

Thesis of doctoral (PhD) dissertation

**REMEDICATION AND RESTORING MARGINAL LANDS WITH
BIOTECHNOLOGICALLY PROPAGATED GIANT REED (*Arundo donax* L.)**

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1. INTRODUCTION AND AIMS OF STUDY

Giant reed (*Arundo donax* L.) is a perennial rhizomatous grass (Poaceae family), native to the freshwater regions of Eastern Asia, but nowadays considered as a sub-cosmopolitan species given its worldwide distribution. It is a hydrophyte, growing along lakes, streams, drains and other wet sites. The genus *Arundo* is able to reach the height of 14 m and is among the fastest-growing terrestrial plants. It can produce more than 50 t ha⁻¹ aboveground dry biomass. As a consequence of its high and fast biological productivity, giant reed is widely cultivated to yield non-food crop that can meet requirements for energy, paper pulp production, bio-fuels and construction of build materials, but it has other different uses such as music tools with stem, medicine with roots and soil erosion control through re-vegetation.

Giant reed displays unique physiological features whereby it readily absorbs and concentrates toxic chemicals from contaminated soil with no appreciable harm to its own growth and development. It is one of the mostly used plants as a trace element bio-accumulator, especially via phytoremediation processes, due to its capacity of absorbing contaminants such as metals that cannot be easily biodegraded. Giant reed can grow in different environments with spacious ranges of pH, salinity, drought and trace metals without any symptoms of stresses and can easily adapt to different ecological conditions and grow in all types of soils. However, because of its great adaptability to different ecological conditions, giant reed is considered noxious invasive weeds in riparian habitats throughout the world.

In 2010 the pond dam of an aluminium manufacturing plant in Hungary broke and flooded many towns with toxic red mud. At least 10 people were dead and over 150 hospitalized. Bauxite residue is often referred as red mud due to the colour of the bauxite ore and iron oxides. Red mud is separated during the refining process. The production of 1 t of alumina generally results in the creation of 1–1.5 t of red mud. Red mud is toxic for the environment due to high alkalinity, salinity and trace metals.

Soil quality refers to the capacity of soil to perform agronomic and environmental functions. Changes in soil quality, resulting from, for example, bush fires or the presence of waste residues from bauxite mining (through the Bayer process) can be measured through physical, chemical and biological indicators. The chemical indicators include pH, EC, soil organic carbon, phosphorus availability, nutrient

cycling, and the presence of contaminants such as heavy metals, organic compounds, and radioactive substances. These indicators determine the presence of soil-plant-related organisms, and nutrient availability. The biological indicators that have been widely studied are the chemical compounds or metabolic products of organisms, particularly enzymes such as cellulases, arylsulfatase, phosphatases, urease, dehydrogenase related to specific functions of substrate degradation or mineralization of organic N, S or P. Soil enzymatic activity assays act as potential indicators of ecosystem quality being operationally practical, sensitive, integrative, described as "biological fingerprints" of past soil management that relate to soil tillage and structure.

The objective of this study was to monitor the short- and long-term effects of the red mud on the Hungarian soil quality using synthetic plantlets of *A. donax* L. and the effects of red mud on growth and chemical composition of giant reed plantlets using red mud, mud-polluted soil, mud/control soil mixture and control soil. The trace metal uptake and translocation by seedlings were highlighted as well.

In addition, we aimed to investigate the influence of giant reed plants on ecosystems of red mud-amended soil (in case of adding red mud to marginal and wetland soils) especially after exposure to high temperatures, to monitor the changes in soil microbial community and soil enzyme activities under giant reed cultivation in case of heated soil compared to control soil, and to investigate the effects of soil heating on giant reed seedling growth and biomass production.

2. MATERIALS AND METHODS

2.1. Sampling and preparation of soils and red mud

In early spring 2011, two composite top-soil samples (0-25 cm) were collected from the demonstration Garden of Debrecen University, Debrecen city (47° 32' 0" N; 21° 38' 0" E). The first soil sample (S1) was collected from a field, which had grass coverage for the previous 9 years; S2 was collected from a field of 1-year old giant reed. An additional two composite soil samples and a sample of red mud (also 0-25 cm) were collected from Kolontár town, western Hungary (47° 5' 3.97" N, 17° 28' 30.08" E) in October 2010 after two weeks from the Alumina factory catastrophe: S3 was collected from a field with a maize, sun flower and rapeseed crop rotation; S4 (mud-polluted soil) was collected from a red mud polluted field. The red mud sample was collected from flooded grassland. A mixture (S5) of red mud and S3 soil was made by ratio 1:1 by weight. After sampling, a portion of each fresh soil sample was sieved through 8 mm

and used directly for enzyme activity measurements and microbial counts. The rest was air dried 25°C, ground in stainless steel crushing machine and sieved at 2 mm, then kept in plastic bags for further chemical analysis at 25°C.

2.2. Plant Material

The plant material used for the current study was somatic embryo-derived plantlets of the Blossom ecotype of giant reed (*Arundo donax* L.) obtained from the University of South Carolina and propagated in the Ottó Orsós Laboratory, Department of Plant Biotechnology, Debrecen University, Hungary. Sterile plantlets were directly transplanted to autoclaved samples; the other sterile plantlets were acclimatized to the greenhouse environment before potting in non-autoclaved samples.

2.3. Bioinocula treatments

Two different bioinocula were used after 2 days from plantation to enhance the bacterial community around the root system of giant reed; a commercial product (*Azotobacter croococcum* and *Bacillus megaterium*) was obtained from Hungarian market. The other bioinoculum was prepared by extracting the bacterial community in rhizosphere and rhizoplane fractions of giant reed's roots. To prepare this bioinoculum, the roots of giant reed were collected from 1-year-age giant reed from the experimental farm at Debrecen University; the root extraction method used was based on that described by Wieland et al. (2001). After isolation of both rhizosphere and rhizoplane fractions' bacteria, the fractions were suspended in sterile distilled water to get total bacterial count density of 1×10^8 CFU (colony-forming units) per ml.

2.4. Plant Analysis

The giant reed plants were harvested three months after planting. Fresh mass and length of shoot and root portions were measured. For NPK content, a 0.5 g sample was digested by addition of 10 ml H₂SO₄ and 1.0 ml HClO₄. The tissue concentration of N, P, and K was determined both colorimetrically, and by flame photometer.

2.5. Soil Chemical Analysis

Available N, P, K and soil organic carbon (SOC) analyses were carried out in triplicate for each treatment. Available P was determined colorimetrically. Available N was evaluated by the macro Kjeldahl digestion procedure. Available K was determined

using flame photometry. SOC was determined using the modified Walkley-Black wet combustion method.

2.6. Soil Microbiological Components Analysis

Soil microbial activities were measured in fresh samples before potting and after harvesting giant reed. The serial dilution pipette method was used for the microbial counts on different selective media (Allen, 1953). Phosphatase activity was measured as described by Szegi (1979). Dehydrogenase, urease and catalase activities were determined using the procedures of (Tabatabai, 1994; Kandeler and Gerber, 1988; Guwy et al., 1999) respectively.

2.7. Germination assay

To determine the effect of red mud on germination percent of some vegetables, a germination assay was carried as follow; 250 g of S1, S2, S3, S4, S5, and red mud were transferred to plastic pots then irrigated by tap water to saturation capacity. Per each pot, 30 seeds of radish (*Raphanus sativus*), carrot (*Daucus carota*) and Tobacco (*Nicotiana tabacum*) were sowed. The numbers of germinated seeds were counted. This work was repeated three times.

2.8. Statistical Analysis

Data analysis was performed using Microsoft Excel 2003 (mean values and standard deviation). The effect of giant reed on soil properties before and after experiment was compared with paired t tests. An effect was considered significant at the 5% level. The three-way fixed-effects analysis of variance (ANOVA) was conducted using the SPSS 13.0 software package (SPSS Inc., Chicago, IL). Dependent variables were checked for normality and homoscedasticity and transformed as necessary. Separation of means was performed by post hoc test (Scheffe test), and significant differences were accepted at the level $p < 0.05$. All values are presented as untransformed means and standard deviations.

3. RESULTLS AND DISCUSSION

3.1. Phytoremediation of bauxite-derived red mud

3.1.1. Characteristics of red mud

The dry red mud is alkaline with pH 9.8, the pH of semisolid red mud at flooding was 12 (Ruyters et al., 2010), the pH of control soil, near to catastrophe's site, was 7.86, and the pH of mud-polluted soil is drastically increased after accident (pH 8.24) as a result of covering soil surface with thick layer of wet red mud more than 10 cm; at that time, red mud had enough time to penetrate the soil profile and affects soil pH strongly. EC of red mud (1.74 dS m^{-1}) and mud-polluted soil, S4, (0.91 dS m^{-1}) was four- and twofold more than the EC of control soil (S3), respectively, as a result of high Na content in red mud (Brunori et al., 2005). The liquid phase of red mud has an EC of 20 dS m^{-1} . Ruyters et al. (2010) reported that dry red mud has EC of 2.4 dS m^{-1} but EC of liquid phase of red mud was 20.2 dS m^{-1} . High organic carbon content was measured from red mud (23.2 g kg^{-1}) compared with control soil, S3, (19.5 g kg^{-1}). Moreover, the available N, P and K contents of red mud were also higher than in mud-polluted soil (S4) and control soil (S3) (Table 1).

The highest number of total bacterial count ($9.56 \times 10^7 \text{ CFU ml}^{-1}$) was recorded at mud-polluted soil (S4). However, red mud also contained higher number of bacteria than control soil (S3). The *Azospirillum sp.*, *Azotobacter sp.* and spore-forming bacilli were separately grown, and both of them were in the highest number in red mud. As far as fungi are concerned, the red mud contained 2.6-fold less than control soil (S3), maybe because the pH was too high for fungal growing. However, there is a tiny relationship between the total number of microbes and soil enzyme activity. Table 1 showed that among the measured soil enzyme activities the dehydrogenase and urease activities were higher in red mud than in other soils. On the contrary, the phosphatase and catalase were lower in red mud than in other soils.

Table 1: Characterization of soils and red mud after *Arundo donax* L. plantation as affected by Arundo's root extraction (Ar. ex.) and commercial biofertilizers (CB)

Property	S3			S4			Red mud			S5		
	Cont.	Ar. ex.	CB	Cont.	Ar. ex.	CB	Cont.	Ar. ex.	CB	Cont.	Ar. ex.	CB
pH	8.33	8.38	8.30	9.03	9.04	8.97	9.70	9.66	9.70	9.30	9.23	9.30
EC (dS m ⁻¹)	0.35 ^{a,a}	0.35 ^{a,a}	0.32 ^{a,a}	0.88 ^{b,a}	0.87 ^{b,a}	0.83 ^{b,a}	1.27 ^{c,a}	1.26 ^{c,b}	1.39 ^{c,c}	1.12 ^{d,a}	0.85 ^{b,b}	0.99 ^{d,c}
SOC %	26.3 ^{a,a}	24.9 ^{a,a}	24.6 ^{a,a}	23.8 ^{a,a}	24.9 ^{a,a}	23.5 ^{a,a}	23.1 ^{a,a}	23.7 ^{a,a}	24.6 ^{a,a}	24.8 ^{a,a}	24.7 ^{a,a}	25.7 ^{a,a}
NH ₄ ⁺ -N (mg kg ⁻¹)	10.8 ^{a,a}	12.8 ^{a,a}	13.2 ^{a,a}	17.9 ^{b,a}	17.1 ^{ab,a}	18.9 ^{b,a}	16.8 ^{b,a}	19.0 ^{b,a}	18.2 ^{ab,a}	8.1 ^{a,a}	6.0 ^{c,a}	6.4 ^{c,a}
Avail. P (P ₂ O ₅ g. kg ⁻¹)	6.3 ^{a,a}	6.8 ^{a,a}	8.0 ^{a,a}	9.0 ^{ab,a}	10.0 ^{ab,a}	7.6 ^{a,a}	13.9 ^{b,a}	11.8 ^{b,a}	11.6 ^{a,a}	10.2 ^{ab,a}	9.8 ^{ab,a}	9.6 ^{a,a}
Avail. K (K ₂ O mg kg ⁻¹)	267 ^{a,a}	367 ^{b,a}	633 ^{a,a}	367 ^{a,a}	300 ^{a,a}	367 ^{a,a}	767 ^{a,a}	733 ^{a,a}	667 ^{a,a}	400 ^{a,a}	467 ^{a,a}	467 ^{a,a}
Dehydrogenase activity	105 ^{a,a}	116 ^{a,a}	126 ^{a,a}	143 ^{a,a}	108 ^{a,a}	167 ^{ab,a}	161 ^{a,a}	142 ^{a,a}	216 ^{b,a}	117 ^{a,a}	119 ^{a,a}	145 ^{ab,a}
Phosphatase activity	0.17 ^{b,a}	0.17 ^{b,a}	0.16 ^{a,a}	0.18 ^{ab,a}	0.17 ^{a,a}	0.16 ^{a,a}	0.16 ^{a,a}	0.14 ^{b,b}	0.17 ^{b,a}	0.19 ^{b,a}	0.17 ^{ab}	0.21 ^{b,c}
Urease activity	1190 ^{a,a}	2247 ^{a,b}	317 ^{a,c}	1255 ^{a,a}	1262 ^{b,a}	1138 ^{b,a}	2877 ^{b,a}	732 ^{b,b}	471 ^{ab,b}	1825 ^{a,a}	744 ^{b,b}	490 ^{ab,b}
Catalase activity	17 ^{a,a}	14 ^{a,a}	16 ^{a,a}	106 ^{b,a}	106 ^{b,a}	108 ^{b,a}	18 ^{a,a}	15 ^{a,a}	21 ^{a,a}	41 ^{c,a}	42 ^{c,a}	46 ^{c,a}

S3: Control soil, **S4:** Mud-polluted soil and **S5:** Mud/control soil mixture by ratio (1:1) by weight. **Ar. ex:** arundo' root extraction, **CB:** commercial biofertilizer, **Cont:** control. pH of 1:2.5 suspension red mud/ water or soil/ water, EC of 1:5 extraction red mud/ water or soil/ water
Different letters on the left show significant differences among the same treatment for the different soil types in same row. Different letters on the right show significant differences among the same soil types for the different treatments in same row.

Dehydrogenase activity in µg TPF g⁻¹ soil, phosphatase activity in mgP₂O₅/100g/2h, urease activity in NH₄⁺ mg/100 g and catalase in O₂ ml/ 2 min

3.1.2. *Arundo* growth and red mud

The main problems with red mud are high salinity, pH and trace metal contents. Ruyters et al. (2010) have reported that the red mud reduced the shoot yield of barley seedlings by 25 % when red mud was applied to normal soil with 5 %. The biotechnologically propagated *A. donax* L. plantlets growing on pure red mud did not show any toxic symptoms. The plant toxicity, trace metal availability and biomass production for *A. donax* L. were tested with red mud, mud-polluted soil (S4), mud/control soil mixture (S5) and control soil (S3).

The vegetative parameters of *A. donax* L. are showed in Table 2, after 3 months of pot experiment in greenhouse. To ensure the principle of natural attenuation, our treatments were using microbial extraction prepared from giant reed roots (Ar. ex) compared with the commercial biofertilizer (CB). A two-way ANOVA revealed insignificant differences between all treatments for plant fresh weight, plant length and number of new buds. However, ANOVA has revealed significant effects for red mud on giant reed fresh weight, whereas red mud recorded the highest fresh weight for plants compared to mud-polluted soil (S4) and control soil (S3). Mud/control soil mixture (S5) has induced the giant reed growth compared with control soil (S3); the plants were so dark green (unpublished data) and have higher fresh weight than control soil (S3). As mentioned above, it becomes so clear that giant reed is red mud-tolerant plant.

Table 2: Vegetative parameters and contents of Fe and Ni in giant reed (*Arundo donax* L.) grown on different soils and red mud using different biofertilizers

Treatments		Fresh weight (g plant ⁻¹)	Plant length (cm)	Number of new buds per plant	Fe (mg kg ⁻¹)		Ni (mg kg ⁻¹)	
					Shoot	Root	Shoot	Root
S3	Cont.	1.63 ^{a,a}	41.0 ^{a,a}	4.0 ^{a,a}	182 ^{a,a}	5022 ^{a,a}	49 ^{ab,a}	76 ^{a,a}
	Ar. ex.	1.49 ^{a,a}	34.3 ^{a,a}	4.0 ^{a,a}	206 ^{a,a}	5429 ^{a,a}	46 ^{a,a}	121 ^{a,b}
	CB	1.23 ^{a,a}	31.5 ^{a,a}	2.8 ^{a,a}	188 ^{a,a}	6461 ^{a,b}	28 ^{a,a}	156 ^{a,c}
S4	Cont.	1.87 ^{a,a}	30.3 ^{a,a}	4.8 ^{a,a}	779 ^{b,a}	11937 ^{b,a}	46 ^{ab,a}	99 ^{b,a}
	Ar. ex.	1.68 ^{a,a}	29.7 ^{a,a}	4.8 ^{a,a}	863 ^{b,b}	10142 ^{b,b}	67 ^{a,a}	97 ^{b,a}
	CB	1.70 ^{a,a}	28.9 ^{a,a}	4.3 ^{ab,a}	635 ^{b,c}	13422 ^{b,c}	61 ^{b,a}	103 ^{b,a}
Red mud	Cont.	2.97 ^{a,a}	34.0 ^{a,a}	6.0 ^{a,a}	899 ^{c,a}	36422 ^{c,a}	67 ^{a,a}	97 ^{bc,a}
	Ar. ex.	3.40 ^{a,a}	39.4 ^{a,a}	5.0 ^{a,a}	1199 ^{c,b}	48822 ^{c,b}	63 ^{a,a}	55 ^{c,b}
	CB	3.44 ^{a,a}	36.1 ^{a,a}	5.5 ^{b,a}	1262 ^{c,b}	50422 ^{c,c}	61 ^{b,a}	193 ^{c,c}
S5	Cont.	3.03 ^{a,a}	37.0 ^{a,a}	4.3 ^{a,a}	641 ^{d,a}	11622 ^{d,a}	37 ^{b,a}	88 ^{c,a}
	Ar. ex.	2.07 ^{a,a}	30.4 ^{a,a}	4.8 ^{a,a}	482 ^{d,b}	8182 ^{d,b}	67 ^{a,b}	79 ^{d,a}
	CB	2.67 ^{a,a}	33.4 ^{a,a}	4.5 ^{ab,a}	632 ^{b,a}	15122 ^{d,c}	51 ^{b,ab}	100 ^{b,b}

S3: Control soil, S4: Mud-polluted soil and S5: Mud/control soil mixture by ratio (1:1) by weight. Cont: control, Ar. ex: arundo' root extraction, CB: commercial biofertilizer.

Different letters on the left show significant differences among the same treatment for the different soil types in same column. Different letters on the right show significant differences among the same soil types for the different treatments in same column

Giant reed has potential effects on the chemical properties of red mud, mud-polluted soil (S4), mud/control soil mixture (S5) and control soil (S5). Three months after giant reed plantation in pots, most of the chemical properties of red mud and soils were influenced positively (Table 1). The electrical conductivity (EC dSm^{-1}) of soils and red mud was decreased in comparison with initial values before plantation by 24.9 % for red mud, 18.1 % for control soil (S3) and 5.9 % for mud-polluted soil (S4). Giant reed has decreased the EC of red mud and mud-polluted soil (S4) by 37 and 4 %, respectively. Moreover, the treatments have significant effects on EC, especially in red mud and mud/control soil mixture (S5). Furthermore, pH of red mud was 9.80 before experiment and decreased to 9.69 by the end of the experiment. It encourages data for using giant reed as phytoremediation plant for soil health point of view; it was the organic carbon content (OC %) in soils after giant reed harvesting. Giant reed increased the OC content of all soils, as a result of the robust growth of root system and huge plant's residues.

3.1.3. Soil enzymes activity

Soil quality becomes one of the most subjects that gained much attention to ensure the sustainability of agricultural systems. Moreover, soil enzymes are used to estimate the adverse effects of various anthropogenic and natural activities on soil health (Masto et al., 2008). Giant reed showed potential affects on soil quality indicators such as organic carbon content as discussed above. Furthermore, most of the tested soil enzyme activities are significantly increased after plantation except phosphatase activity that was decreased in compared to its status before experiment (Table 1). The giant reed took up phosphorous from soil with high rate, which could cause decrease in phosphatase activity. On the other hand, significant increasing was recorded in case of dehydrogenase, urease and catalase activities. Giant reed induced the growth of microflora around roots because of the robust growth of its root system. Free-living bacteria that fix atmospheric nitrogen grow well under such circumstances; therefore, much amount of nitrogen is expected to be fixed in soils after giant reed plantation, and the previous data on available nitrogen emphasize this explanation and also data on urease, dehydrogenase and catalase.

3.1.4. Trace metal uptake, removal and translocation by *Arundo donax* L.

Many articles have reported that giant reed has the ability to uptake and accumulate huge amounts of different trace metals with different concentrations in its root and shoot tissues (Balogh et al., 2012; Papazoglou et al., 2007; Tzanakakis et al., 2009; Mirza et al., 2011). It can grow in contaminated environments without symptoms of toxicity.

Uptake rate depends on certain concentration of such pollutants. The trace metals' tolerance capacity for giant reed has been tested with red mud and different soils through Cd, Co, Pb, Ni and Fe. The first three metals were not detected in both giant reed root and shoot samples. Many reasons may explain why Cd, Co and Pb were undetected: (1) low available concentrations in soil samples, (2) high pH of red mud, mud-polluted soil (S4) and mud/control soil mixture (S5) and (3) high phosphorous content in red mud. The pH of the soil is usually the most important factor that controls metal uptake, with low pH favouring Cd accumulation, and phosphate and zinc decrease Cd uptake (Kirkham 2006). pH of red mud was 9.80 and decreased to be 9.66 inducing Fe and Ni uptake by giant reed roots. Statistical analyses showed that there were significant differences in Fe and Ni contents between treatments itself and between soils itself. The highest Fe and Ni contents in both root and shoot were measured in red mud. Addition of commercial biofertilizer has significantly increased ($P < 0.05$) Fe and Ni uptake by plant roots with soils and red mud. On the other hand, mud/control soil mixture (S5) has increased Fe contents than control soil (S3), but this effect was not observed with Ni (Table 2).

Three months after giant reed plantation, the available concentration of tested trace metals was decreased in soils; however, there were significant differences between treatments and soils (Table 3). The reduction percentage of these metals in soils and red mud was found in the following order:

S3: Co (100) > Fe (80.4) > Cd (78.8) > Ni (57.8) > Pb (55.3)

S4: Co (100) > Cd (83.3) > Fe (74.8) > Ni (61.9) > Pb (60.4)

Red mud: Pb (85.7) > Fe (79.1) > Cd (73.8) > Ni (25.3)

Table 3: Available concentrations using DTPA-extraction of trace metals (mg kg⁻¹) in soils and red mud after giant reed (*Arundo donax* L.) plantation using different biofertilizers

Treatments		Cd	Ni	Pb	Co	Fe
S3	Cont.	(0.22) 0.04 ^{a,a}	(1.20) 0.45 ^{a,a}	(1.46) 0.56 ^{a,a}	(0.64) nd	(52.0) 9.8 ^{a,a}
	Ar. ex.	0.06 ^{a,a}	0.60 ^{a,b}	0.84 ^{a,b}	nd	11.0 ^{a,b}
	CB	0.04 ^{a,a}	0.46 ^{a,a}	0.56 ^{a,a}	nd	10.0 ^{a,a}
S4	Cont.	(0.18) nd	(0.84) 0.28 ^{b,a}	(1.06) 0.36 ^{b,a}	(0.22) nd	(36.0) 8.3 ^{b,a}
	Ar. ex.	0.02 ^{b,a}	0.30 ^{b,a}	0.40 ^{bc,ab}	nd	8.9 ^{b,b}
	CB	0.06 ^{a,b}	0.38 ^{b,b}	0.50 ^{a,b}	nd	10.1 ^{a,c}
Red mud	Cont.	(0.28) 0.06 ^{c,a}	(0.50) 0.32 ^{c,a}	(3.44) 0.51 ^{a,a}	(nd) nd	(74.0) 14.1 ^{c,a}
	Ar. ex.	0.06 ^{a,a}	0.36 ^{b,a}	0.52 ^{b,a}	0.02	14.9 ^{c,b}
	CB	0.10 ^{b,b}	0.44 ^{ab,b}	0.44 ^{a,a}	0.02	17.2 ^{b,c}
S5	Cont.	(-) 0.06 ^{c,a}	(-) 0.38 ^{a,a}	(-) 0.36 ^{b,a}	(-) nd	(-) 10.1 ^{d,a}
	Ar. ex.	0.02 ^{b,b}	0.30 ^{b,b}	0.30 ^{c,a}	nd	7.7 ^{d,b}
	CB	0.02 ^{c,b}	0.30 ^{c,b}	0.26 ^{b,a}	nd	6.4 ^{c,c}

S3: Control soil, **S4:** Mud-polluted soil and **S5:** Mud/control soil mixture by ratio (1:1) by weight. **Cont:** control, **Ar. ex:** arundo' root extraction, **CB:** commercial biofertilizer.

Values in parentheses are data before the experiment, whereas the rest represents data after experimentation

Different letters on the left show significant differences among the same treatment for the different soil types in same column.

Different letters on the right show significant differences among the same soil types for the different treatments in same column.

nd: not detected.

3.1.5. Germination assay

The main goal of germination percentage test was to evaluate the effect of pure red mud and red mud contaminated soil on the edible crops cultivation. Soil (S3) was used as a reference. 30 seeds of each following crops Radish, Carrot and Tobacco were used for this purpose. Table 4 showed the germination percent (%) of different seeds with different soils (S3, S4) and red mud before giant reed plantation, and red mud after giant reed plantation to investigate the residual effect of giant reed on pure red mud.. For Radish and Carrot, the highest percent was recorded with S3, 76.7 and 56.7%, respectively. While, the lowest percent was found with red mud, 26.7 and 0.0%, respectively. Concerning Tobacco, the highest percent, 96.7%, was recorded with S4 and the lowest percent, 16.7%, was existed under red mud. After three months of giant reed cultivation, it was clear that the giant reed has induced the germination percent of all tested crops. However, the germination percent was increased from 26.7 to 46.7% (Radish), from 0.0 to 46.7% (Carrot) and from 16.7 to 60.0% (Tobacco). This data could clearly emphasize the ability of giant reed to reconstruct and decontaminate the polluted soils from different types of pollutants such as high pH, salinity and heavy metals (Fig. 1).

Table 4: Germination percent (%) of some vegetable crops in red mud and mud-polluted soil

Sample	Radish	Carrot	Tobacco
S3	76.7	56.7	93.3
S4	60.0	40.0	96.7
Red mud before giant reed plantation	26.7	0.0	16.7
Red mud after giant reed plantation	46.7	46.7	60.0

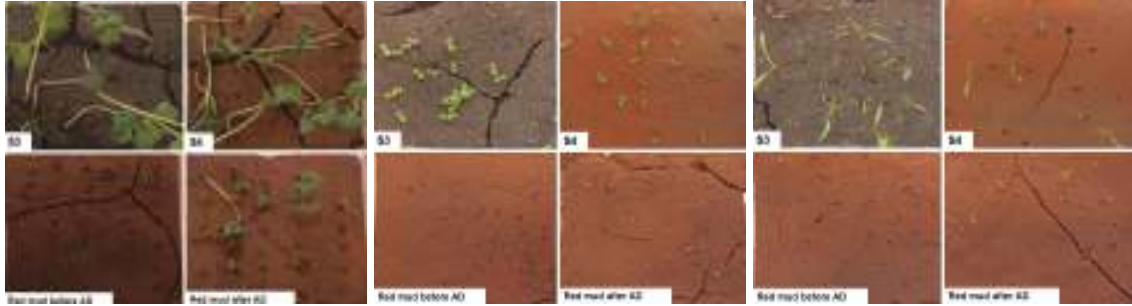


Fig. 1: Germinated seeds of radish (Left), tobacco (Middle) and carrot (Right) in different soils and red mud. (S3: Soil after maize, sun flower, and rapeseed rotation, S4: Mud-polluted soil and AD: giant reed)

3.2. Restoring soil ecosystem by giant reed under microbial communities-depleted soil

3.2.1. Soil properties

Planting giant reed affected some soil chemical and biochemical properties after 3 months (Table 5). For instance, soil pH increased by 5 - 9 % in all soils by the end of experiment, except for red mud where planting giant reed decreased pH by 1% (Table 5). Electrical conductivity (EC dS m^{-1}) for all soils was decreased by 24 – 82 % in non-autoclaved and autoclaved soils, respectively; the EC of red mud planted with giant reed was decreased by 63 %. Planting the test soils with giant reed had a variable effect on SOC after 3 months. For instance, without addition of inorganic fertilizers, SOC slightly decreased in most soils, e.g. by 11 % in soil S3, but SOC was increased in S1 and S4 (Fig. 2). With respect to available soil P, a sharp decrease was observed in all autoclaved and non-autoclaved soils. The amount of available potassium was increased more than 15 – 150 % in most soils relative to levels before experimentation, but there were no significant differences between treatments.

The presence of giant reed significantly increased the amount of available N in all soils (Table 5). It was observed that non-autoclaved samples had significantly higher

concentrations of available N than autoclaved samples. These results are in agreement with results from Alshaal et al. (2013), who reported that giant reed increased available nitrogen in red mud and mud-polluted soil. The effects of autoclaving on remineralization of labile inorganic P were clear, giant reed growth was much higher in autoclaved soils than non-autoclaved. In this study, P concentrations were between 1.3 - 2.6 % in shoots, and 0.7 - 1.7 % in roots produced on autoclaved soils. Plant available P is a critical growth determining factor, and it has been observed that the densest, tallest stands of giant reed growing along streams and drains in Egypt do so beside drains with high concentrations of soluble P. In addition, giant reed was one of the fast growing plants in fields that had been burned deliberately to control vegetation. These results are in agreement with results from Alshaal et al. (2013), who reported that giant reed increased available NPK in red mud sample and mud-polluted soil.

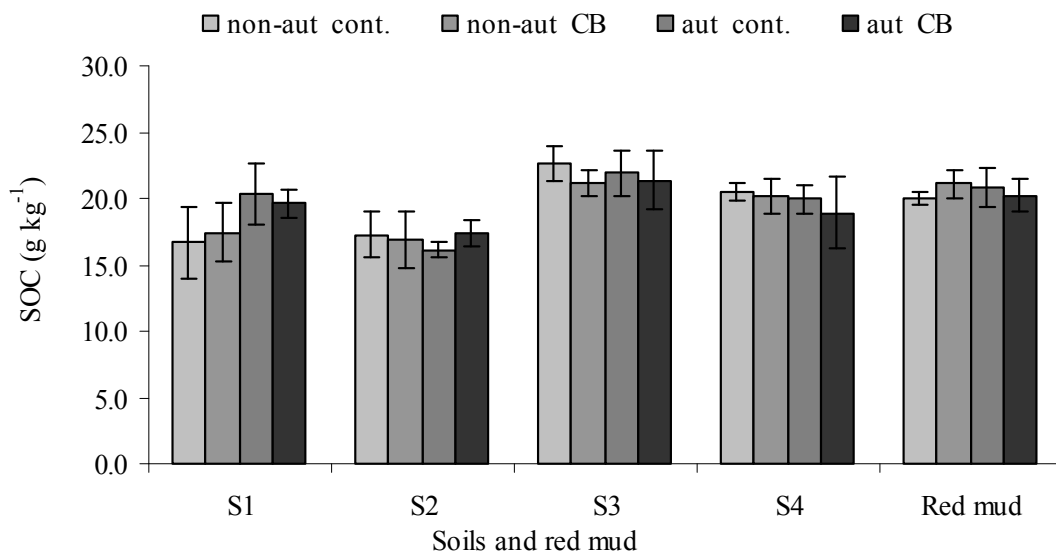


Fig 2: Soil organic carbon (SOC) content after giant reed grew in different soils and red mud with and without soil autoclaving. S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil, non-aut: non-autoclaved; aut: autoclaved; CB: commercial biofertilizer. *Vertical bars* represent the standard error (n=3)

Table 7: Soil chemical properties after giant reed cultivated in different soils treated by biofertilizer with and without autoclaving compared to before experiment

Soils	Treatment		pH*		EC** (dS m ⁻¹)		avail.N (mg kg ⁻¹)		avail.P (mg kg ⁻¹)		avail.K (mg kg ⁻¹)	
	non-aut	Aut	Non-aut	aut	non-aut	aut	non-aut	aut	non-aut	aut	non-aut	aut
S1	Control	8.52 ^{a,a,a}	(0.45) 0.34 ^{a,a,a}	0.43 ^{a,a,a}	(0.16) 12.2 ^{a,a,ab}	6.9 ^{a,a}	26.2 ^{a,a,c}	1630) 21.8 ^{a,a,d}	26.2 ^{a,a,c}	300 ^{a,a,a}		
	CB	8.25 ^{a,a,a}	8.49 ^{b,a,a}	0.34 ^{a,a,a}	0.40 ^{a,a}	16.7 ^{a,a,ab}	8.3 ^{a,a,a}	25.2 ^{a,a,c}	18.3 ^{a,b,b}	467 ^{a,a,a}		
S2	Control	(7.93) 8.34 ^{a,a,a}	8.67 ^{b,ab}	(0.50) 0.34 ^{a,a,a}	0.35 ^{a,a,a}	(0.15) 16.6 ^{a,ab}	8.5 ^{b,a,a}	17.9 ^{a,abc}	11.2 ^{b,ab}	500 ^{a,a,a}		
	CB	8.35 ^{a,a,a}	8.57 ^{b,b,b}	0.36 ^{b,a,a}	0.34 ^{a,a,a}	11.0 ^{a,a,a}	7.7 ^{a,a,a}	17.9 ^{a,abc}	13.8 ^{a,ab}	533 ^{a,a,a}		
S3	Control	(7.86) 8.33 ^{a,a,a}	8.54 ^{b,a,a}	(0.80) 0.35 ^{a,a,a}	0.40 ^{a,a,a}	(0.13) 10.8 ^{a,ab}	8.5 ^{b,a,a}	(170) 6.3 ^{a,a}	3.0 ^{a,a}	367 ^{a,a,a}		
	CB	8.30 ^{a,a,a}	8.57 ^{b,a,a}	0.32 ^{a,a,a}	0.37 ^{a,a,a}	13.3 ^{a,ab}	8.1 ^{a,a,a}	8.0 ^{a,a,a}	4.5 ^{a,a,a}	400 ^{a,a,a}		
S4	Control	(8.24) 9.03 ^{a,ab}	9.15 ^{b,ab}	(1.82) 0.88 ^{a,ab}	0.85 ^{a,ab}	(0.14) 17.9 ^{a,ab}	8.1 ^{b,a,a}	(130) 9.0 ^{a,ab}	5.6 ^{a,ab}	533 ^{a,a,a}		
	CB	8.97 ^{a,ab}	9.09 ^{b,ac}	0.83 ^{a,ab}	0.82 ^{a,ab}	18.9 ^{a,ab}	10.4 ^{b,a,a}	7.6 ^{a,a,a}	4.4 ^{a,a,a}	400 ^{a,a,a}		
Red mud	Control	(9.80) 9.70 ^{a,ac}	9.75 ^{b,ac}	(3.44) 1.27 ^{a,ac}	1.40 ^{b,ac}	(0.21) 16.8 ^{a,ab}	9.9 ^{a,a}	(520) 14.0 ^{abc}	10.5 ^{a,ab}	733 ^{a,a,a}		
	CB	9.70 ^{a,ac}	9.76 ^{a,ad}	1.39 ^{b,bc}	1.29 ^{b,bc}	18.2 ^{a,ab}	7.1 ^{b,a,a}	11.7 ^{a,abc}	10.7 ^{a,ab}	667 ^{a,a,a}		

Values in parentheses are data before the experiment, whereas the rest represents data after experimentation

* pH measured in soil:water suspension (1:2.5).

** Electrical conductivity (EC) measured in soil:water extract (1:5).

S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil. non-aut: non-autoclaved; aut: autoclaved; CB: commercial biofertilizer.

Different letters on the left show significant differences between the non-autoclaved and autoclaved treatment in case of the same soil type and control or the same soil type and commercial biofertilizer. Different letters in the centre show significant differences between the control and commercial biofertilizer treatment in case of the same soil type and non-autoclaved or the same soil type and autoclaved.

Different letters on the right show significant differences among soil types for the same treatment: non-autoclaved - autoclaved and control - commercial biofertilizer.

3.2.2. Autoclaving and soil enzymes activity

3.2.2.1. Soil enzymes activity

Soil enzymes are good indicators of soil quality because: a) they are closely related to organic matter, physical characteristics, microbial activity and biomass in the soil, b) provide early information about changes in soil quality, and are assessed more rapidly (Eldor, 2007)

3.2.2.1.1. Dehydrogenase activity

Overall, we detected high increasing in dehydrogenase activity for all soils and red mud compared to initial status before giant reed plantation (Fig. 3). Twelve weeks after planting of the giant reed was enough to significantly increase the activity of dehydrogenase in all soils and the red mud. Compared to the status of samples before the treatment, it is clear that giant reed has induced the intracellular enzyme (dehydrogenase) activity. In general, dehydrogenase activity has increased by 187 – 425 % in non-autoclaved soils compared to 262 – 705 % increases after autoclaving. Significant differences were found between autoclave and non-autoclaved treatments, where dehydrogenase possessed higher activity in autoclaved soils than non-autoclaved soils. Moreover, using commercial biofertilizer did not significantly affect dehydrogenase activity.

Dehydrogenase requires an intracellular environment (viable cells) to express its activity (Dick, 2000), the higher activity found in planted soils compared to unplanted controls indicates that giant reed has a unique microbial community which associates with its robust root system, where, after using sterile seedlings of giant reed and autoclaved samples, high numbers of bacteria and fungi were counted indicating that ability of giant reed to induce the growth of microbial community around its root system.

3.2.2.1.2. Urease activity

Urease activity is sensitive to autoclaving. A significant decrease in urease activity was found under autoclaved tested soils and red mud in comparison with non-autoclaved soils and red mud (Fig. 3). Planting giant reed in non-autoclaved soils significantly increased urease activity compared to the activity seen before giant reed plantation. Increasing of urease activity ranged between 195 - 591 and -35 – 46 % in non-autoclaved and autoclaved soils, respectively. Similarly, there were no significant

differences between control and biofertilizer in most soils and red mud with or without soil autoclaving. Urease has been widely used to evaluate changes on soil quality related to management. This enzyme is an extracellular enzyme representing up to 63% of total activity in soil. It has been show that its activity depends on microbial community, physical, and chemical properties of soil (Corstanje et al., 2007).

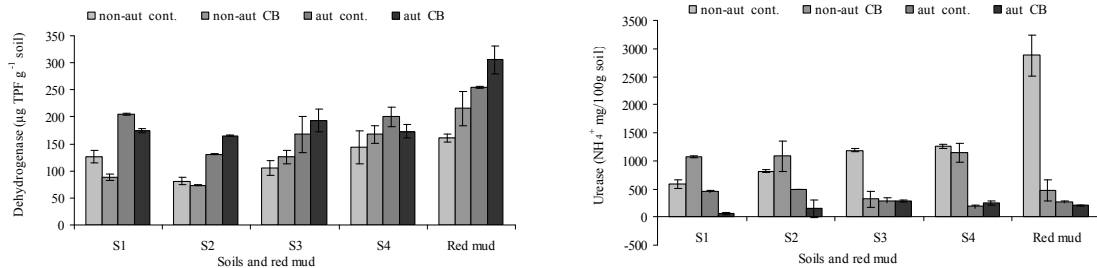


Fig 3: Soil dehydrogenase activity (Left) and urease activity (Right) in different soils and red mud cultivated by giant reed using commercial biofertilizer. S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil. non-aut: non-autoclaved; aut: autoclaved; CB: commercial biofertilizer. Vertical bars represent the standard error (n=3)

3.2.2.1.3. Alkaline phosphatase activity

Alkaline phosphatase activity decreased in all tested soils and red mud compared to that observed in soils before experimentation (Fig. 4). In autoclaved and non-autoclaved samples, biofertilizer did not affect alkaline phosphatase activity significantly. The reduction of phosphatase activity ranged between 0.3- 0.8 folds in both autoclaved and non-autoclaved studied soils. Phosphatases are a group of enzymes that catalyze hydrolysis of esters and anhydrides of phosphoric acid. Its activity, as extracellular enzymes, can be free in the soil water phase or stabilized in the humic fraction or clay soil content (Turner and Haygarth, 2005). Phosphatase activity in temperate grassland was investigated by Turner and Haygarth (2005), and they found a strong correlation between enzyme activity and soil properties such as pH, total N, organic P and clay content. The significant decreasing of alkaline phosphatase activity in tested soils maybe refers to shortage of available P concentration in the soils after experiment. These results emphasize that *A. donax* L. is a good candidate for marginal and wetland soil construction with soil health point of view.

3.2.2.1.4. Catalase activity

In general, the variation of catalase activity in all soils was similar to the variation in dehydrogenase activity. However, catalase activity increased in tested soils and red mud by 51 - 385 and 87 – 207 % in non-autoclaved and autoclaved tested soils

respectively, comparing with activity before giant reed plantation (Fig. 4). Autoclaving treatment significantly affected catalase activity in all soils except in case of red mud since higher activity for catalase was recorded in non-autoclaved red mud. On the other hand, biofertilizer has no significant effects on catalase activity in both autoclaved and non-autoclaved samples.

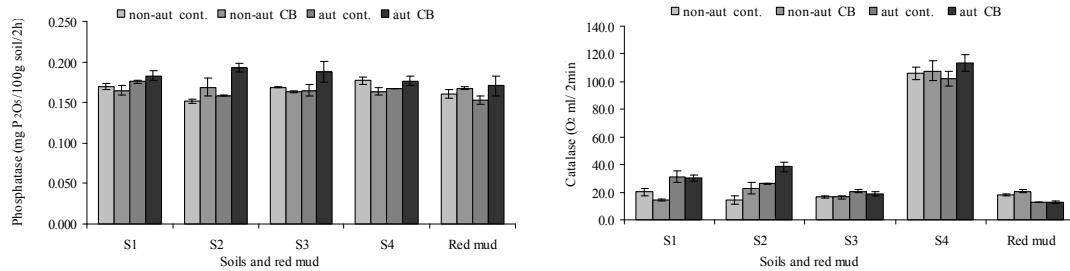


Fig 4: Soil phosphatase activity (Left) and catalase activity (Right) in different soils and red mud cultivated by giant reed using commercial biofertilizer. . S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil. non-aut: non-autoclaved; aut: autoclaved; CB: commercial biofertilizer. Vertical bars represent the standard error (n=3)

3.2.2.2. Microbial communities

3.2.2.2.1. Total bacterial count

Generally, total bacterial count decreased after planting giant reed, these variations ranged between 29 – 93 %. The highest reduction was recorded under mud-polluted soil (S4), as a result for unfavourable conditions after contamination with red mud (Fig. 5). An increasing in total bacterial count was found with soil S3 (maize, sun flower and rapeseed rotation). On the contrary, total microbial count was positively affected by soil autoclaving than non-autoclaved samples.

3.2.2.2.2. Total fungal count

Total fungal count was higher in autoclaved samples than in non-autoclaved samples (Fig. 5). In general, after giant reed experiment the total fungi number has increased in most soils. This increasing ranged between 45 and 136 %, except soil S1 (9-years grass) and soil S4 (mud-polluted soil). Similar, biofertilizer did not affect total fungi count in both autoclaved and non-autoclaved tested soils and red mud.

Microorganisms play a key role in nutrient cycling and energy flow. Microbial communities respond to environmental stress or ecosystem disturbance, affecting the availability of energetic compounds that support microbial population (Marinari et al.,

2007) Soil autoclaving did not negatively affect significantly the microbial communities (e.g. bacterial, fungal and actinomycetes counts) growth under giant reed, but autoclaved treatment recorded higher numbers than non-autoclaved treatment, maybe because autoclaving allowed a large number of bacteria in autoclaved soil to grow within a less diverse community. These findings maybe demonstrate that giant reed has special microbial community, so further studies about microbial communities that associate with giant reed root system are needed. Also biofertilizer addition did not affect the total bacterial count in significant way.

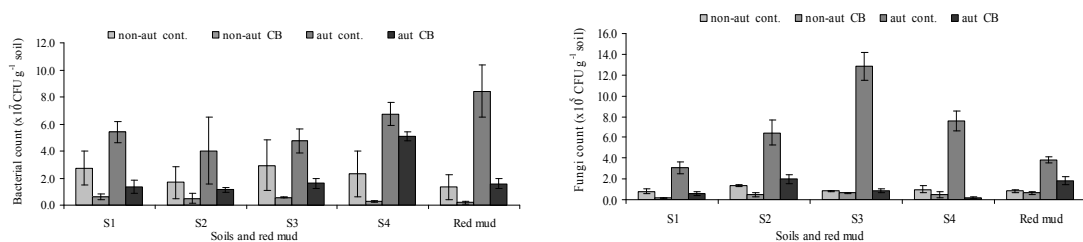


Fig. 5: Total bacterial (Left) and fungal (Right) counts after growing giant reed in different autoclaved soils and red mud under commercial biofertilizer. S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil. non-aut: non-autoclaved; aut: autoclaved; CB: commercial biofertilizer. Vertical bars represent the standard error (n=6)

3.2.3. Growth performance of *Arundo donax* L.

Giant reed plants in autoclaved soils possessed more robust root systems (short, very branched) than those in non-autoclaved soils (long, little branched), which means the former plants have a greater ability to uptake NPK and other micronutrients from the soils. The wet weight (Fig. 6) and length (Fig. 7) of shoots and roots were higher in autoclaved treatments than non-autoclaved. Giant reed stem weight and length were greater when grown in red mud-contaminated samples compared to the non-contaminated soils, with or without autoclaving. Macronutrient (NPK) concentrations were higher in the shoots than in the roots of giant reed (Figures 8, 9 and 10), although there were no differences between autoclaved and non-autoclaved treatments. However, the highest N concentrations in shoots and roots were 1.3 and 0.8 % under S4 (red mud-contaminated soils) and S2, respectively, whereas the highest concentrations for P were 3.4 and 2.5 % for shoots and roots under S2 and S4. On the other hand, the highest K concentrations in shoots and roots, 1.7 and 1.4% respectively, were recorded in S1.

Effects of heating and / or autoclaving on the soil properties have been widely documented (Neary et al., 1999; Ketterings et al., 2000). However, heating affects the nature of soil minerals and most probably their interactions with other plant nutrients. Soil temperatures in excess of 500 °C can be reached during fires, which will modify many hydroxylated soil minerals thereby changing their nature and nutrient retention properties (Anderson and Magdoff, 2005). Heat generated during fire not only induces chemical oxidation of soil organic matter by altering carbon and nitrogen transformations but also has potential effects on the soil microbial communities, since, higher temperatures than 50 °C are enough to kill the heat-sensitive microbes specially fungi, and temperatures higher than 70 °C can directly affect cover vegetation (Neary et al., 1999; Ketterings et al. 2000; Anderson and Magdoff, 2005). However, exposure soil to high temperature and pressure for long time has negative effects on soil ecology where almost enzyme activity is stop and microbial groups are going to disappear, but on the other hand, there are some positive effects such remineralisation of nutrients such as P. Anderson and Magdoff (2005) reported that autoclaving soil resulted in almost 60% more available P compared to non-autoclaved samples, with 78% more orthophosphate monoesters, 60% more orthophosphate diesters, and 54% more soluble inorganic P. Although at the same time other nutrients, like N and C, may be lost by volatilization. In this study, giant reed grew more vigorously in autoclaved soils and red mud than non-autoclaved soils and red mud. The most promising and encourage data was the higher biomass production of giant reed growing in autoclaved tested soils and especially in pure red mud. These findings demonstrate that under these conditions, high temperature and pollution by red mud (a caustic medium with high EC and trace metal content), giant reed could be effective solution to restore this soil soon and at same time produce significant biomass production. The root system architecture was entirely different and extraordinary between autoclaved and non-autoclaved soils. There were no clear explanations for why giant reed roots were short and denser in autoclaved treatment comparing with long and few roots in non-autoclaved. Anyway, the root architecture for giant reed still needs more investigations to get clear view for using giant reed as construct plant in marginal soils. This data encourages using giant reed as a good candidate to restore soil ecosystems after exposure to high temperatures for long time.

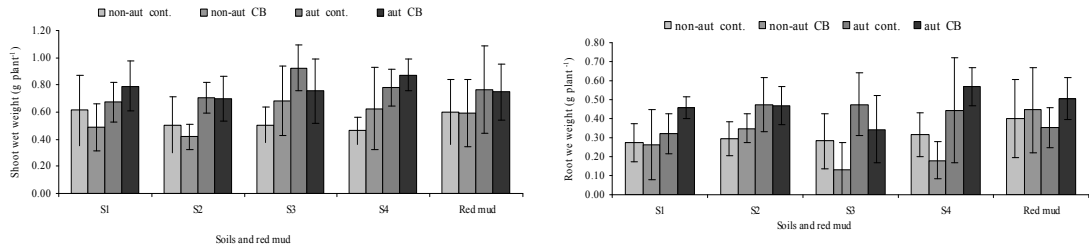


Fig. 6: Shoot and root wet weight of giant reed plants grew in different soils and red mud with and without soil autoclaving. S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil, non-aut: non-autoclaved; aut: autoclaved; CB: commercial biofertilizer. Vertical bars represent the standard error (n=8)

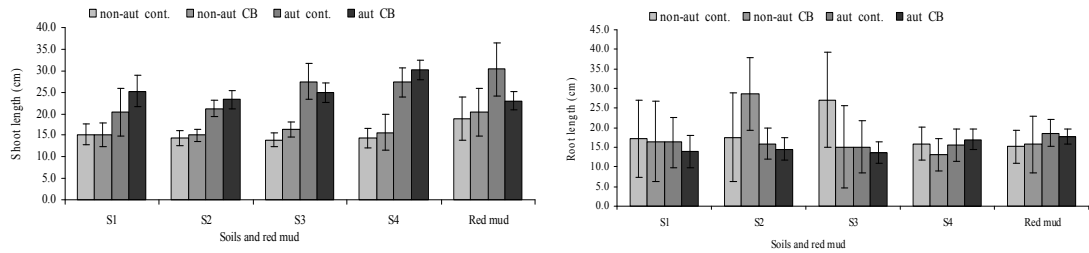


Fig. 7: Shoot and root length of giant reed plants grew in different soils and red mud with and without soil autoclaving. S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil, non-aut: non-autoclaved; aut: autoclaved; CB: commercial biofertilizer. Vertical bars represent the standard error (n=8)

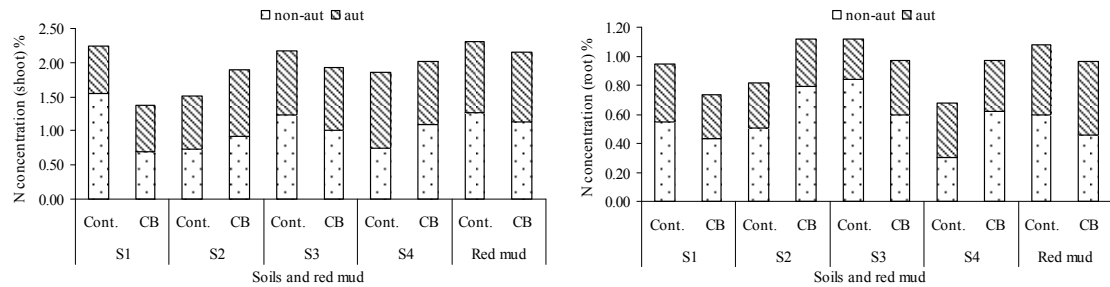


Fig. 8: Shoot and root nitrogen content of giant reed plants grew in different soils and red mud with and without soil autoclaving. S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil, non-aut: non-autoclaved; aut: autoclaved; CB: commercial biofertilizer

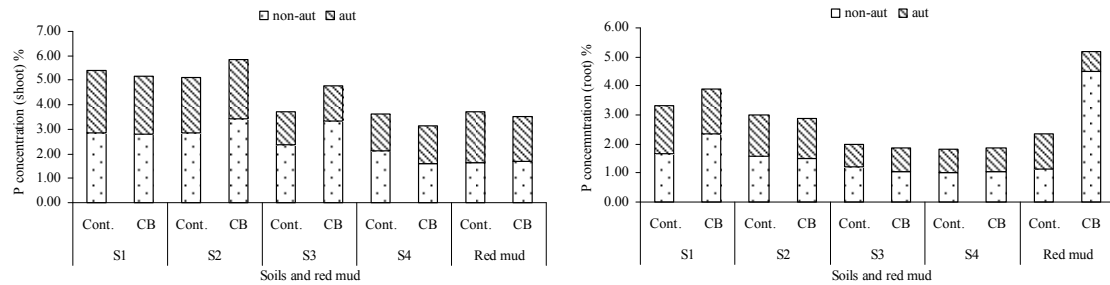


Fig. 9: Shoot and root phosphorous content of giant reed plants grew in different soils and red mud with and without soil autoclaving. S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil, non-aut: non-autoclaved; aut: autoclaved; CB: commercial biofertilizer

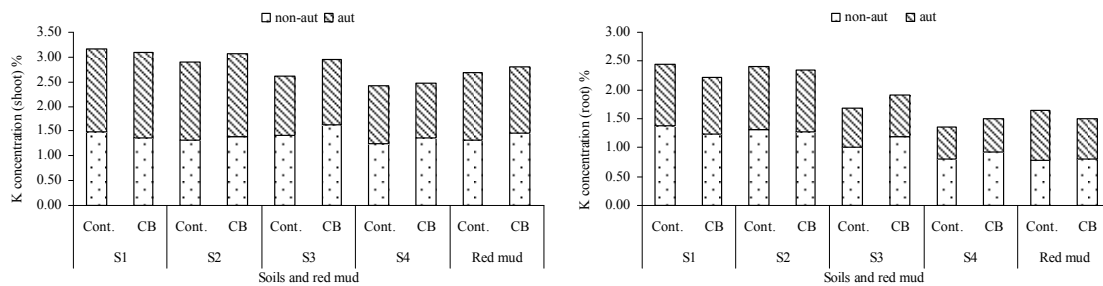


Fig. 10: Shoot and root potassium content of giant reed plants grew in different soils and red mud with and without soil autoclaving. S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil, non-aut: non-autoclaved; aut: autoclaved; CB: commercial biofertilizer

3.2.4. Restoring ecosystem of red mud-contaminated soil by *Arundo donax* L.

Recycling of bauxite derived-red mud becomes urgent where huge amounts of red mud are generated every year, since, 1 t of alumina generally results in the creation of 0.5–2.5 t of red mud. There is limited information on the use of red mud as soil amendment. However, red mud was used to reduce the availability of trace metals in different environments because of its high pH (Brunori et al., 2005). Moreover, leaching of phosphorus was decreased by adding red mud to soil as a result for the high retention capacity of fine particles in red mud and its high pH. Our study aimed to investigate the possibility using of red mud in wetland and marginal soils and the ability of giant reed to recover red mud-affected soil after exposure to high temperatures. The data showed that giant reed is able to improve soil quality after heating and after contamination with red mud. No wild fires were recorded in the soil sampling area in Hungary, but bauxite mining occurs in other parts of the world where wild fires do occur, and the results from this study suggest that in those areas giant red can be used to rehabilitate soil containing red mud (Figures 11, 12 and 13).



Fig. 11: Giant reed plants growing on different non-autoclaved soils and red mud with using *Arundo*' root extraction (Ar ex.) and commercial biofertilizer (CB) compared to control . (S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil, S5: Mixture of red mud and S3 soil by ratio 1:1 by weight)



Fig. 12: Giant reed plants growing on different autoclaved soils and red mud with using *Arundo*' root extraction (Ar ex.) and commercial biofertilizer (CB) compared to control . (S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil, S5: Mixture of red mud and S3 soil by ratio 1:1 by weight)



Fig. 13: Root structure of giant reed plants growing on different non-autoclaved and autoclaved soils and red mud. (S1: soil after grass; S2: soil after giant reed; S3: soil after maize, sun flower, and rapeseed rotation; S4: mud-polluted soil, S5: Mixture of red mud and S3 soil by ratio 1:1 by weight)

4. CONCLUSIONS AND RECOMMENDATIONS

4.1. Effects of red mud on soil and plant

- In spite of, high pH and salinity content of red mud, giant reed not only was able to survive in pure red mud sample, but also it can give a significant biomass production compared to non-contaminated soil with red mud.
- High organic carbon content as well as high total numbers for bacteria, fungi, actinomycetes, *Azotobacter sp* and *Azospirillum sp* were recorded in pure red mud maybe due to natural selection that could occur at Alumina ponds.
- As a consequence for adding red mud into soil, the nutrition status of this soil was improved, especially phosphorous and potassium contents.
- Before the giant reed plantation, Radish, Carrot and Tobacco seeds could not germinate on pure red mud.
- After three months giant reed cultivation on pure red mud. The germination percents of the previous seeds were increased, indicating that giant reed was able to reconstruct red mud and make it suitable for germinating different vegetable seeds.

From the information mentioned above, it can conclude that application of red mud has pros and cons. **So, pre-treatment for red mud to decrease its high pH and salt content could be helpful for using red mud as soil amendment.**

An environmentally friend and cost-effective technique to rehabilitate red mud is using giant reed plant.

5.2. Restoring ecosystem of heated soil

- Heating soil such as in natural fires (bushfires) almost kills all types of living organisms in soil as well as most of soil enzyme activities are disappeared.
- Giant reed showed high and fast potential to recover the microbial communities as well as soil enzyme activities in heated soils.
- For heated red mud-polluted soil, giant reed recorded same effects, where the microbial communities were fast recovered.
- Three months of giant reed cultivation were enough to reconstruct the heated soils.

- Giant reed showed potential growth and biomass production after soils autoclaving compared to non-autoclaved soils. Where, the root structure for giant reed plants in autoclaved samples was stronger and branched than in non-autoclaved samples giving ability for better growing.

From all of the above, we can recommend that using *Arundo donax* L. (giant reed) as phytoremediation plant for red mud-polluted soil as well as to reconstruct heated soils and environments.

5. NEW SCIENTIFIC RESULTS

- Using giant reed for decontaminate red mud-polluted environments due to its ability to translocate the metal species from roots to shoots.
- Possible use of red mud as soil amendment to decrease phosphorous leaching especially under excessive phosphorous fertilization and irrigation as well as to decrease the mobility of trace metals in soils, after pre-treatment for it.as well as enrich the soil nutrients especially phosphorous and potassium.
- Recovering heated soils by using giant reed where the experiments demonstrated robust and branched root system and dark green plants, so giant reed is appropriate for recovering heated soils.

6. SCIENTIFIC RESULTS UTILIZABLE IN THE PRACTICE

- Giant reed plants showed considerable effects to remediate the red mud contaminated soil where giant reed decreased pH, salinity content as well as the heavy metal contents for red mud sampl.
- The possibility of using giant reed plants in saline soil and Na-affected soil, since it showed high tolerance for high salinity content in soils.
- Giant reed is considered as good candidate for restoring the steaming soil, where it enhanced the microbial growth in microbial-depleted soil.
- Using giant reed as bioenergy crop, where it can produce significant biomass production especially in marginal soil where the using of food crops is not possible.

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