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Effect of Steel Slag to Improve Soil Quality of Tsunami-Impacted Land while Reducing the Risk of Heavy Metal Bioaccumulation

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Abstract

After the tsunami caused by the Great East Japan Earthquake, marine sediment was taken from the sea bottom and deposited over local agricultural fields. The marine sediment already contained an unknown amount of heavy metals, due to anthropogenic activities prior to the tsunami, which might affect plants, animals, and humans. Furthermore, soil salinity in tsunami-inundated land greatly increased. Three different amounts of steel slag were employed as pretreatment agent in order to improve agricultural soil quality. The soil samples treated with 2% of steel slag present a remarkable increase of *A. thaliana* biomass production with low BCF and TF values for most of the heavy metals. It was concluded that steel slag pretreatment used in the tsunami-inundated agricultural lands produced a noteworthy improvement in

soil quality which lead to a positive stimulative effect on plant growth, and the slag addition treatment proved to be a promising treatment that might be used for phytostabilization of slightly contaminated soils.

Keywords

Soil rectification

Steel slag

Tsunami sediment

Arabidopsis thaliana

Heavy metals

1. Introduction

The Great East Japan Earthquake, which is ranked in the top 5 major earthquakes ever recorded (USGS 2016), and the subsequent tsunami caused severe damage in the Tohoku region of Japan. Due to the intensity of the tsunami, large amounts of marine sediment were taken from the seabed and deposited over farmlands in the coastal area. It is known that sea sediment in Japanese bays can contain toxic compounds such as heavy metals due to past and current industrial activity (Kabir et al. 2006; Baba et al. 2012). In addition, the tsunami severely damaged various industrial facilities, which may have caused further contamination with hazardous materials in the affected areas (Bird and Grossman 2011).

Heavy metals are a group of metals with a density greater than 5.0 g/cm³. Elements such as arsenic (As), cadmium (Cd), chromium (Cr), cobalt (Co), copper (Cu), iron (Fe), lead (Pb), manganese (Mn), mercury (Hg), molybdenum (Mo), nickel (Ni), and zinc (Zn) are included in this classification (Sanita di Toppi and Gabbrielli 1999; McLaughlin 2006). Although some of them act as essential micronutrients for the development of living organisms, other heavy metals are toxic and can cause severe problems even at very low concentrations (Ali et al. 2013).

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It is important to note that because heavy metals are considered some of the most toxic inorganic pollutants in the world, they are recognized as a serious environmental issue. Four of them make the top-ten priority list of toxic pollutants issued by the Agency for Toxic Substances and Disease Registry

(ATSDR), and the United States Environmental Protection Agency (USEPA). As, Pb, Hg, and Cd rank in the first, second, third, and seventh place, respectively (ATSDR 2015).

When reviewing the literature related to worldwide large-scale tsunamis of the past (December 2004, Indian Ocean and March 2011, Japanese Tohoku Area), it can be seen that the majority of research is focused on the damage suffered to the coastline and urban infrastructure, (Ramanamurthy et al. 2005; Bird and Grossman 2011; Udo et al. 2012, Tanaka et al. 2012), rehabilitation of agriculture land (FAO 2005; Rachman and Agus 2005; Subagyono et al. 2005; Haque 2006; Tchiadje 2007) as well as the effect of sea water salinity on farmlands and the quality of their surface water and groundwater (Chandrasekharan et al. 2005, 2008; Kume et al. 2009; Raja et al. 2009; McLeod et al. 2010; Roy et al. 2014). On the other hand, information related to heavy metal pollution in tsunami-inundated areas and the subsequent handling of the contaminated soil, specifically in agricultural areas, is relatively scarce (Szcucinski et al. 2005; Ranjan et al. 2008; Srinivasalu et al. 2008; Tsuchiya et al. 2012).

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Therefore, the purpose of this study was to research the effect of using a pretreatment consisting of steel slag on tsunami-inundated soil samples in order to improve the agricultural soil quality. Also, the influence of the steel slag to ameliorate the effect of salinity in soil, and limit the uptake of heavy metals from contaminated sea sediment were studied. Within all the industrial wastes available for improving soil conditions, steel slag was selected due to its known capability to correct soil acidity, its potential use as an optional fertilizer and its cost-effectiveness (Wang and Cai 2006; Das et al. 2007).

During the experimental work, the model plant *Arabidopsis thaliana* was employed. Such a plant is often used in the agriculture and biotechnology research fields. The use of *A. thaliana* is advantageous because its growth stages and phenotypic characteristics are very well established, and any changes or modifications in these phases due to specific conditions can be easily identified.

2. Material and Methods

2.1. Soil

Soil was collected from agricultural areas near the central coast of Miyagi, Japan. Samples were oven dried at 65 °C for 48 h and then ground, weighted, and digested. This last process was performed in a microwave digestion system (Speed wave MWS–3⁺, Berghof) using 5 mL of aqua regia. Soil metal concentrations were determined by inductively coupled plasma mass spectroscopy (ICP–MS, Hewlett–Packard) with certified reference materials (Japanese Method based in EPA Method 6020A).

2.2. Steel Slag

During the experiment, a determinate amount of steel was added to the tsunami-impacted soil after the latter was washed with tap water. Originally, the steel slag had a particle size of 20–30 mm; however, it was crushed with a hammer, sieved and the particle size was adjusted to 2 mm or less. Table 1 shows the chemical composition of the steel used in the above-mentioned systems.

Table 1

Chemical composition of steel slag (weight %)

SiO ₂ (%)	CaO (%)	T-Fe (%)	Al ₂ O ₃ (%)	MnO (%)	MgO (%)	P ₂ O ₅ (%)	TiO ₂ (%)	S (%)	Others (%)
28.0	31.6	16.7	5.7	8.3	4.7	3.6	1.1	0.12	0.18

2.3. Plant Used in Experiment

For all the systems used in this experiment, *A. thaliana* Columbia species (Col – 0) plants were chosen due to their well-known phenotype and life cycle. Several growth variables, such as light, water supply, and nutrient addition were controlled in order to assure homogeneous plant growth in early stages and allow observing any changes on plant development when treatments were applied.

2.4. Experimental Design

A couple of *A. thaliana* seeds were planted in individual 1.5 in. cubes of Grodan Stonewool A–OK (hydroponic substrate matrix). Several cubes were placed in a growth chamber at a constant temperature of 20 °C, supplied with 24 h of light, and fed with a 1-M Hyponex® solution every 1–2 days according to the requirements of each plant. After germination, all 3-week-old plants showing

similar development were transplanted to individual pots filled with contaminated soil previously collected from tsunami-inundated zones.

Before transplant, two pretreatment processes consisting of soil washing (with tap water) and a single addition of basic oxygen furnace (BOF) slag were performed. The slag amount incorporated to each soil sample was about 2, 5, and 10% of their total soil dry weight. Five systems of soil samples; three washed and slagged samples, one tap–water washed, one raw sample, and the control (vermiculite) system were tested. One extra soil system, which consisted on non-tsunami impacted soil from a near location, was included for plant growth comparison. During the experiment, artificial light was provided in a 12-h cycle and the samples were kept in a temperature controlled chamber at 20 °C.

2.5. Plant Sample Preparation and Analysis

Twenty days after being transplanted to individual pots, plants were harvested and divided between roots, leaves, and stem (including seeds). Roots were extracted carefully and washed with deionized water. Fresh, dry biomass weight was determined, and all divided plant samples were dried in a constant temperature oven (Yamato DKN 402) at 65 °C for 24 h. The amount of biomass produced by most experiments was not enough to perform chemical analysis for each replicate. Therefore, in order to increase sample quantity, a group of mixed samples were prepared by combining the biomass and taking three replicates of each system. Chemical analysis was triplicated when sample amount allowed it.

Total heavy metal concentrations in plant tissue were determined by inductively coupled plasma mass spectroscopy (ICP–MS, Hewlett–Packard) with certified reference materials. Prior to the measurement, a subsample of each plant sample was taken, ground, weighted, and digested. This last process was performed in a microwave digestion system (Speed wave MWS–3⁺, Berghof), using 5 mL of HNO₃ before decomposition.

2.6. Statistical Analysis

Statistical analyses of the experimental data were performed using the R version 3.1.3 package on OS X Maverick. Evaluation of significant differences among the mean total content of heavy metals, nutrients in the soil, and some soil physicochemical parameters were performed using a one-way ANOVA followed by Tukey's post hoc test, with $p < 0.05$ indicating statistical differences. Also a principal component analysis (PCA) was carried out between soil samples.

3. Result and Discussion

3.1. Treatment Effect on Soil Samples

In Table 2, HM concentration in tsunami sediment was compared with the HM naturally abundant in Japan and the world, and also with Japanese environmental quality standards (EQS). As shown, the tsunami sample complied easily with EQS 1; however, it surpassed the acceptable level of As stated in the EQS 2. It is important to note that despite the compliance of Cd concentration, tsunami sediment should be considered a potential hazard due to its specific characteristics.

Table 2

Heavy metal concentration in soil and comparison with EQS in Japan

	As (mg/kg)	Cd (mg/kg)	Cu (mg/kg)	Pb (mg/kg)
Sediment	19.4 ± 1.1	0.31 ± 0.01	52.70 ± 1.2	37.9 ± 1.2
Japan agricultural soil average*	10.3 (a)	0.33 (b)	48.0 (b)	24.0 (b)
World–soil average* (c)	6.83	0.41	38.9	27.0
EQS 1 Japan	≤ 150.0	≤ 150.0	ND	≤ 150.0
EQS 2 Japan	< 15.0	< 0.4**	< 125.0	ND
*Background contents of HM in surface soil [redacted] ND: No Designation				
**Concentration in brown rice [redacted] a: Arao et al. (2011)				
EQS 1: Soil contamination countermeasures act [redacted] b: Takeda et al. (2004)				
EQS 2: Agricultural land soil pollution prevention act [redacted] c: Kabata-Pendias (2011)				

Table 3 contains a summary of basic physicochemical characteristics for the normal soil, control and tsunami sediment samples, including those washed with tap–water and slag treated. Table 2 also shows each treatment effect on pH, electrical conductivity (EC), sodium adsorption ratio (SAR) and redox potential (Eh). As shown, the raw soil samples had an extreme acidic pH (3.81) with EC (6.76 dS/m) and SAR (14.56 meq/l) values similar to those reported in others areas which had also been affected by large-scale tsunami disasters. Such values

are a consequence of seawater flooding and seabed sediment deposits on agricultural land (Subagyono et al. 2005; Chaudhary et al. 2006).

Table 3

Characteristics of raw soil and treated soil samples ($n = 3 \pm SD$)

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System	Treatment	pH	EC (dS/m)	SAR (meq/L) ^{1/2}	Eh mV
Control	No (vermiculite)	7.02 ± 0.17d	0.17 ± 0.01de	1.28 ± 0.21c	30
Normal soil	No (regular soil)	5.52 ± 0.08e	0.14 ± 0.01e	2.57 ± 0.15bc	78
Raw soil	No (tsunami affected soil)	3.81 ± 0.02f	6.76 ± 0.48a	14.56 ± 1.85a	288
Washed	Washed with tap water	3.87 ± 0.02f	2.24 ± 0.08b	5.29 ± 0.84b	350
Slag 2%	Washed + 2% slag	7.36 ± 0.07c	0.69 ± 0.06cd	4.10 ± 0.30bc	170
Slag 5%	Washed +5% slag	8.14 ± 0.02b	0.93 ± 0.09c	3.35 ± 0.00bc	147
Slag 10%	Washed +10% slag	8.69 ± 0.19a	0.99 ± 0.11c	2.66 ± 0.31bc	85

EC, electric conductivity; *SAR*, sodium adsorption ratio; *Eh*, redox potential, same letter means no statistical significant differences (95%)

Such soil conditions are not favorable for plant growth since salt tolerant plant species are only able to grow in salinity levels of 3.0–5.0 dS/m, and if SAR values rise above 12 to 15, serious physical soil problems arise and plants have difficulty absorbing water (Munshower 1994). These values are to be expected because after a tsunami floods agricultural land, seawater has a chance to infiltrate the soil profile and increase soil salinity, in effect deteriorating the soil structure due to high sodium content.

After the washing process, the raw sample pH levels were not improved significantly. On the other hand, a marked increase in soil pH levels was observed after the addition of steel slag, which also showed a significant difference among each slag treatment. Regarding EC, a decrease in level due to the tap–water washing and slag addition was evident, with a clear reduction of

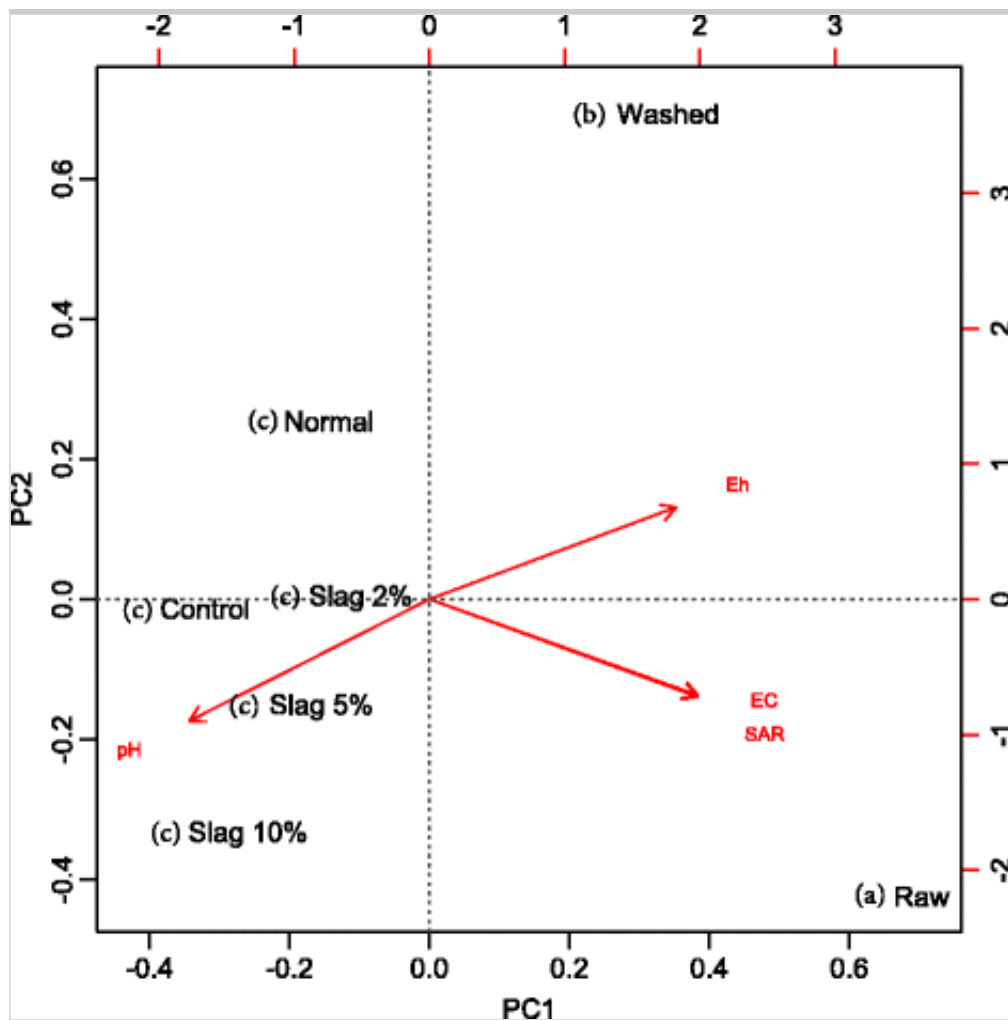
EC from 6.76 dS/m (raw sample) to 0.69 dS/m (Slag 2%). However, when the slag percentage was increase, the EC values only augmented slightly from 0.69 dS/m to 0.99 dS/m (Slag 10%). It should be noted that there were significant differences between normal soil, raw soil, washed soil and slag–treated samples; but between the slag samples there was no significant difference.

SAR levels in soil also were reduced, from $14.56 \text{ (meq/L)}^{1/2}$ to $2.66 \text{ (meq/L)}^{1/2}$ for raw and slag 10% samples, respectively. Only raw samples showed a significant difference compared with the rest of the samples. Finally, the Eh increased slightly after the washing process but when the steel slag was added, a decreasing trend was observable.

On all data content in Table 3, a principal component analysis (PCA) was performed in order to identify possible grouping of samples into sub–populations. As shown in Fig. 1, soil samples can be assembled into three group corresponding to: (a) raw, (b) washed soil, and (c) normal, control, slag 2%, slag 5%, and slag 10% systems. Analyzing Fig. 1, it is evident that groups (a) and (b) can be clearly distinguished from group (c). Group (a) showed the highest value of SAR and EC, meanwhile group (b) presented the highest value of Eh. Such data explains in part the opposite location of group (c) in the biplane compared with the other two groups. Group (c) had the highest values of pH, especially the slag 10% sample.

Fig. 1

Principal component analysis (PCA) for soil samples

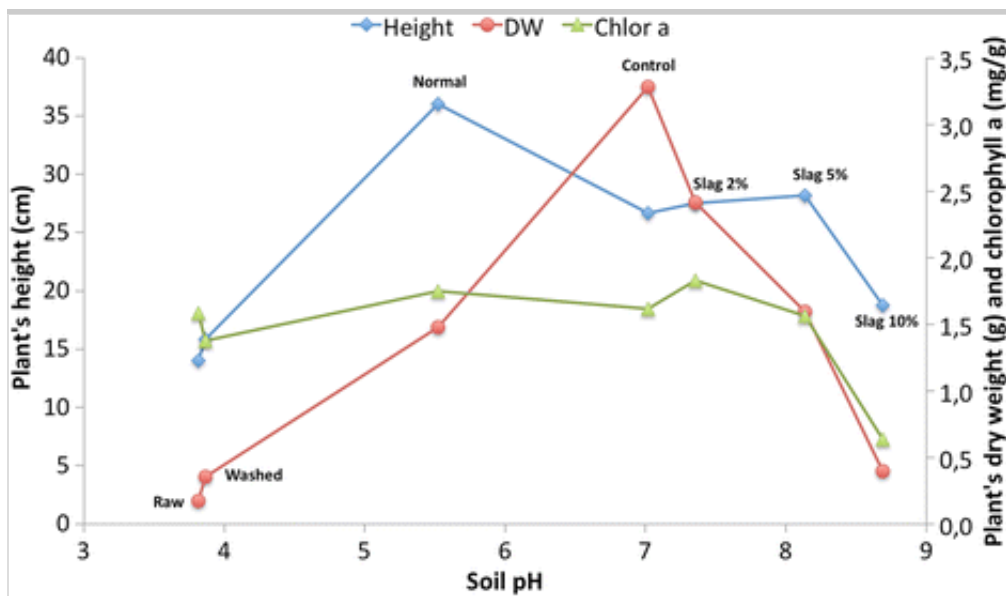


3.2. Plant Growth

Figure 2 shows the plant growth response (height, dry weight and chlorophyll a) to pH changes due to the treatments applied to the soil. The total biomass of the *A. thaliana* plants grown in the raw system presented an alarming decrease from 3.28 g–dry weight (DW) and 1.79 g–DW to 0.17 g–DW compared with the control samples (– 94.8%) and normal soil (– 88.4%) respectively. Such significant reduction of biomass remained similar, 0.35 g–DW (– 89.2%), when plants were grown in tap–water washed soil. This clearly showed that soil washing was not enough to ensure proper growth of plants and that further correction of soil was required.

Fig. 2

Relationship between soil pH and plant height, dry weight and chlorophyll a



After treatment by adding steel slag the soil acidity was rectified, the pH values significantly increased and plant biomass was enhanced. In the samples grown in soil treated with slag 2%, a total weight of 2.41 g–DW was reached which represented an increase of +1308% of biomass compared with the raw samples (0.17 g–DW). Even though an enhancement in biomass of 1.59 g–DW was obtained when steel slag treatment was increase to 5%, it was evident that a negative trend appeared under alkaline soil conditions (pH over 7.5). For slag 10% treatment, the declining trend went even further whit a weight of 0.40 g–DW shown.

Final height of the plant displayed a similar trend to the dry weight, where raw and washed systems had similar values, but steel slag systems presented a significant increase in the plant height compared with the normal and control system (with the exception of the slag 10% system). The raw and water-washed systems did not present a significant difference between each other (14.0 and 15.8 cm, respectively). On the other hand, the highest value within the treated systems, 28.2 cm, was obtained for the Slag 5% system.

Regarding chlorophyll a, it was not possible to distinguish a trend between systems. Apparently there is no significant difference between all the systems since their values (except Slag 10%) remained almost constant. As mentioned, the slag 10% samples showed a clear reduction in chlorophyll a, which could be related to the notable increase of soil pH (Fig. 2). After analyzing the relationship between the dry weight of plant, height of plant and each treatment, it should be noted that Slag 2% system presented the best growth performance for all systems and was the closest to the control and normal soil systems.

As previously shown in Table 3, the addition of steel slag increased soil pH from slight to moderate alkaline levels (range of 7.36–8.69). This effect, combined with an elevated Ca concentration, will trigger a deficiency in nutrients. Mn, Fe, and P (due to high pH); K and Mg (due to high Ca levels) will be relatively unavailable for plant uptake. This explains the dramatic biomass decrease for the steel slag 10% system (Fig. 2). Initially, the pH increase and most importantly, the increase in Ca concentration will assist development in the plant's roots. Due to 10% steel slag addition, soil pH reached basic levels and Mg/K concentration remained nearly constant; and in consequence, the development of the plant slowed.

As described by Boyes et al. (2001), altered environmental stress conditions may affect several plant characteristics, resulting in morphological changes in *A. thaliana* such as a decrease in height or reduction in leaf number. Table 4 shows a deficiency in some essential soil nutrients, which is not completely remedied by an increase in soil pH and the steel slag addition, per se. In addition, heavy metal uptake also triggers an extra stress condition, which could have an impact on plant development.

Table 4

Total content of some essential nutrients in soil samples ($n = 3 \pm SD$)

System	Na (g)	Mg (g)	K (g)	Ca (g)	Mn (g)	Fe (g)
Normal	0.79 ± 0.06c	3.09 ± 0.07d	3.76 ± 0.19b	3.91 ± 0.03c	0.25 ± 0.03b	24.0 ± 1.2d
Raw soil	6.50 ± 0.02a	6.95 ± 0.09b	4.01 ± 0.01ab	4.98 ± 0.19c	0.31 ± 0.01b	40.0 ± 0.6c
Washed	2.14 ± 0.38b	6.17 ± 0.05c	5.13 ± 0.12a	5.33 ± 0.49c	0.32 ± 0.22b	39.8 ± 0.4c
Slag 2%	1.89 ± 0.12b	6.30 ± 0.02c	5.01 ± 0.01ab	9.82 ± 0.35bc	0.69 ± 0.01b	43.0 ± 0.1bc
Slag 5%	1.72 ± 0.12b	6.83 ± 0.16b	4.68 ± 0.17ab	13.86 ± 3.21b	1.23 ± 0.45b	47.7 ± 1.2b
Slag 10%	1.66 ± 0.15b	8.35 ± 0.20a	4.73 ± 0.77ab	23.26 ± 1.73a	2.42 ± 0.46a	56.0 ± 2.4a

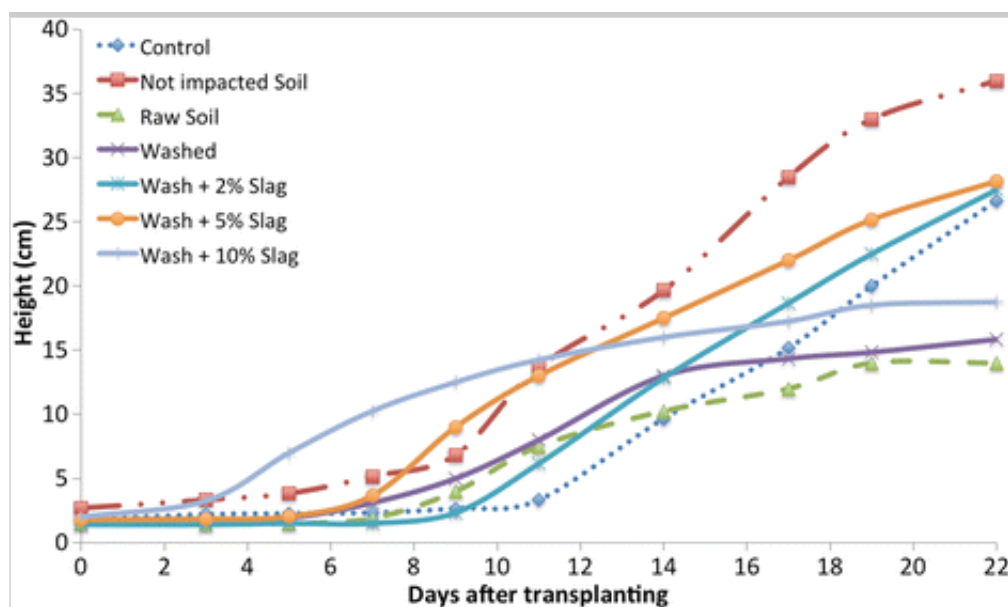
Same letter means no statistical significant differences (95%)

During normal *A. thaliana* growth stages, inflorescence and meristems development (stem), and the subsequent production of flowers and silique occurs between 30 and 40 days after seed germination. Such process takes place at a relatively slow but steady rate. A combined stress inflicted by a lack of nutrients and the uptake of heavy metals (such as As and Cd) threatens the survival of the plant. In order to complete its life cycle, the plant's best defensive response is to accelerate its growth (Keunen et al. 2011).

Figure 3 shows the height of *A. thaliana* for each system over the course of the experiment. All plants, except the control samples, began to develop their stems between 33 and 37 days after germination (3–7 days after transplantation). The control growth curve presented a steady development, however, 10–12 days after transplantation, an acceleration of plant growth was observed. The growth curve of plants grown in raw, water-washed, and the slag 2% and slag 5% systems, followed the initial development described above. However, 37 days after germination, growth performance started to change significantly and the growth curve was modified.

Fig. 3

Height response of *A. thaliana* grown in tsunami sediment treated with steel slag



Modification of the growth curve was reflected by accelerated plant growth, where the development of the stem and consequently, the plant's height, began to increase quickly. This phenomenon occurred for all systems, especially for the steel slag 10% system. Raw and washed systems showed signs of stunted

plant development and 19 days after transplanting, serious damage was perceptible. On the other hand, steel slag 2% and 5% systems exhibited an outstanding growth performance when compared with the raw, washed and slag 10% systems. Both systems equaled the control system's plant height values, which suggests that the addition of slag improved soil conditions.

3.3. Metal Content in Plant Tissue

After observing plant production improvement by steel slag addition, the analysis of heavy metal content in the plant tissue was in order, given that when a higher biomass production is shown, an increase in the uptake of heavy metals is expected. Table 5 presents a summary of the total content of heavy metals for each system in three different tissue types. It should be noted again that prior to chemical analysis, all plants sampled were divided into roots, rosette leaves and stems (including seeds).

Table 5

Total heavy metal content in plant tissue in each experiment system

System	Dry weight (g)	As (µg)	Cd (µg)	Cu (µg)	Pb (µg)
		Root			
Control		1.03	0.15	34.00	5.32
Raw soil*		–	–	–	–
Washed*		–	–	–	–
Wash + Slag 2%		19.44	0.32	119.86	29.48
Wash + Slag 5%		25.09	0.11	105.08	14.69
Wash + Slag 10%		4.30	0.03	24.63	5.39
		Leaves (rosette)			
Control		0.19	0.05	4.32	0.10
Raw soil		0.03	0.00	0.12	0.04
Washed		0.00	0.00	0.01	0.00
Wash + Slag 2%		2.11	0.18	10.71	2.14
Wash + Slag 5%		2.00	0.06	6.37	1.43
Wash + Slag 10%		0.35	0.01	1.42	0.27
		Stem			

Control		0.13	0.13	11.91	0.13
Raw soil*		–	–	–	–
Washed		0.20	0.01	0.78	0.01
Wash + Slag 2%		0.40	0.10	5.47	0.05
Wash + Slag 5%		0.64	0.04	2.65	0.04
Wash + Slag 10%		0.04	0.00	0.16	0.00
	Total				
Control	3.282	1.36	0.33	50.23	5.55
Raw soil	0.172	0.03	0.001	0.12	0.04
Washed	0.354	0.20	0.01	0.80	0.01
Wash + Slag 2%	2.414	21.95	0.60	136.03	31.67
Wash + Slag 5%	1.593	27.73	0.20	114.10	16.16
Wash + Slag 10%	0.397	4.70	0.05	26.21	5.65
*No detectable during analysis					

As, Cd, Cu and Pb distribution in the plant for the three steel slag systems, from highest to lowest content, were as follows: root >> rosette leaves > stem. It is noted that for As, Cu and Pb, most amounts were found in the root (more than 88%). It is clear that the roots accumulated the highest amount of these metals and therefore, transport to the aerial parts was limited. In the case of Cd, even though the same trend was apparent, the Cd content in the roots was about 53.2–64.9%. This reveals that these particular soil conditions favored the transport of this metal within the plant.

With respect to the control system, Cu and Pb distribution in *A. thaliana* was surprisingly different: root >> stem > rosette leaves. Cd also followed such a trend (root > stem > rosette leaves), however, the content in the root was not significantly higher when compare with the stem. As for As, metal distribution was as follows: root >> rosette leaves > stem. The low concentration of heavy metals in this system, as well as the neutral soil pH, should allow their transport from the root to the stem and seeds.

With regard to total heavy metal content found in plant tissue, the raw and washed systems presented the lowest values of all samples. This is partly

explained by the low biomass production, which complicated the quantification of such elements. However, it is noted that damage in the plants of both systems were observed, which suggested that metal uptake and the content found in plant tissue were reduced. As can be seen from Table 4, the steel slag 2% system had the highest total content for Cd, Cu and Pb in their tissues samples, while the highest As content found in plant tissue was obtained from the steel slag 5% system. This confirmed that the bump in biomass production also slightly increased the uptake of heavy metals by *A. thaliana*.

3.4. Soil Chemistry and Uptake Mechanism

Taking into account the physicochemical characteristics of the soil samples, where soil pH ranged from 4.05 to 8.69 with a redox potential between 85 and 350 mV, they indicated that all soil samples were under acid-reduced soil conditions or alkaline-reduced soil conditions depending on their respective pH values. After reviewing the four Pourbaix diagrams corresponding to each metal system (Hermann and Neumann-Mahlkau 1985; Brookins 1988), the chemical forms that may be present in the soil solution of all raw soil and washed systems are H_3AsO_3 (aq), Cd^{+2} (aq), Cu^{+2} (aq), and PbSO_4 . On the other hand, for the steel slag systems the probable existent chemical forms are Cd^{+2} (aq), Cu_2O (c), PbSO_4 and HAsO_4^{-2} (aq). Due to its high alkaline pH, steel slag 10% system might present different chemical forms for lead and cadmium such as PbCO_3 and CdCO_3 , respectively.

Chemical forms for the raw soil samples and the washed system were present in their cationic forms, especially Cd^{+2} and Cu^{+2} , which should allow easy uptake by the plant. However, the high salinity and SAR in the soil caused extensive damage to *A. thaliana*. Due to such harm, plants were not able to absorb metals, which might explain the low total content of metals in these systems.

Conversely, for the three systems where steel slag was added, the slight increase of metal extraction uptake was expected due to an improvement in biomass production, especially for the steel slag 2 and 5% systems. Nevertheless, the addition of steel slag increased soil pH, which would reduce all heavy metal mobility and prevent a larger uptake by plant roots.

Pb in soil primarily exists in the +2 oxidation state complexed with inorganic constituents (i.e., HCO_3^- , CO_3^{-2} , SO_4^{-2} and Cl^-) as we found by using the Pourbaix diagrams. Pb uptake by *A. thaliana* decreased conversely as soil pH increased by steel slag addition. Such trend could be explained by the alkalinity

of the soil, which may reduce the available content of heavy metals in the soil for uptake by plants (Teng et al. 2015). Also, Ca could inhibit Pb absorption in the plant roots; such phenomenon is associated with competition between these two cations for calcium channels (Huang and Cunningham 1996) and supports the notable decrease of Pb total content in the plants observed under the 5% and 10% steel slag systems.

The slight increase of As uptake in the 5% slag system might be explained by the reduction in Cd uptake and because $\text{HAsO}_4^{-2}(\text{aq})$ is chemically similar to phosphate, which in such conditions might share same transporters for uptake by plant (Meharg and Macnair 1990). Under simultaneous contamination of As and Cd in soil, it has been found that As in soil could promote Cd absorption in the plant. Therefore, in polluted sites where a moderate co-contamination of Cd and As exists, a synergetic effect appears to be apparent for Cd uptake (Zhou and Gao 1994; Sun et al. 2007).

The above statement supports the results obtained for the slag 2% system, but when Ca concentration in soil increased due to the addition of steel slag, as found in the slag 5% and 10% systems, the uptake of Cd by *A. thaliana* is limited since both elements are in antagonistic competition for uptake (Eller and Brix 2015). However, some researchers found that Cd becomes more mobile in alkaline soils (in our case, soil alkaline pH ranged from 8.0 to 9.0) due to the formation of complexes or metal chelates, which is contradictory to the well-known effect of low soil pH on Cd mobility (Kabata-Pendias 2011).

Both systems (2% and 5% slag systems) had a similar height response, however, there was a significant difference in the dry weight. Such a response could be explained by the combined effect of Cd and As pollution, but also by a probable lack of nutrients that might have occurred due to the increase in pH by steel slag addition. Finally, as expected by the low biomass production, the 10% slag system had a significant reduction in the concentration of heavy metals uptake by plant. This is explained by the high alkalinity in soil (pH = 8.69), which is not a suitable environment for plant growth and also reduces the mobility of heavy metals in the soil (Kabata-Pendias 2011).

3.5. Bioconcentration Factor and Translocation Factor

The bioconcentration factor (BCF) has been used to evaluate the ability of plants to accumulate heavy metals in the easily harvestable parts of the plants, such as leaves, stem and seeds (Zhang et al. 2002; McGrath and Zhao 2003;

Yanqun et al. 2005). On the other hand, the translocation factor (TF) of heavy metals indicates the efficiency of the plants to translocate those elements from roots to over ground plant parts (Baker and Whiting 2002; Mattina et al. 2003; Yoon et al. 2006; Niu et al. 2007; Shi and Cai 2009). The BCF and the TF were calculated as follows:

$$BCF = \frac{[HM]_{harvestable\ tissue}}{[HM]_{Soil}} \quad 1$$

Where $[HM]_{harvestable\ tissue}$ is the concentration of the target heavy metal in the harvested tissues of the plant (mg/kg), and $[HM]_{soil}$ is the concentration of the same metal in the soil (mg/kg).

$$TF = \frac{[HM]_{harvestable\ tissue}}{[HM]_{Root}} \quad 2$$

Where $[HM]_{harvestable\ tissue}$ is the concentration of the heavy metal in the harvestable plant parts (mg/kg), and $[HM]_{root}$ is the concentration of the same heavy metal in the plant roots (mg/kg). Both parameters have been used to quantify the efficiency of phytoextraction; they are two especially important and critical standards for screening and selecting hyperaccumulator plants for uptake heavy metals. Plant species with both a BCF and TF greater than 1 have the potential to be used for phytoextraction purposes (Baker and Brooks 1989; McGrath and Zhao 2003; Cluis 2004; Yoon et al. 2006; Liu et al. 2011).

In Table 6, the BCF and TF for the control system and each test systems are presented. As shown, the highest values for both parameters were obtained mostly from the steel slag 2% systems, where only the BCF for As, the TF for Cd, and the TF for Pb were slightly higher for the slag 5% system. This supports the results obtained by the heavy metals uptake test, where the slag system had a 2% higher biomass production, which allows a greater accumulation of metals. It is also evident that the BFC and TF values for the raw and washed systems were extremely low, mostly due to the marked deterioration suffered by the samples grown in the unfavorable soil conditions of both systems.

Table 6

Bioconcentration factor (BCF), translocation factor (TF) and metal extraction ratio (MER) of each experimental system

System	TF	BCF	MER (%)	TF	BCF	MER (%)
	As			Cd		
Control	0.31	0.41	2.15	1.22	7.73	17.57
Raw soil*	–	< 0.01	< 0.01	–	< 0.01	< 0.01
Washed*	–	0.01	0.01	–	0.02	0.02
Wash + Slag 2%	0.13	0.14	1.50	0.86	0.57	1.53
Wash + Slag 5%	0.11	0.15	1.91	0.88	0.19	0.52
Wash + Slag 10%	0.09	0.02	0.34	0.54	0.03	0.12
	Cu			Pb		
Control	0.48	2.10	8.11	0.04	0.06	1.85
Raw soil*	–	< 0.01	< 0.01	–	< 0.01	< 0.01
Washed*	–	0.01	0.02	–	< 0.01	< 0.01
Wash + Slag 2%	0.13	0.31	3.29	0.07	0.06	1.05
Wash + Slag 5%	0.09	0.16	2.59	0.10	0.04	0.55
Wash + Slag 10%	0.06	0.03	0.70	0.05	0.01	0.18
*Not detectable during analysis for root tissue of plants						

As described in a previous section, the As, Cu and Pb absorbed in all systems were mostly retained in the root, suggesting that soil conditions and slag addition limited the translocation of those heavy metals to aerial plant parts (leaves and stem). Such metal immobilization in root cells was emphasized by the extremely low TF values (less than 0.15) in each system, which implies an exclusion mechanism (Baker 1987).

In the case of Cd, translocation was surprisingly high, particularly for the steel slag 2% and 5% systems. Such a finding was interesting when taking into account *A. thaliana* is a model that is not distinguished for its use for phytoextraction purposes. However, when steel slag addition increased, the TF and BCF values also reduced, especially for the steel slag 10% system. This result also strengthens the fact that the increase of Ca concentration in soil is partly responsible for the decrease of Cd uptake by the plant. It is concluded that under moderate alkaline conditions, where the production of plant biomass is very low, the bioaccumulation and translocation of heavy metals is seriously

affected.

3.6. Metal Extraction Ratio

Metal extraction ratio (MER) was used to express the extraction capacity, taking into account the biomass produced and the amount of soil used to grow a plant,

$$MER = \frac{[HM]_{Plant} \times DW_{Plant}}{[HM]_{Soil} \times DW_{Soil}} \quad 3$$

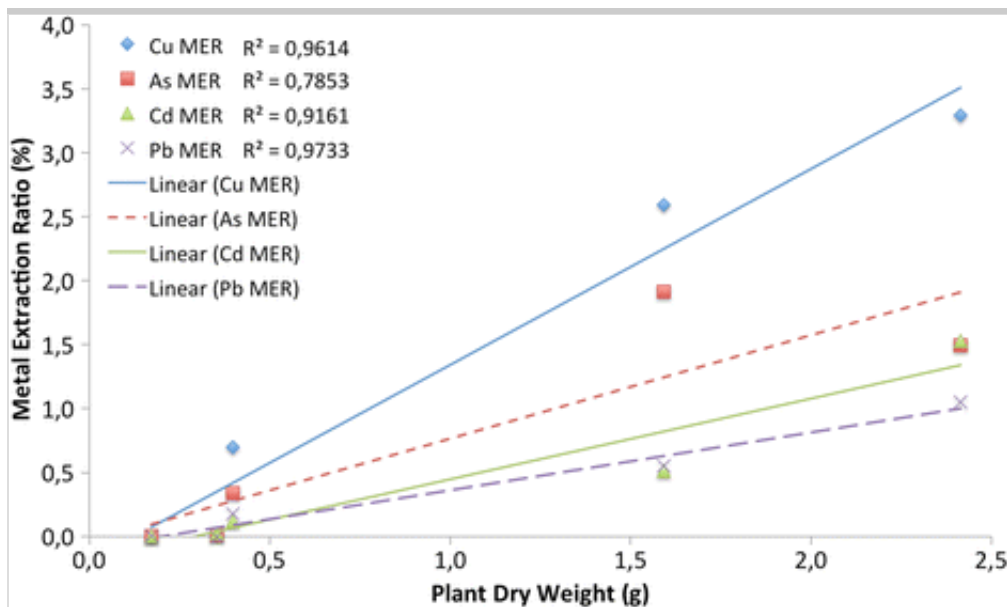
Where DW_{plant} is the mass of the harvested biomass (above ground) produced during experiment, $[HM]_{plant}$ is the metal concentration in the harvested plant tissue, $[HM]_{soil}$ is the metal concentration in the soil volume, and DW_{soil} is the rooted – soil volume influenced by the root of the plant species under study (Mertens et al. 2005; Rezvani and Zaefarian 2011; Andreazza et al. 2014).

Reviewing the calculated values of MER, it was found that the highest values for Cu, Cd and Pb were obtained for the steel slag 2% system. On the other hand, the best result for As was found in steel slag 5% system (Table 5). However, the same negative trend obtained for BCF and TF in each heavy metal was also found in the MER.

Figure 4 shows the relationship between the amount of biomass and the MER for each heavy metal. It is clear that the higher the production of biomass occurs, the greater the percentage of metal extraction ratio is obtained. Lineal regression was done and after doing a correlation between MER and dry weight, where the obtained R-square values were higher than 0.91 for Cd, Cu and Pb; but meanwhile the lowest R-square value was found for As ($R^2 = 0.7853$). The above result might suggest that the treatment worked better for Cd, Cu and Pb; but in the case of As, interference by the other heavy metals is suggested. Such interference or antagonism happens especially with Cd, a scenario that has been mentioned by other researchers (Liu et al. 2007; Sun et al. 2008, 2009). However when Cd availability was reduced (due to increase in Ca concentration) and coupled with a probable lack of nutrients (caused by an increase in soil pH), As uptake might be favored.

Fig. 4

Relationship between dry weight and metal extraction ratio (MER)



It is noted that a typical steel slag composition includes substantial amounts of minerals rich in Mn, Mg, Al, Si, and especially Ca and Fe (Shi 2004); which significantly increases soil pH and reduces the bioavailability of heavy metals. Soil pH is the key factor controlling the solubility and bioavailability of metals, and therefore, increasing the pH could cause the improvement in the adsorptive ability of most oxides for cations and this will promote the formation of less soluble metal precipitates such as silicates, phosphates and hydroxides (Kumpiene et al. 2007; Zhao and Masaihiko 2007; Lee et al. 2011; Gu et al. 2013).

It is clear that the incorporation of the steel slag into the tsunami-impacted soil improved soil conditions, which prompted an increase in the production of *A. thaliana* biomass. When this happened, the translocation and bioaccumulation of some metals such as cadmium improved markedly. These results allow us to infer that if a model plant like *A. thaliana* managed to improve BCF and TF values after this treatment; using a specific hyperaccumulator plant for a heavy metal (for example, *A. halleri* for Cd), it could improve the performance of phytoextraction in an area with relatively low contamination.

In summary, the use of steel slag combined with the selection of a specific and adequate hyperaccumulator plant would allow to refine the use of phytoremediation in lands impacted by a tsunami. This hybrid technique could be used in areas with a moderate level of metals or as a final polishing step to reduce the cost and impact of other methods.

4. Conclusions

The soil treated with steel slag 2% system showed the best growth performance and a good biomass production. Also, this system obtained the highest total concentration of Cd, Cu and Pb. On the other hand, the highest total concentration for As was obtained for the steel slag 5% system. Although the biomass production of the slag 2% system was similar to the control system and close to the normal system, the deficiency of key nutrients such as K and Mg, hampered a better biomass increase. From the observed improvement of the soil physicochemical characteristics and the low BCF and TF obtained values, it can be concluded that steel slag addition is an effective method to rectify soil conditions of agricultural land inundated by a tsunami, and also limited the uptake of heavy metals.

Although the performance of the *A. thaliana* plants was not comparable with heavy metal hyperaccumulators in regard to heavy metals uptake, obtained results suggest that steel slag treatment might be used for phytostabilization of a slightly contaminated soil in combination with a heavy metal-tolerant species with a high BCF and a low TF. However, the slag addition should be controlled to avoid alkalinity problems and to maintain soil conditions that allow a normal plant development.

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