of xenobiotics, and other hazardous elements, as well as assessing ecotoxicities, is highly important. Depending on contamination levels, remediation actions are required to reduce the environmental threat. Phytotechnologies, especially phytoremediation methods, appear as proper and practically suitable solutions. Phytoremediation, as an alternative to chemical and physical methods, is a cheap, non-invasive, and an efficient industrial-scale approach aimed at environmental recovery and reclamation of soils. It is based on the use of green plants for pollutant phytoextraction, phytostabilization, rhizoremediation, or biotransformation (Ali et al. 2013; Favas et al. 2014; Ansari et al. 2018; Śliwa-Cebula et al. 2020; Yan et al. 2020). For metal-contaminated soils, the

plants should be tolerant both to non-essential elements (Pb, Cd, As, Cr, and Hg) and to high concentrations of the bioavailable forms of essential metals (Cu, Fe, Mn, Ni, Zn). Among the phytoremediators, metal accumulators and hyperaccumulators are particularly useful. These plants are capable of growth in toxic environments and able to take up metals with uptake followed by efficient translocation from roots to shoots (Van der Ent et al. 2013; Śliwa-Cebula et al. 2020; Yan et al. 2020).

The purpose of this work was to propose a phytoremediation strategy for utilization of bottom sediments contaminated with toxic or potentially toxic metals. We have tested the applicability of the common ice plant, *Mesembryanthemum crystallinum* L. of the Aizoaceae family. The applied model plant is a low-demanding, fast-growing semi-halophyte, producing an extensive root system. As shown earlier, this plant reveals high biotechnological potential by being capable of adapting to extreme conditions while developing resistance mechanisms against numerous environmental stresses including drought, salinity, high temperature, poor-quality substrates, oxidative stress, and high light intensity. Most of the studies carried out thus far have focused on the high plasticity of *M. crystallinum* photosynthetic metabolism, manifested by the ability to shift between C3 and CAM (Crassulacean Acid Metabolism) types in response to high salinity (Adams et al. 1998; Kornaś et al. 2010; Surówka et al. 2016; Libik-Konieczny et al. 2019). More recently, the common ice plant in model experiments proved its ability to adapt and grow in the presence of high concentrations of Cd, Ni, Zn, and Cu ions. The described extraordinary properties of *M. crystallinum* suggest the plant's environmental usability for efficient NaCl accumulation; Cr, Ni, and Cu phytoremediation; and Cd phytostabilization (Kuznetsov et al. 2000; Shevyakova et al. 2003; Kholodova et al. 2005; Amari et al. 2014; Nosek et al. 2019; Śliwa-Cebula et al. 2020).

The present study is a continuation of earlier research aimed at demonstrating agronomic applicability of bottom sediments originating from the Chechło reservoir (Koniarczyk et al. 2022a) as well as developing methods for metal contamination remediation (Koniarczyk et al. 2022b). In the latter case, an efficient process was proposed for chemical immobilization of metals. Here, we have focused on testing a bio-based approach that is phytostabilization of bottom sediments accompanied by metal phytoextraction. We hypothesized that the common ice plant could grow on substrates containing anthropogenically affected bottom sediments and develop populous rhizospheric microbiota. We also tended to verify whether the applied plant model might be used for efficient sediment phytoremediation by accumulating toxic elements and thus leading to the reduction of substrate ecotoxicity.

2 Materials and methods

2.1 Bottom sediments

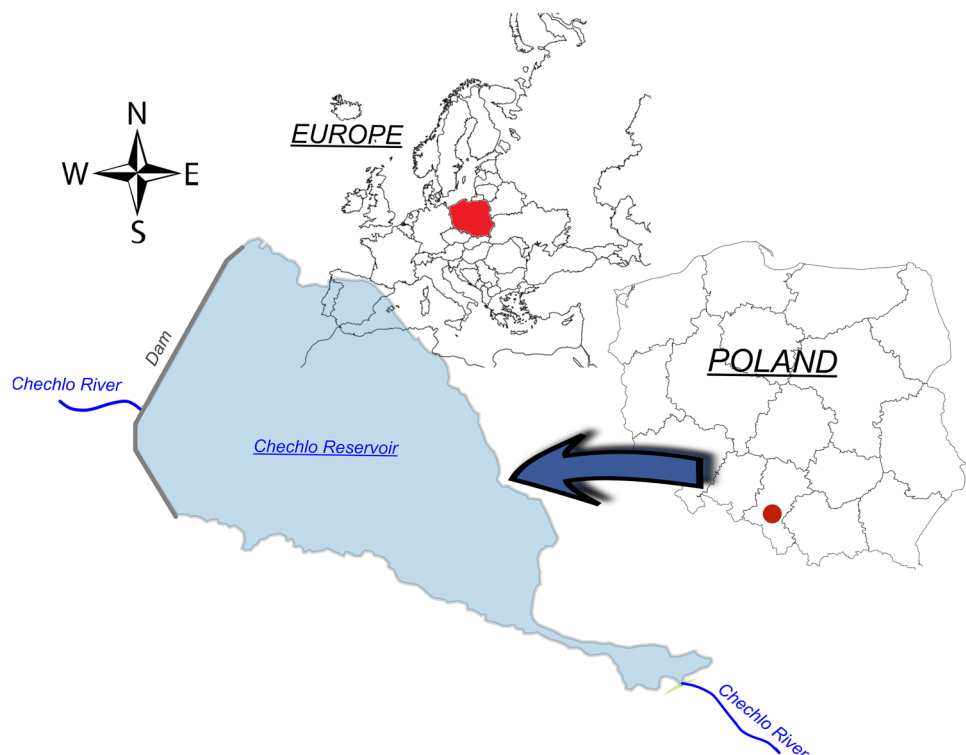
The bottom sediments were obtained from the Lake Chechło reservoir. This reservoir is located in the north-western part of the province of Małopolska, near Trzebinia and Chrzanów cities (50°8'29"N, 19°30'24"E), in a highly industrialized part of Poland (Fig. 1). The adjacent soils contain high levels of lead, cadmium, and zinc. In the Trzebinia municipal district, numerous post-industrial waste products, mainly post-mining and metallurgical ones, are collected and dumped. The above factors pose risk of migration of metal contaminants. The Chechło reservoir was artificially formed within 1944–1945 to provide water supplies for industry and firefighting. Nowadays, its additional role is to level the runoff of the Chechło river bed and to perform recreational functions. The total capacity of the lake is 587 500 m³ (Bogdał et al. 2014). Currently, the inflow of biogenic substances from the coastal and backwater zones causes rapid vegetation overgrowth. The bottom sediments of Lake Chechło were shown to contain high levels of toxic organic carbon (average of 92 g kg⁻¹ d.w.) and total nitrogen (2.5 g kg⁻¹ d.w.), whereas the chemical analyses revealed high content of several metals, compared to other reservoirs of this type (Zawisza et al. 2014). In this study, we have tested both the raw sediments as well as several substrate admixtures

containing universal soil and other materials added to reduce the original ecotoxicity and to improve plant growth conditions.

2.2 Plant cultivation in substrates containing bottom sediments

The experiment was carried out in a summer season (July/August), in a greenhouse under the temperature ranging typically from 20 °C (morning hours) to 30 °C (early and late afternoons) and occasionally reaching 40 °C. *M. crystallinum* seeds were sown and the sprouts grown for four weeks. Plants with a fully developed second leaf pair were divided into five groups and transferred to new pots with volume 0.5 L (100 × 75 × 75 mm). The first group (U) consisted of control plants grown in the substrate obtained upon mixing of the universal soil substrate “Hartmann” with sand (proportions: 7.5 L substrate:1 kg sand) and then adding 0.25 kg to each pot. This mixture was also used as an additive to bottom sediments in experiments applying mixed substrates. Plants in the second group (S) were grown on raw bottom sediments (1.2 kg per pot). The remaining plant groups were grown on mixtures containing the following components (mixed by volume % due to differences in bulk densities of each material): (SU) bottom sediments (50%) + universal soil substrate with sand (50%); (SLU) bottom sediments (25%) + limed soil (25%) + universal soil substrate with sand (50%); (SPU) bottom sediments (25%) + plant ash (25%) + universal soil

Fig. 1 Location of the Chechło Reservoir, the study area

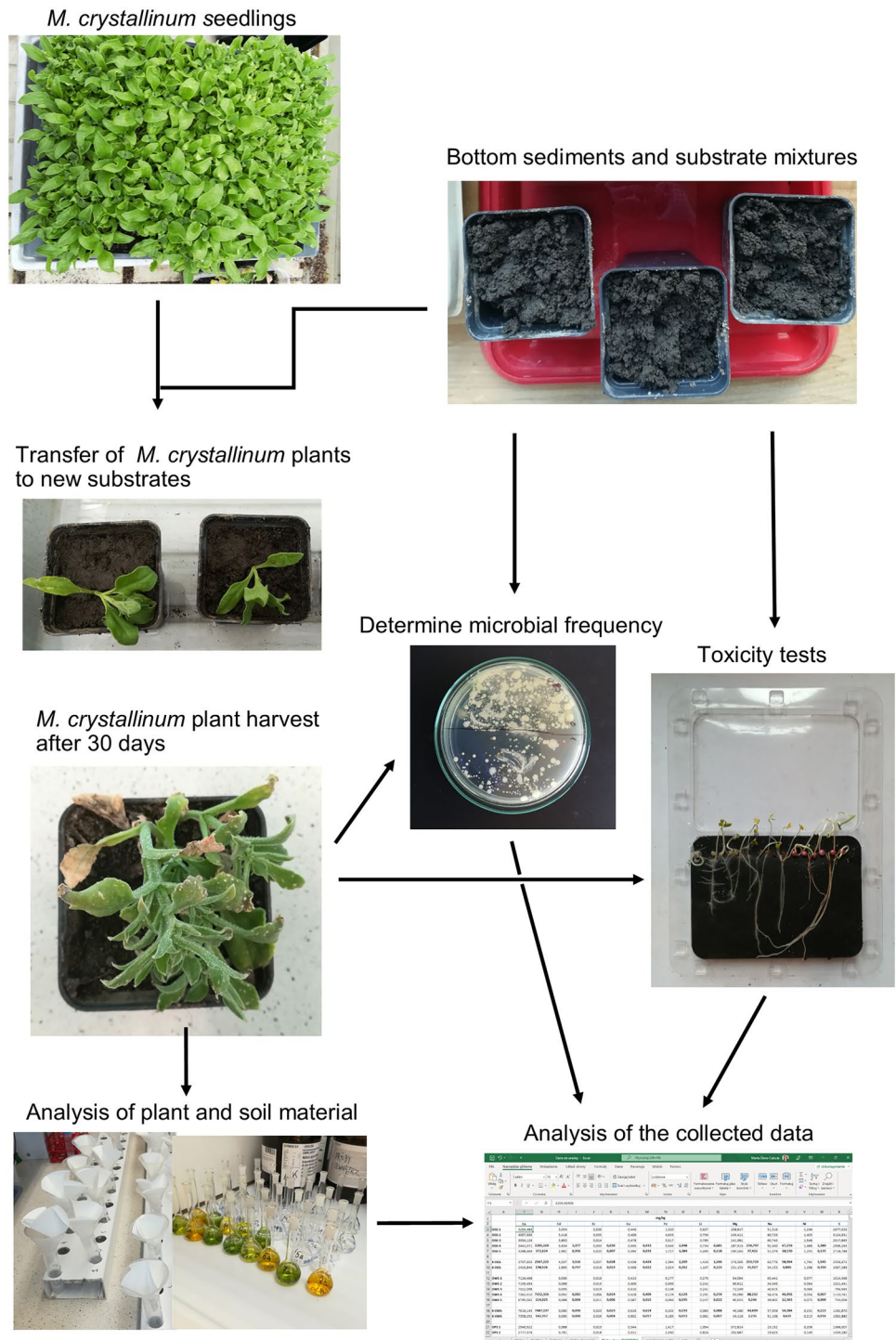


substrate with sand (50%). Typically, for each experimental series, 5 pots were tested (5 replicates, one plant per pot), which required a total of 1.5 kg (25%) or 3.0 kg (50%) of raw bottom sediment, 0.63 kg of soil/sand substrate, and 0.3 kg of the other additives. Figure 2 presents the scheme of the experiment.

The market available universal substrate “Hartmann” (Hartmann, Poznań, Poland) consisted of milled high

peat, fraction 0–20 mm, pH 5.5–6.5, supplemented ($1.0\text{--}1.3\text{ kg m}^{-3}$) with the all-in-one powdered multicomponent fertilizer “PG Mix NPK 14:16:18” (Yara, Szczecin, Poland). The final elemental content of the substrate was determined by ICP-OES (see below). Biologically available macro- and micro- nutrients were extracted from the soil substrate with acetic acid; the concentrations of P, K, Mg, Ca, S, Mn, Al, Ba, Li, Na, Sr were: 29.89, 60.69, 61.71,

Fig. 2 Scheme of the pot experiment, data collection, and analysis



1062.9, 14.93, 0.37, 0.76, 0.31, 0.01, 25.69, 0.68 mg per L, respectively. Nitrogen was present as nitrites (III) and nitrates (V) (a total of 13 mg L⁻¹), and ammonia (53 mg L⁻¹). The substrate acidity (pH of 5.5–5.7) and overall salinity (EC of 1–1.5 mS cm⁻¹) were determined with potentiometric and conductometric techniques, respectively, in a soil:water mixture (20:40 cm³).

The plant tolerance to the tested substrates was established based on determination of physiological and morphological characteristics such as growth, necrosis, and chlorosis. After 30 days, having completed the experiments, the plant material was harvested, and the fresh and dry weights were measured.

2.3 Determination of elemental content in sediments, substrates, and plants

After thorough cleaning, the harvested roots were rinsed with distilled water. The fresh weight of the plants was determined. The plant material was then dried for 3 days at 105 °C, and the dry weight (d.w.) was assessed. The plant tissue samples were ground in liquid nitrogen with a mortar. Next, the material was mineralized in 65% HNO₃ (suprapure, MERCK) with the use of the CEM MARS-5 Xpress mineralizer. The analyses of element concentration in bottom sediments and mixed substrate samples were done according to the method of Rinkis, based on the extraction of 10 g specimens with 1 mol L⁻¹ HCl (Ostrowska et al. 1991; Gediga et al. 2015). It should be noted here that the applied extraction protocol is considered a standard method typically used to determine the bioavailable metal fractions (Ostrowska et al. 1991; Stanislawska-Glubiak and Korzeniowska 2010; Baran et al. 2019b; Pusz et al. 2021; Koniarz et al. 2022a). The total content of elements in soil was not assessed since our study focused on accumulation indices calculated for plant bioaccessible metal forms. The extracts were subjected to the inductively coupled plasma–optical emission spectrometry (ICP-OES, Prodigy Teledyne Leeman Labs, Mason, Ohio, USA), based on the appropriate calibration curves with the Certipur® reference standards (the “ICP multielemental standard IV”, Merck, Darmstadt, Germany).

2.4 Definition of metal accumulation capabilities: bioaccumulation, and translocation factors

M. crystallinum bioaccumulation factors (BAFs) and translocation factors (TFs) were calculated for all the elements determined in the tested substrates. BAF was defined as a given metal concentration [mg kg⁻¹ d.w.] in plant organs (roots or shoots) per concentration in the soil substrate [mg kg⁻¹ soil d.w.]. TF was calculated as the ratio of an element accumulated in shoots to that determined in roots [mg kg⁻¹ d.w.].

2.5 Bacteria colonizing the rhizosphere of *M. crystallinum*

To determine microbial frequency within the root zone of the *M. crystallinum* plants, the root systems were carefully pulled out and the soil material shaken off and collected. Bacterial cell population was monitored in aqueous soil extracts using a standard method of Koch which involves surface-plating onto Petri dishes with the media solidified with 2.5% enriched agar (Biocorp, Poland). Colony-forming units (CFUs) were macroscopically evaluated and counted after 3-day incubation at room temperature, and the resultant cell numbers were expressed as CFUs per g d.w. of the original soil samples (Kaszycki et al. 2014).

2.6 Toxicity tests

The toxicity assessment of the samples was performed employing three biotests: Phytotoxkit, Ostracodtoxkit F, and Microtox. Phytotoxkit is a plant test based on the evaluation of germination and early plant growth of three plant species: *Sinapis alba*, *Lepidium sativum*, and *Sorghum saccharatum*. Ostracodtoxkit F is a direct contact test used to assess chronic toxicity using a crustacean *Heterocypris incongruens* as a test organism. The assessment of mortality and increase in body length of crustaceans were carried out after 6 days of exposure to pollutants contained in the tested samples (Szara et al. 2020). The Microtox test is used to measure acute toxicity and consists in using *Aliivibrio fischeri* luminescent bacteria as test organisms. Upon exposure to a toxic substance, a decrease in luminescence is observed, which is a result of a cellular respiration inhibition. The procedures for the Phytotoxkit, Ostracodtoxkit F, and Microtox tests were described in detail in our previous studies (Baran et al. 2019a, 2020; Szara et al. 2020). The toxicity assessment of samples was carried out on the basis of the estimated percentage value of toxic effect (PE) for the performed biotests. The samples were assigned to the appropriate toxicity classes: class I (PE ≤ 20%, no significant toxic effect)—no acute hazard; class II (20% < PE ≤ 50%, significant toxic effect, low toxic sample)—low acute hazard; class III (50% < PE < 100%, significant toxic effect, toxic sample)—acute hazard; class IV (PE = 100%, single test)—high acute hazard; and class V (PE = 100%, all tests)—very high acute hazard (Persoone et al. 2003; Szara et al. 2020).

2.7 Statistical data analysis

All results were reported as mean values ± standard deviation. The results were statistically evaluated with the

one-way ANOVA module of the Statistica 13.3 software (StatSoft Polska, Kraków, Poland), employing a Duncan's post hoc test at the significance threshold level $p \leq 0.05$.

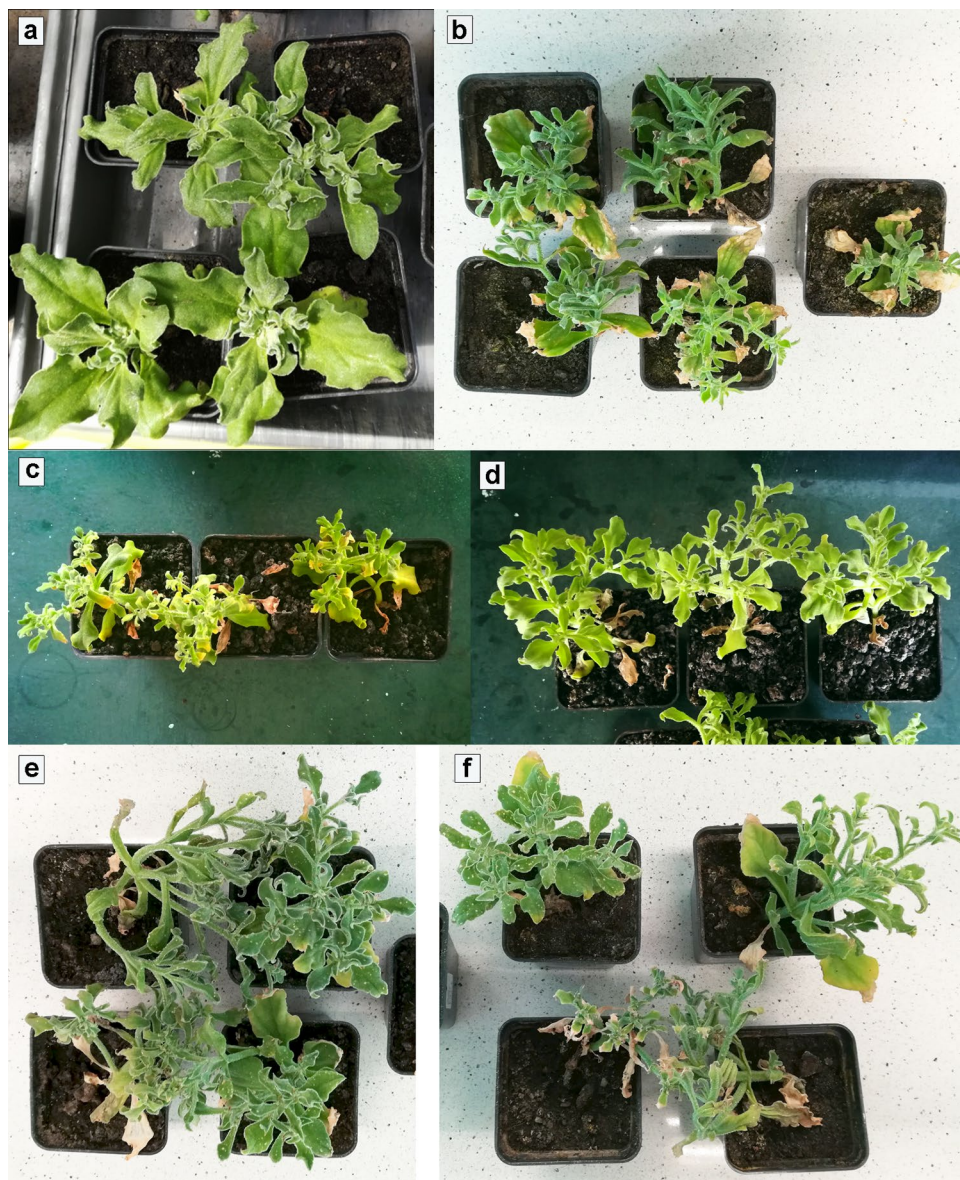
3 Results

3.1 *M. crystallinum* growth on substrates containing bottom sediments

The plant tolerance to the Lake Chechło metal-contaminated bottom sediments and its ability to grow in the tested substrates were determined based on evaluation of the morphological visible symptoms (photos in Fig. 3) and the resultant plant biomass formed after cultivation (Fig. 4).

In Fig. 3a, an example is shown of a typical plant appearance after 10 days of quilting into pots containing either raw bottom sediments, or any of the variant with amending materials. Figure 3b presents the control experiment involving the common ice plant grown for 30 days in universal soil (U). Then, Figs. 3c–f show plant morphologies after 30-day growth in the raw bottom sediment (S), and the variants SU, SLU, and SPU, respectively. Leaving aside the technical differences of independently taken photos, it can be clearly seen that no visible toxicity effects were developed upon cultivation in substrates SU, SLU, and SPU and the plants aerial organs were similar to the control. Only for the case of the sediment alone (S) did the shoots appear relatively weaker; however, no considerable necrosis nor chlorosis could be observed, and in general the plant remained unaffected by the toxic environment (Fig. 3c).

Fig. 3 Morphological evaluation of the common ice plant (*M. crystallinum*) aerial parts appearance after cultivation in substrates containing bottom sediments from Lake Chechło. **a** Typical morphology of plants cultivated for 10 days after quilting as exemplified by the SPU substrate variant; **b** control experiment: plants cultivated for 30 days in universal soil (U); **c–f** plants cultivated for 30 days in substrate variants: S (bottom sediments), SU (bottom sediments + universal soil substrate with sand), SLU (bottom sediments + limed soil + universal soil substrate with sand), SPU (bottom sediments + plant ash + universal soil substrate with sand), respectively



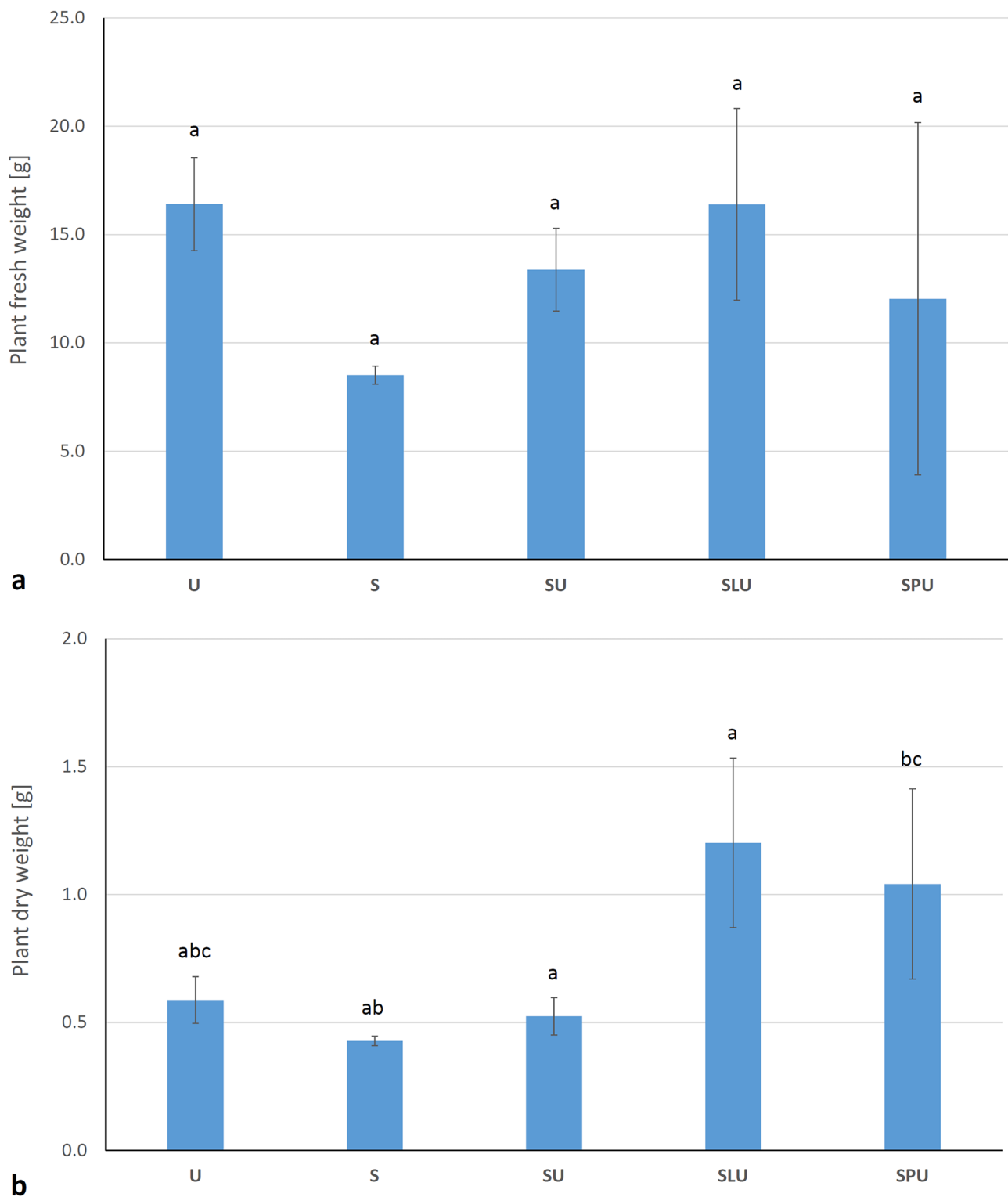


Fig. 4 Fresh **a** and dry **b** weight of *M. crystallinum* plants after 30-day growth in different substrates containing bottom sediments from Lake Chechlo. Bars topped by the same letter do not differ significantly at $p \leq 0.05$; error bars show standard error. Description of substrate samples: U, universal soil substrate with sand; S, bottom

sediments; SU, bottom sediments + universal soil substrate with sand; SLU, bottom sediments + limed soil + universal soil substrate with sand; SPU, bottom sediments + plant ash + universal soil substrate with sand

The direct photographic observations are supported by plant biomass determination after 30-day cultivation on variant substrates (Fig. 4). No significant differences in relation to the control group were found upon assessment of fresh and dry weights (Fig. 4a, b, respectively). Based on the dry matter evaluation, higher biomass was observed only for plants grown on a substrate with the addition of lime (SLU) and plant ash (SPU), but still no statistically significant differences were noted between the growth on the bottom sediment alone (S) and on the universal soil (control, U).

3.2 Analyses of metal content in bottom sediments and substrate formulations

The total content of selected elements in the investigated bottom sediments is given in Table 1, row S1. The levels of several metals (Cd, Cu, Cr, Fe, Na, Ni, Pb, and Zn) were considerably higher compared to the universal soil/sand mixture (Table 1, row U). For the case of Na, its concentrations determined in the tested bottom sediments ($247.1 \pm 13.5 \text{ mg kg}^{-1} \text{ d.w.}$, Table 1, row S1) and in the other substrate mixtures (ranging from 122.9 ± 7.4 to $182.7 \pm 13.6 \text{ mg kg}^{-1} \text{ d.w.}$, Table 1, rows SU1, SLU1, and SPU1) were considerably higher than in typical soils of a moderate climate. Na content usually does not exceed $25 \text{ mg kg}^{-1} \text{ d.w.}$ (Kabata-Pendias 2010) and therefore should be regarded as potentially toxic to some plants. Three other metals, namely Cd, Pb, and Zn occurred at concentrations that should be considered toxic or potentially toxic to plants: their respective levels were determined as: 45.04 ± 0.81 , 244.40 ± 3.63 , and $9243 \pm 207 \text{ mg kg}^{-1} \text{ d.w.}$ (Table 1, row S1). Admixing of bottom sediments with other materials caused a decrease in the content of particular metals in the resultant substrates (Table 1, rows SU1, SLU1, and SPU1) but only in a few cases it allowed for environmental toxicity

risk elimination (see below). The substrates analyzed after 30-day growth of the common ice plant exhibited further decrease of metal concentration (Table 1, rows S2, SU2, SLU2, and SPU2); however, the resultant values suggested the persistence of elevated content of selected contaminating elements. Based on the evaluation of statistical importance, the significant differences of concentrations determined before and after treatment were collated to give evidence for efficient metal removal by *M. crystallinum*. The data of Table 1 are expressed as % of the original metal content removed (phytoextracted) from a given substrate variant: for Cd, variant S, removal of 18.1%; for Cu, 47.6% removed from SLU and 32.9% from SPU; for Cr, 31.9% removed from SLU and 32.7% from SPU; for Pb, 14.5% from S and 36.6% from SPU; and for Zn, 24.1% removed from SU.

3.3 Assessment of metal concentration in plant tissues

The plants grown in contaminated substrates were tested for their capabilities of metal (Na, Cd, Cu, Cr, Fe, Ni, Zn) accumulation. Tables 2 and 3 present the metals content in the roots and shoots, respectively. The obtained values, combined with the undisturbed growth potential, prove the ability of *M. crystallinum* to cope with the presence of metals, even with regard to those elements that occurred above toxicity thresholds.

For the case of sodium, the common ice plant, as expected for a halophyte, tended to accumulate this metal efficiently, both in roots and shoots (Tables 2 and 3). Depending on the tested substrate variant, the content of accumulated Na ranged from 2617 ± 0.182 to $5210 \pm 0.270 \text{ mg kg}^{-1} \text{ d.w.}$ and from $34,669 \pm 2408$ to $104,665 \pm 10,144 \text{ mg kg}^{-1} \text{ d.w.}$ for roots and shoots, respectively. Note that the highest value of $104,665 \text{ mg kg}^{-1} \text{ d.w.}$ was accumulated in the aerial parts

Table 1 The content of metals in different substrates containing bottom sediments from Lake Chechło, before (1) and after (2) growing *Mesembryanthemum crystallinum* plants. Different letters next to the numbers indicate statistically significant differences at $p \leq 0.05$

Substrate sample	mg kg ⁻¹ d.w.							
	Cd	Cu	Cr	Fe	Na	Ni	Pb	Zn
U	7.27 ± 0.47 a	18.62 ± 0.07 ae	0.19 ± 0.18 a	216 ± 19 a	43.2 ± 12.9 a	0.53 ± 0.12 a	0.10 ± 0.04 a	11.8 ± 2.9 a
S 1	45.04 ± 0.81 c	56.55 ± 1.17 b	3.86 ± 0.11 b	13492 ± 367 b	247.1 ± 13.5 d	9.10 ± 0.15 b	244.40 ± 3.63 b	9243 ± 207 b
S 2	36.91 ± 2.50 d	51.29 ± 1.84 b	3.57 ± 0.11 b	17667 ± 474 c	254.8 ± 42.1 d	9.96 ± 0.68 b	208.89 ± 11.19 c	8271 ± 654 b
SU 1	27.13 ± 0.09 b	36.98 ± 1.36 c	2.08 ± 0.29 cde	8348 ± 1114 d	182.7 ± 13.6 cd	6.19 ± 0.27 c	148.71 ± 4.84 d	4624 ± 150 c
SU 2	23.25 ± 3.26 b	35.06 ± 3.43 c	1.49 ± 0.24 ef	8046 ± 703 d	125.6 ± 42.6 abc	5.82 ± 0.66 c	131.62 ± 15.43 d	3509 ± 744 d
SLU 1	7.04 ± 0.12 a	27.17 ± 2.86 d	2.48 ± 0.01 c	3558 ± 217 ef	122.9 ± 7.4 abc	3.34 ± 0.32 de	53.56 ± 2.92 e	1419 ± 42 e
SLU 2	7.40 ± 0.67 a	14.24 ± 1.32 a	1.69 ± 0.17 def	2332 ± 187 f	100.1 ± 19.0 abc	2.52 ± 0.26 e	39.73 ± 3.42 e	844 ± 93 ae
SPU 1	10.20 ± 0.07 a	28.38 ± 2.46 d	2.20 ± 0.37 cd	4170 ± 581 e	142.3 ± 16.8 bc	3.95 ± 0.40 e	84.20 ± 9.39 f	1622 ± 162 e
SPU 2	10.44 ± 0.39 a	19.04 ± 0.81 e	1.48 ± 0.06 f	3364 ± 177 ef	83.2 ± 45.5 ab	3.11 ± 0.13 ef	53.42 ± 1.96 e	1025 ± 49 ae

U universal soil substrate with sand, S bottom sediments, SU bottom sediments + universal soil substrate with sand, SLU bottom sediments + limed soil + universal soil substrate with sand, SPU bottom sediments + plant ash + universal soil substrate with sand

Table 2 Metal content in roots of *Mesembryanthemum crystallinum* plants after 30-day growth in different substrates containing bottom sediments from Lake Chechlo. Different letters next to the numbers indicate statistically significant differences at $p \leq 0.05$

Substrate sample	mg kg ⁻¹ d.w.						
	<i>Cd</i>	<i>Cu</i>	<i>Cr</i>	<i>Fe</i>	<i>Na</i>	<i>Ni</i>	<i>Zn</i>
U	0.43 ± 0.12 a	12.23 ± 0.98 a	2.19 ± 1.35 a	136 ± 9 a	4404 ± 1174 ab	5.72 ± 0.87 a	67 ± 14 a
S	48.08 ± 2.62 c	37.88 ± 5.88 c	26.40 ± 15.68 b	1886 ± 526 b	4264 ± 139 ab	22.19 ± 1.48 b	6668 ± 1286 c
SU	23.97 ± 0.56 b	14.16 ± 1.92 a	4.52 ± 1.48 a	760 ± 119 a	3141 ± 304 ac	5.91 ± 0.85 a	1744 ± 183 b
SLU	0.97 ± 0.03 a	24.49 ± 7.05 ab	0.07 ± 0.03 a	349 ± 57 a	2617 ± 182 c	3.64 ± 0.15 a	174 ± 51 a
SPU	26.82 ± 3.00 b	30.22 ± 4.96 bc	6.25 ± 2.61 a	672 ± 146 a	5210 ± 270 b	22.12 ± 5.87 b	574 ± 90 ab

U universal soil substrate with sand, S bottom sediments, SU bottom sediments + universal soil substrate with sand, SLU bottom sediments + limed soil + universal soil substrate with sand, SPU bottom sediments + plant ash + universal soil substrate with sand

of *M. crystallinum* after growth in bottom sediments mixed with the universal soil (SU variant), where the total Na level was determined as 182.7 ± 13.6 mg kg⁻¹ d.w. (Table 1).

Despite the relatively low contents of Cu, Cr, and Ni in the bottom sediment and the other derived substrates (Table 1), the accumulation of these metals in some experimental variants was also efficient (Tables 2 and 3). The maximum values of the root-accumulated metals (Cu, Cr, and Ni) were obtained in the sole bottom sediment (S): 37.88 ± 5.88 , 26.40 ± 15.68 , and 22.19 ± 1.48 mg kg⁻¹ d.w., respectively. The maximum accumulation levels found in shoots were: 25.02 ± 3.40 mg kg⁻¹ d.w. (in SU), 4.16 ± 1.36 mg kg⁻¹ d.w. (in SU), and 5.62 ± 0.38 mg kg⁻¹ d.w. (in S).

For cadmium, present at potentially toxic concentrations in the tested bottom sediments, the plant tended to accumulate this metal both in roots and shoots in the test variants S, SU, and SPU. The respective levels of Cd were determined in roots (Table 2) as: 48.08 ± 2.62 , 23.97 ± 0.56 , and 26.82 ± 3.00 mg kg⁻¹ d.w., whereas in shoots (Table 3) as: 20.44 ± 3.68 , 35.23 ± 5.43 , and 12.89 ± 2.65 mg kg⁻¹ d.w.

M. crystallinum was particularly capable of accumulating large Zn amounts, either in roots or in aerial parts. Zinc accumulation was most pronounced for the case of treatment of the sole bottom sediment (S), where the Zn content was the highest (9243 ± 207 mg kg⁻¹ d.w., Table 1) and

the level of accumulated metal reached 6668 ± 1286 and $11,052 \pm 2132$ mg kg⁻¹ d.w., for roots and shoots, respectively (Tables 2 and 3).

For lead, it can be seen that the common ice plant was able to accumulate Pb preferentially in the roots (Table 2), but the effect could be observed only for the case of higher concentrations of this element in the substrate. The Pb accumulation values in roots were recorded for S and SU treatments as 41.53 ± 2.92 and 9.85 ± 0.83 mg kg⁻¹ d.w., respectively. For lower concentrations of Pb in the substrates, the applied analytical method made it impossible to assess the content of this element in plant tissues and for that reason the data regarding Pb accumulation have not been listed in Tables 2 and 3 and were not considered further.

In Table 4, bioaccumulation factors (BAF) of the metals are given for roots and shoots to reveal the tendencies of the common ice plant to take up and accumulate these elements. The BAF values varied depending on the experimental variant and on the metal considered. They were particularly high for Na accumulation, where the BAF parameter ranged from 17.26 ± 0.56 to 36.61 ± 1.90 for roots, and from 314.85 ± 16.32 to 572.87 ± 55.52 for shoots (this is discussed later in the context of the plant's halophytic potential). In the case of metals present at high levels (Cd and Zn) BAF > 1 was observed for Cd accumulation, that is root variants Sr

Table 3 Metal content in shoots of *Mesembryanthemum crystallinum* plants after 30-day growth in different substrates containing bottom sediments from Lake Chechlo. Different letters next to the numbers indicate statistically significant differences at $p \leq 0.05$

Substrate sample	mg kg ⁻¹ d.w.						
	<i>Cd</i>	<i>Cu</i>	<i>Cr</i>	<i>Fe</i>	<i>Na</i>	<i>Ni</i>	<i>Zn</i>
U	0.40 ± 0.05 a	9.25 ± 0.26 a	5.76 ± 0.71 a	263 ± 144 ab	49,877 ± 14,348 a	3.26 ± 0.91 a	192 ± 24 a
S	20.44 ± 3.68 b	15.34 ± 2.38 a	2.55 ± 1.52 abc	353 ± 98 a	79,829 ± 2608 c	5.62 ± 0.38 b	11,052 ± 2132 c
SU	35.23 ± 5.43 c	25.02 ± 3.40 b	4.16 ± 1.36 b	262 ± 41 ab	104,665 ± 10,144 d	5.31 ± 0.76 b	7014 ± 737 b
SLU	0.39 ± 0.18 a	14.27 ± 4.11 a	0.52 ± 0.25 c	159 ± 26 b	34,669 ± 2408 ab	2.76 ± 0.11 a	431 ± 127 a
SPU	12.89 ± 2.65 b	16.24 ± 2.67 a	2.53 ± 1.06 bc	215 ± 47 ab	44,807 ± 2322 b	4.10 ± 1.09 ab	536 ± 84 a

U universal soil substrate with sand, S bottom sediments, SU bottom sediments + universal soil substrate with sand, SLU bottom sediments + limed soil + universal soil substrate with sand, SPU bottom sediments + plant ash + universal soil substrate with sand

Table 4 Metal bioaccumulation factors (BAF) of *Mesembryanthemum crystallinum* roots (*r*) and shoots (*s*) after 30-day treatment of substrates containing bottom sediments

Substrate sample	Bioaccumulation factor (BAF)						
	<i>Cd</i>	<i>Cu</i>	<i>Cr</i>	<i>Fe</i>	<i>Na</i>	<i>Ni</i>	<i>Zn</i>
<i>S r</i>	1.07 ± 0.06	0.67 ± 0.10	6.84 ± 4.06	0.14 ± 0.04	17.26 ± 0.56	2.44 ± 0.16	0.72 ± 0.14
<i>S s</i>	0.45 ± 0.08	0.27 ± 0.04	0.66 ± 0.39	0.03 ± 0.01	323.08 ± 10.56	0.62 ± 0.04	1.20 ± 0.23
<i>SU r</i>	0.88 ± 0.02	0.38 ± 0.05	2.17 ± 0.71	0.09 ± 0.01	17.19 ± 1.67	0.95 ± 0.14	0.38 ± 0.04
<i>SU s</i>	1.30 ± 0.20	0.68 ± 0.09	2.00 ± 0.66	0.03 ± 0.00	572.87 ± 55.52	0.86 ± 0.12	1.52 ± 0.16
<i>SLU r</i>	0.14 ± 0.00	0.90 ± 0.26	0.03 ± 0.01	0.10 ± 0.02	21.29 ± 1.48	1.09 ± 0.04	0.12 ± 0.04
<i>SLU s</i>	0.05 ± 0.03	0.53 ± 0.15	0.21 ± 0.10	0.04 ± 0.01	282.05 ± 19.59	0.83 ± 0.03	0.30 ± 0.09
<i>SPU r</i>	2.63 ± 0.29	1.06 ± 0.17	2.84 ± 1.19	0.16 ± 0.04	36.61 ± 1.90	5.60 ± 1.49	0.35 ± 0.06
<i>SPU s</i>	1.26 ± 0.26	0.57 ± 0.09	1.15 ± 0.48	0.05 ± 0.01	314.85 ± 16.32	1.04 ± 0.28	0.33 ± 0.05

UU universal soil substrate with sand, *S* bottom sediments, *SU* bottom sediments + universal soil substrate with sand, *SLU* bottom sediments + limed soil + universal soil substrate with sand, *SPU* bottom sediments + plant ash + universal soil substrate with sand

(BAF = 1.07 ± 0.06) and *SPU r* (2.63 ± 0.29), as well as shoot accumulation *SPU s* (1.26 ± 0.26) and *SU s* (1.30 ± 0.20). For Zn, the shoots tended to accumulate this metal efficiently in *S s* (BAF = 1.20 ± 0.23) and in *SU s* (1.52 ± 0.16).

Consequently, Table 5 shows translocation factors (TF_{root-to-shoot}) to indicate which plant organs (roots or above-ground parts) served as targets for the accumulated metals. As expected for a halophyte phytoextractor, Na was translocated to shoots the most efficiently, yielding TF values ranging from 6.96 (for universal soil U) to 33.33 (for the variant SU). Cadmium was preferentially translocated to shoots only in SU (TF = 1.47), while Zn in variants S, SU, and SLU (TF = 1.66, 4.02, and 2.48, respectively).

3.4 Development of the rhizospheric microbiota

Bacterial cell frequencies determined in the substrates prior to the *M. crystallinum* growth (bottom sediment sample, S(C), and bottom sediment mixed with the universal soil + sand, SU(C)) and after 30-day plant cultivation (universal soil, U, and all the tested variants, S, SU, SLU, SPU) are shown in Fig. 5. As it can be seen, the microbial population inhabiting the substrates before the treatment was approximately three orders of magnitude lower than after plant growth. The

colonization rate at the end of the test was similar to that observed in universal soil (of the order of 10⁷ CFU g⁻¹ d.w.) except for the sole sediment S (10⁶ CFU g⁻¹ d.w.), the most polluted and potentially toxic sample.

3.5 Ecotoxicity of substrata

Ecotoxicological tests were carried out to reveal potential environmental risks borne by the bottom sediment-containing mixtures before and after treatment with *M. crystallinum*. The responses of organisms were dependent on the type of biotest, treatment, and incubation conditions (before and after test plant growth, Table 6). For *S. alba* and *L. sativium*, the inhibition of germination and plant root growth inhibition were between -29 to 33% and -117 to 64%, respectively; mortality and growth inhibition of *H. incongruens* were between 0 to 100% and -46 to 100%, respectively; and *A. fischeri* luminescence inhibition ranged from -62 to 97%. From among all the species tested, *H. incongruens* appeared to be the most sensitive in comparison to other species, while *S. alba* was the least sensitive to substrates containing bottom sediments. Depending on the tested variant, the significant highest toxic responses were observed for organisms in the S and SU treatments. The significant lowest toxicity in the tests organisms

Table 5 Metal translocation factors (root-to-shoot, TF) after 30-day treatment of substrates containing bottom sediments with *Mesembryanthemum crystallinum*

Substrate sample	Translocation factor (TF)						
	<i>Cd</i>	<i>Cu</i>	<i>Cr</i>	<i>Fe</i>	<i>Na</i>	<i>Ni</i>	<i>Zn</i>
S	0.43	0.40	0.10	0.19	18.72	0.25	1.66
SU	1.47	1.77	0.92	0.34	33.33	0.90	4.02
SLU	0.40	0.58	7.85	0.46	13.25	0.76	2.48
SPU	0.48	0.54	0.40	0.32	8.60	0.19	0.93

U universal soil substrate with sand, *S* bottom sediments, *SU* bottom sediments + universal soil substrate with sand, *SLU* bottom sediments + limed soil + universal soil substrate with sand, *SPU* bottom sediments + plant ash + universal soil substrate with sand

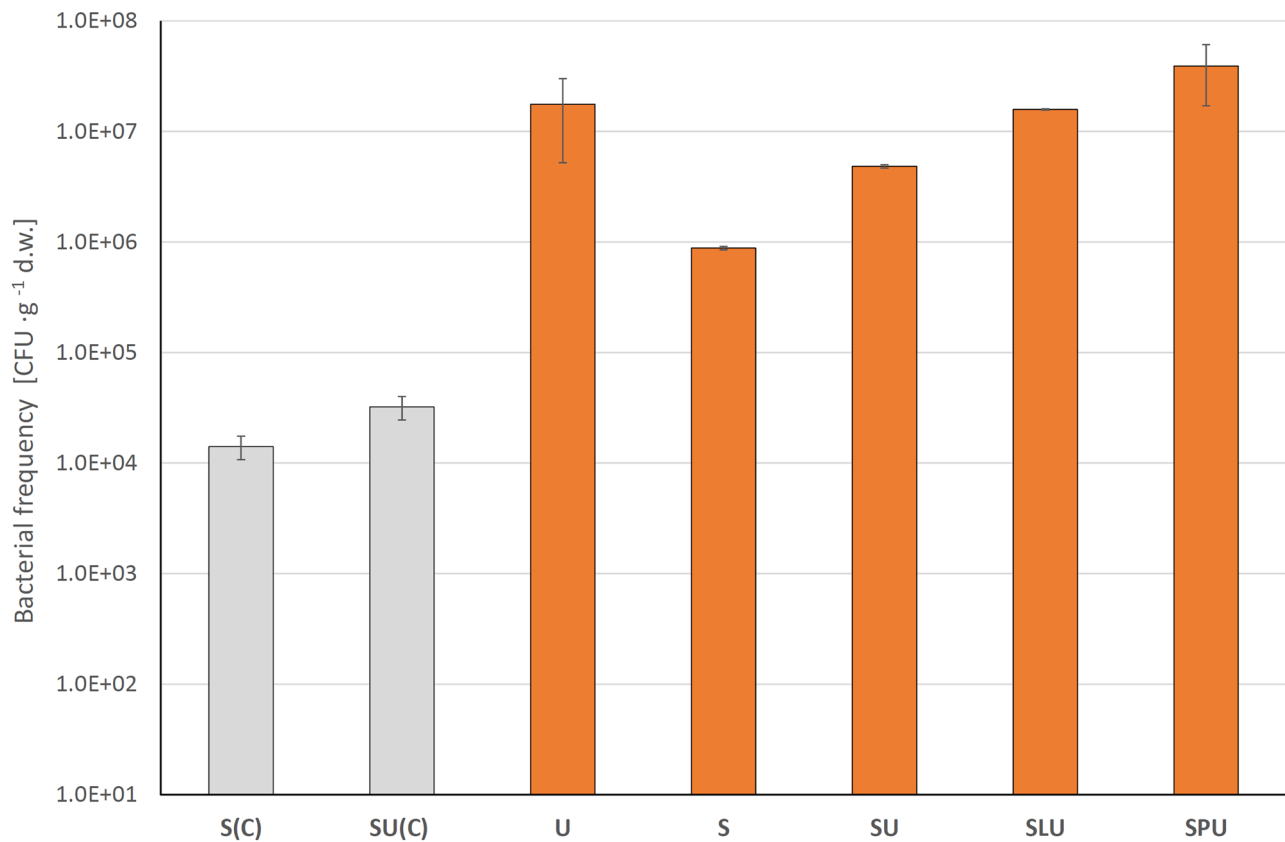


Fig. 5 Changes of bacterial population in the root zone of *M. crystallinum* plants after 30-day cultivation, as dependent on the growth in different substrates containing bottom sediments from Lake Chechło. Error bars show standard error. Substrate sample descriptions: U, universal soil substrate with sand; S, bottom sediments; SU, bottom sediments+universal soil substrate with sand; SLU, bottom

sediments + limed soil + universal soil substrate with sand; SPU, bottom sediments + plant ash + universal soil substrate with sand. Bars S(C) and SU(C) represent the frequency of bacteria determined in substrates before plant growth: bottom sediment and bottom sediment + universal soil and sand, respectively

was observed in combination with bottom sediments + limed soil + universal soil substrate with sand (SLU) and bottom sediments + plant ash + universal soil substrate (SPU) (except for *A. fischeri*). Regardless of the treatments, the incubation conditions (before and after the test plant growth) did not significantly affect the response of the test organisms; however, changes in toxicity classification of the analyzed substrates were observed. The treatments with S and SU were classified as toxicity class III (acute toxicity) or IV (high acute toxicity), treatments with SPU were classified as toxicity class II (slight acute toxicity) and III (acute toxicity), whereas the SLU combination as class I (no acute toxicity) (Table 6).

4 Discussion

The rationale for proposing *M. crystallinum* as a model suitable for phytoremediation of metal-contaminated bottom sediments was based on its unusual biochemical and physiological characteristics. Some of the common ice

plant capabilities seem to be particularly advantageous in terms of reclamation of degraded soils, especially remediation of heavy metal contamination. Namely, maintenance of high levels of antioxidant systems (both enzymatic and low molecular ones) in *M. crystallinum* may contribute to its resistance against heavy metal toxicity resulting from various cellular reactions, especially those involving free radicals and reactive oxygen species (Libik et al. 2019). Then, large amounts of two- and tricarboxylic acids (malate and citrate) produced under stress, especially in the CAM phase, can be pumped out through the roots as soil-enriching exudates. These compounds could interact with the metal ions occurring within the rhizosphere, especially by binding cations. On the other hand, they are expected to have strong positive biostimulating action on soil microbiota, whose interaction with soils involves various mechanisms and may lead to altered bioavailability of elements, either by their mobilization or immobilization.

In this study, for the first time, phytoremediation tests with the common ice plant were carried out by applying

Table 6 Toxicity classification of the tested substrates containing bottom sediments from Lake Chechło before (1) and after (2) growing *Mesembryanthemum crystallinum* plants. Different letters next to the numbers indicate statistically significant differences at $p \leq 0.05$

Biotest		S 1	S 2	SU1	SU2	SLU1	SLU2	SPU1	SPU2
<i>S. alba</i>	Germination inhibition %	-29 ± 0 a	-7 ± 30 a	-7 ± 30 a	-29 ± 0 a	-29 ± 0 a	4 ± 45 a	-18 ± 15 a	-18 ± 15 a
	Roots growth inhibition %	23 ± 8 d	-24 ± 9 cd	-54 ± 23 bc	-80 ± 32 abc	-100 ± 15 ab	-68 ± 5 abc	-76 ± 37 abc	-117 ± 27 a
<i>L. sativium</i>	Germination inhibition %	0 ± 0 a	8 ± 12 a	0 ± 0 a	8 ± 12 a	33 ± 24 a	8 ± 12 a	8 ± 12 a	0 ± 0 a
	Roots growth inhibition %	64 ± 6 b	0 ± 32 ab	25 ± 9 ab	4 ± 34 ab	12 ± 4 ab	-6 ± 1 ab	-13 ± 8 a	-2 ± 20ab
<i>S. saccharatum</i>	Germination inhibition %	4 ± 15 a	-29 ± 0 a	-7 ± 30 a	-29 ± 0 a	-29 ± 0 a	-7 ± 30 a	-29 ± 0 a	-29 ± 0 a
	Roots growth inhibition %	59 ± 15 c	16 ± 11 ab	-35 ± 17 a	-9 ± 1 ab	16 ± 5 ab	8 ± 11 ab	27 ± 26 b	10 ± 5 ab
<i>H. incongruens</i>	Mortality	90 ± 14 c	100 ± 0 c	85 ± 21 c	100 ± 0 c	0 ± 0 a	0 ± 0 a	35 ± 7 b	85 ± 7 c
	Growth inhibition	97 ± 4 b	100 ± 0 b	60 ± 57 b	100 ± 0 b	-31 ± 15 a	-46 ± 16 a	56 ± 3 b	75 ± 14 b
<i>A. fischeri</i>	Luminescence inhibition %	97 ± 1 d	93 ± 2 d	85 ± 1 cd	73 ± 6 c	-22 ± 2 b	-17 ± 5 b	-62 ± 6 a	-21 ± 7 b
Toxicity class		III	IV	III	IV	I	I	II	III

U universal soil substrate with sand, S bottom sediments, SU bottom sediments+universal soil substrate with sand, SLU bottom sediments + limed soil + universal soil substrate with sand, SPU bottom sediments + plant ash+ universal soil substrate with sand

the plants to treat real polluted environmental samples, not artificially doped with any contaminants. The sample material was formed upon long-term spontaneous processes of cumulating organic matter together with various contaminants including metals and other potentially toxic elements (Zawisza et al. 2014; Baran et al. 2017; Koniarz et al. 2022a, b). It is to note, however, that the experiment was carried out in pot tests under controlled conditions and that the plants were grown on variant substrates composed of the sediments admixed with alternative materials, added to improve nutrient availability and water–air conditions. Still, the ecological risk evaluation based on combined biotests showed acute (class III) toxicity of the studied samples.

We believe that the results of our work can contribute to development of proper phytoremediation-based methods for the management of various bottom sediments contaminated with trace metals. However, it is difficult to discuss our data in light of the achievements of other authors who dealt with sediment remediation. This is because most of the documented studies involved different material such as urban sludge, sewage sludge, or sediment mixtures with soil, usually uncontaminated or containing low concentrations of metals (Eid and Shaltout 2016; Wyrwicka et al. 2019). For the case of bottom sediments collected from rivers, lakes, and retention reservoirs, the majority of studies deal with the metal pollution risk assessment and bioindication (Parzych et al. 2016; Małachowska-Jutcz and Gumińska 2018) as well as phytoremediation with aquatic plants (Wu et al. 2014; Parzych 2016; Parzych et al. 2016; Małachowska-Jutcz and

Gumińska 2018; Skorbiłowicz et al. 2018). In a recent thorough review, Nawrot et al. (2021) present a worldwide perspective on the problem of metal contamination in anthropogenically affected bottom sediments. The authors discuss geochemical and ecotoxicological indices for sediment pollution evaluation, provide a comprehensive list of plant bioindicators useful for biomonitoring and propose *Salix* hybrid cultivars as plants capable of phytostabilization and rhizofiltration of Cu, Zn, and Ni.

The data regarding *M. crystallinum* biomass formed after 30-day growth confirmed that the plant was very tolerant to harsh conditions of numerous coexisting contaminants and to potential toxicity of the substrates tested, including the sole bottom sediment. None of the substrates, except for a moderate growth-inhibitory effect caused by raw bottom sediments, induced visible morphological symptoms of the metal stress. The resultant biomass produced by the common ice plant was comparable to that obtained at optimal conditions. The observed growth potential suggests the applicability of *M. crystallinum* for phytostabilization of soils containing bottom sediments. Along with the plant growth, in all the cases a dynamic development of rhizospheric microbiota was observed, allowing for population density increase up to the levels of 10^7 CFU g^{-1} , that is values achieved in the reference soil sample U. Frequent occurrence of soil bacteria is crucial in terms of soil successful rehabilitation, but it also appears as an important factor modulating metal accumulation processes by changing bioavailability of many elements through microbe–metal interactions in soils (Huang 2004).

Clearly, the root zone of *M. crystallinum* provided local non-inhibitory microenvironment enabling bacteria development in substrates containing toxic and potentially toxic contaminants. It is also very likely that the plants revealed biostimulatory effect on microbiota population growth due to the complex metabolism as discussed above.

The collected material contained 247.1 mg kg^{-1} d.w. of sodium, which is a value much elevated compared to the universal soil (agricultural substrate U, Na content of 43.2 mg kg^{-1} soil d.w.) used as a reference. Such a concentration is potentially inhibitory or toxic to many plant species (Kronzucker et al. 2013) but this was not the case for *M. crystallinum* which grew well and revealed high Na accumulation yield together with very high translocation capabilities from roots to shoots (TF values ranging from 8.50 to 33.33, Table 5). These capabilities of Na uptake, accumulation, and translocation are in accordance with the earlier observations on *M. crystallinum* as a halophytic plant (Kuznetsov et al. 2000; Libik et al. 2019). In this study, additional evidence has been collected to prove the great potential of Na accumulation (Tables 2 and 3), reaching the value of 104.67 g kg^{-1} d.w. in shoots (the SU variant, Table 3). The observed sodium accumulation patterns resemble that of the plants grown in model laboratory conditions on saline soils (Libik et al. 2019), which provides further support for adaptational capabilities of *M. crystallinum* interacting with metal-contaminated bottom sediments.

Several other metals, i.e. Cd, Cu, Cr, Fe, Ni, Pb, and Zn, were present in the studied bottom sediments at levels considerably higher than in the reference substrate (U) (cf. Table 1). Some of the metals were present in high concentrations or in chemical forms of severe toxicity. The respective values given in Table 1 can be compared to the limits determined by the European or Polish regulations. The criteria of the European Commission were set for growth media, soil enhancers, and horticultural media used in organic farming and define the permissible levels for Cd, Cu, Cr, Ni, Pb, and Zn as 1.0, 100, 100, 50, 100, and 300 mg kg^{-1} d.w., respectively (EU Commission Decision 2015). Polish legislation lacks precise regulations concerning the natural use of bottom sediments, and therefore the total content of metals in sediment-waste mixtures need to be referred to the criteria related to permissible levels in municipal sewage sludge used in agriculture and for land reclamation for agricultural purposes. These values for Cd, Cu, Cr, Ni, Pb, and Zn were established as 20, 1000, 500, 300, 750, and 2500 mg kg^{-1} d.w., respectively (Ordinance of the Minister of the Environment 2015). Also, Polish standards of assessing soil surface pollution, specified in the Regulation of the Minister of the Environment (Ordinance of the Minister of the Environment 2016), can be applied. In these regulations, the upper permissible limits for the total content of Cd, Cu, Ni, Pb, and Zn for class III of surface soils and grounds

(classified as forests, agricultural and wooded lands, ecological grounds, recreation and leisure areas) are the following: 10, 300, 300, 500, and 1000 mg kg^{-1} d.w., respectively. Taken the values listed in Table 1 for the respective metals in bottom sediments, it can be seen that for the cases of Cu, Cr, and Ni, the concentrations detected (56.55 , 3.86 , and 9.10 mg kg^{-1} d.w., respectively) were below the cited limits and it can be assumed that the environmental risk posed by these metals was not severe. In turn, the concentration of lead ($244.40 \pm 3.63 \text{ mg Pb kg}^{-1}$) exceeded the EU limit and might be regarded as potentially toxic, whereas the contents of Cd and Zn (45.04 ± 0.81 and $9243 \pm 207 \text{ mg kg}^{-1}$ d.w., respectively) were well above the limiting values defined for agricultural and wooded soils up to class III as well as the other cited norms. It should be emphasized that, apart from standard regulations, the determined levels of Cd, Pb, and Zn significantly exceed the toxicity thresholds for soils containing pollutants of anthropogenic origin, as established by Kabata-Pendias (2010) (1.0, 70, and $100\text{--}500 \text{ mg kg}^{-1}$ d.w., respectively). Mixing of the bottom sediments with unpolluted materials led to a decrease in contamination level (cf. rows S and SU, SLU, SPU in Table 1); however, while considering the abovementioned values and limits, also these combined substrates should be regarded as hazardous or toxic.

While examining phytoremediation with *M. crystallinum*, it was of particular interest to verify the potential of metal accumulation in plant tissues accompanied by removal of these pollutants from the substrates. This idea was inspired by earlier model studies showing *M. crystallinum* capability of efficient uptake of Ni (Amari et al. 2014), Cd (Shevyakova et al. 2003), Cu and Zn (Kholodova et al. 2005), and more recently, Cd and Cr (Śliwa-Cebula et al. 2020).

As expected, the plant revealed some mechanisms enabling accumulation of several metals and hence the BAF coefficients were determined to characterize the accumulation potential (see Results). For some cases these coefficients were above 1.0 thus confirming efficient accumulation processes. Seemingly, highly variable content in roots or in shoots and differing BAF values point to divergent strategies of *M. crystallinum* response to different metals. In addition, the uptake/accumulation processes and the resultant BAF values were obviously affected by broad concentration ranges of the metals in variant substrates and by different bioavailabilities of these elements. Also, at this point, the cases of Cu, Cr, or Ni should be considered since their contamination levels were not severe relative to non-polluted soils and therefore might not result in generation of physiological stress reactions enough to trigger plant remediation actions, uptake, and accumulation.

Root-to-shoot translocation factors (TF) were calculated to further reveal *M. crystallinum* suitability for phytoremediation of heavy metal contamination. As shown in Table 5, in several

cases, TFs were greater than 1.0, which indicates that plant aboveground parts served as final deposition targets for phytoextracted metals. It should be noted here that efficient root-to-shoot translocation is not enough to ensure extensive phytoremediation effect and the $TF > 1$ criterion must be coupled with a high accumulation yield. Similar to other accumulation-related parameters, the TF values varied broadly and were dependent on particular metal considered and experimental variant tested. As discussed above, the plausible explanation of these facts is the complexity of stress reactions to different heavy metals for different substrates.

From among the studied metals, only in the case of zinc contamination remediation with *M. crystallinum*, the mechanism of hyperaccumulation can be suggested. According to the novel concentration criteria provided by Van der Ent et al. (2013), for Zn, the plant foliage should contain $> 3000 \text{ mg kg}^{-1} \text{ d.w.}$ while the root-to-shoot translocation factor should be greater than 1.0 (Ali et al. 2013; Van der Ent et al. 2013). In this study, values well above the cited thresholds were obtained for two cases: (1) sole bottom sediment (S) where the shoot accumulation level reached the highest value of $11,052 \pm 2132 \text{ mg kg}^{-1} \text{ d.w.}$ and TF was determined as 1.66; (2) in the variant SU (bottom sediment mixed with universal soil + sand), where the Zn content was assessed as $7014 \pm 737 \text{ mg kg}^{-1} \text{ d.w.}$ and $TF = 4.02$. In addition, the respective BAF coefficients determined for shoots were also above 1.0 (1.20 and 1.52) thus confirming high Zn accumulation yields. The reported Zn phytoremediation parameters are novel findings and to our best knowledge there is only one contribution of Kholodova et al. (2005) available, in which the *M. crystallinum* tolerance to Zn and efficient Zn uptake was documented. Here, based on the treatment of real samples, we show the plant particular applicability for rehabilitation of Zn-contaminated soils. This observation is particularly interesting in the context of our previous data proving biotechnological potential of *M. crystallinum* toward reclamation of lands affected by cadmium and chromium (Nosek et al. 2019; Śliwa-Cebula et al. 2020).

We are aware that the study was not carried out under real environmental growth conditions, involving plant natural habitats. The need to confirm accumulation capabilities by naturally populated plants was emphasized by Van der Ent et al. (2013). Also, these authors argue against hyperaccumulation data based on hydroponics, artificially spiked soils or acidified soils. However, we stress that our material was based on real samples, spontaneously formed as sediments in Lake Chechło, and no actions were taken to modify their content except for mixing with other materials. We believe that at least in some respects the present study has brought us close to the real environmental conditions. Nevertheless, further complementary field-studies are still

required to finally prove Zn hyperaccumulation potential of the common ice plant.

After 30-day treatments, the content of metals in the bottom-sediment containing substrates was significantly lower (compare respective rows marked 1 and 2 in Table 1), although the experiment duration was not enough to achieve considerable removal of all the hazardous elements. The maximum removal yields were presented in Results and depended on particular variants of the final substrate composition. It is likely that *M. crystallinum* treatment made it possible to mobilize several of the elements and phytoextract them partially from the bottom sediments. Note that the lowest removal rate was observed for the case of Cd (maximum 18% removal, only for the single variant S; Table 1). This result is in good agreement with our recent data based on model laboratory studies, where *M. crystallinum* was shown to be cadmium-tolerant excluder rather than accumulator (Nosek et al. 2019; Śliwa-Cebula et al. 2020), although in this work it accumulated Cd to some degree both in roots and shoots (in variants S, SU, and SPU).

The effectiveness of phytoremediation process can be fully assessed upon analyses of the elemental content in the substrate before and after reclamation, determination of the degree of contaminant accumulation in plant tissues, and finally, establishing the environmental toxicity risks whose reduction contributes most significantly to full soil rehabilitation.

In the case of ecotoxic samples such as polluted bottom sediments, the application of Phytotoxkit, Ostracodtoxkit F, and Microtox biotests appear as useful tools to evaluate full spectrum of environmental risks (Czerniawska-Kusza and Kusza 2011; Baran and Tarnawski 2015; Baran et al. 2019a). It was shown that the studied bottom sediments posed severe environmental threat due to exhibiting class III toxicity (acute), which was likely a result of numerous, coexistent pollutants of different mobilities and bioavailabilities. Furthermore, treatment with *M. crystallinum* led to single-step toxicity increase (for the cases: S, SU, and SPU). The observed ecotoxicity changes can be accounted for by mobilization of some of the potentially toxic elements. Such mobility enhancement most probably results from a concerted action of the plant itself (e.g. by producing root exudates) together with the biostimulated rhizospheric microbiota and an independent, long-term study is required to verify whether the prolonged phytoremediation would lead to the final loss of toxicity. Nevertheless, if the toxicity problem persists, *M. crystallinum* might still be used for bottom sediment phytostabilization as it revealed high physiological tolerance and adaptation potential allowing for undisturbed growth, while the other ecotoxicologically tested plants such as *S. alba*, *L. sativum*, or *S. saccharatum* were strongly inhibited.

An alternative way enabling decreased substrate toxicities was proposed for the SLU variant in which the mixture of bottom sediment and universal soil was amended with the limed soil. In this case, the toxicity category was reduced to class I (no acute toxicity) and after treatment with the common ice plant it remained unchanged. Importantly, although the described action proved efficient in terms of ecotoxicity mitigation, the substrate enriched with calcium was characterized by markedly reduced mobility (and thus bioavailability) of several elements (cf. Table 1, row SLU 1, for the decreased content of the extractable fractions of Cd, Fe, Ni, Pb, Zn). This, in turn, resulted in considerably lowered accumulation levels of these metals, both in roots (Table 2) and shoots (Table 3), and negatively affected their removal rate (Table 1, row SLU 2).

Taking into consideration all the described approaches for processing and treatment of toxic bottom sediments with *M. crystallinum*, it is clear that efficient phytoremediation requires a choice of the most appropriate variant of substrate formulation followed by the plant growth. In any case, the application of the common ice plant might be beneficial in terms of proper management of the material: either enabling phytoextraction and removal of potentially toxic elements from the sediments, or allowing for sediment-containing soils revitalization via phytostabilization to create vegetation cover.

5 Conclusions

This is the first study reporting potential applicability of the common ice plant *M. crystallinum* to treat substrates containing bottom-sediments obtained from industrially affected areas. The plant interaction with variant substrate mixtures as well as its action on particular mineral content was found to be diverse and complex, which can be explained by unique physiological and biochemical characteristics of the applied model, especially high stress resistance and adaptable photosynthetic metabolism.

The studied plant proved to be capable of growth under very harsh conditions of toxic and poor-quality substrates, while, depending on the experimental variant, accumulating metal contaminants (Na, Cd, Cu, Cr, Ni, Pb, and Zn) and partially removing them (especially Cu, Pb, Cr, and Zn) from the bottom sediment-containing soils. Some of the potentially toxic elements (the most pronounced effect observed for Na and Zn) were translocated from roots to shoots thus indicating a mechanism of efficient phytoextraction. Soil microbiota was significantly biostimulated in the plant root zone and grew to achieve population densities close to that observed in agricultural soils. Ecotoxicological tests of the treated samples showed one-step increase of toxicity degree upon 30-day growth of *M. crystallinum*, which was most likely a result of

the element bio-mobilization by the common action of both the plants and populous rhizospheric bacteria. However, the toxicity effect could be mitigated by admixing the sediment-containing substrates with the limed soil.

Taken together, the results of this study allow us to conclude that the common ice plant should be considered a highly suitable candidate for environmental applications involving phytostabilization and/or phytoremediation of soils containing post-industrial, contaminated bottom sediments.

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Data availability All data will be made available upon request.

Declarations

Conflict of interest The authors declare no competing interests.

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