Assessing the remediation of a contaminated soil ecosystem with some recent chemo biological technologies through bioremediation index

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Abstract---In a soil ecosystem irrigated with low-quality water out at El-Rahway village, Giza Governorate, Egypt, a completely randomized field trial was carried out to assess the suitability of some soil biochemical remediation technologies on minimizing the inorganic contaminants hazards, their bioavailability as well as the relationship between potential toxic elements (PTEs) distribution in remediated soil ecosystem, the bioavailability index (BI) and pollutants uptake by cultivated hyperaccumulated plants. Turnip Brassica rapa and radish Raphanus sativus were separately cultivated in the contaminated after being enriched with elemental sulfur (T1), elemental sulfur fortified with a mixture of Thiobacillus Thiooxidans and Thiobacillus ferroxidans (T2), rock phosphate impregnated with phosphate...
dissolving bacteria (PDB) (T3), bentonite inoculated with Pseudomonas fluorescence (T4), kaolinite associated with Pseudomonas fluorescence (T5), sulfur mixed with rock phosphate and inoculated with a mixture of Thiobacillus thiooxidans, Thiobacillus ferrooxidans and phosphate dissolving bacteria (PDB) (T6), a mixture of bentonite and kaolinite inoculated with Pseudomonas fluorescence (T7) and a combined mixture of bentonite, kaolinite and rock phosphate inoculated with Pseudomonas fluorescence and phosphate dissolving bacteria (PDB) (T8), as well as control treatments represented by cultivated untreated soil ecosystem. Results implied that the concentration of most contaminants was found in F6 (Residual form) in control cultivated, decreased, T1, T2, and increased in the treatments supplied with modified clay minerals. Results also emphasized that T8 is the best management practice in the remediation of targeted inorganic contaminants grown in a contaminated soil ecosystem cultivated with radish or turnip plants that showed the lowest values compared to other trailed treatments. The combination of phytoremediation and chemo-biological techniques was very effective in minimizing the hazards of PTEs bioavailability and the distribution of the inorganic contaminants, and BI was verified to be promising tools in evaluating soil remediation.

**Keywords**---soil pollution, PTEs bioavailability, bioremediation, chemical remediation, phytoremediation, PTEs distribution, chemical fractionation.

**Introduction**

Fashionable water scarcity has forced Egyptian farmers to irrigate their fields with low-quality water contaminated with potentially toxic elements (PTEs). The reuse of such water for prolonged periods intensified the content of many PTEs in the soil ecosystems. Bhattacharya et al. (2013) and Wuana and Okieimen (2011) stated that the existence of high concentrations of certain PTEs such as As, Cd, Cu, Pb, and Zn was frequently recognized in many soil ecosystems and led to significant adverse impacts on the soil ecosystem and grown crops as well. The main hazard of PTEs in a given soil ecosystem is their uptake by cultivated plants and emerge in the edible parts. Loganathan et al. (2018) and Olaniran et al. (2013) confirmed some adverse impacts on plants grown in contaminated soils, *i.e.*, reduction in leaf thickness, changes in cellular organization, slow plant growth, and decreased biomass. They added that irrigation with industrial effluent certainly reduces the germination percentage, root and shoot length, and fresh weight of seedlings.

Shehata et al. (2019) stated that the varied geochemical forms of PTEs existing in a given soil ecosystem influence their potential bioavailability; hence assessing the environmental impacts of PTEs in the soil ecosystems by their aqueous concentrations is incomplete. In other words, the bioavailability of inorganic contaminants in given soil ecosystems is not based explicitly on their readily available forms. Still, it might also depend on other forms like moderately or, in
some cases, hardly available forms (El-Kherbawy et al., 2008); however, aqueous concentrations of PTEs might help get information on the degree of their bioavailability in light texture soil ecosystems (Saber et al., 2016). Many other factors control the mobility, bioavailability, and environmental toxicity of PTEs, such as the type of specific chemical forms, pH in the rhizosphere, and/or release of radical exudates that might change the images of not readily available PTEs to available ones (Malachowska-Jutsz and Gnida, 2015; Qian et al., 1996). The distribution and bioavailability of PTEs also depend on some other factors related to sorbent and sorbate, like types of pollutant(s), soil characterization, organic matter content, redox potential conditions, and soil pH (Li et al., 2019; Shetaya et al., 2019; Pérez-Esteban et al., 2013). Despite the voluminous literature on the advantages of chemical, biological, and phytoremediation techniques in remediating inorganic contaminants, there is little focus on the effect of combined technologies (Mansour et al., 2019).

Through the distribution of PTEs in selected soil treated with remediation materials, Bioavailability index BI calculated and pollutants uptake by Radish and Turnip, the hyperaccumulation plants used, the current work aims to evaluate the selected index in declaring the efficiency of certain microorganisms combined with some chemical and natural amendments in minimizing hazards of PTEs and to select the best management practice to minimize the hazards of inorganic contaminants in contaminated soil ecosystems.

Materials and Methods

Soil sampling

A composite surface (0-30 cm) soil sample irrigated with low quality water for 80 year was sampled in three replicates from El-Rahawy village, Giza governorate. The collected samples were air-dried, ground into 2 mm average size and exposed the chemical fractionation, and the physicochemical properties (Table 1).

Table 1 Some physicochemical properties and PTE’s concentrations in soil ecosystem

<table>
<thead>
<tr>
<th>soil parameter</th>
<th>Surface (0-30)</th>
<th>Subsurface (30-60)</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH (1: 2.5)</td>
<td>7.83±0.01</td>
<td>7.92±0.03</td>
</tr>
<tr>
<td>ECe (dSm⁻¹)</td>
<td>2.40±1.63</td>
<td>2.96±2.50</td>
</tr>
<tr>
<td>OM (%)</td>
<td>1.50±0.01</td>
<td>0.60±0.04</td>
</tr>
<tr>
<td>CEC (meq/100g)</td>
<td>40.04±2.14</td>
<td>40.97±1.75</td>
</tr>
<tr>
<td>Clay content (%)</td>
<td>52.40±0.01</td>
<td>55.60±0.01</td>
</tr>
<tr>
<td>Soil Texture</td>
<td>Clay</td>
<td>Clay</td>
</tr>
<tr>
<td>Total Zn (mg kg⁻¹)</td>
<td>99.20±1.47</td>
<td>99.20±1.95</td>
</tr>
<tr>
<td>Total Cu (mg kg⁻¹)</td>
<td>33.23±1.00</td>
<td>30.63±0.50</td>
</tr>
<tr>
<td>Total Ni (mg kg⁻¹)</td>
<td>99.20±0.88</td>
<td>64.83±1.62</td>
</tr>
<tr>
<td>Total Cd (mg kg⁻¹)</td>
<td>13.30±0.55</td>
<td>11.03±0.60</td>
</tr>
</tbody>
</table>
Experimental

A completely randomized field experiment was carried out at El-Rahway site, Giza Governorate, Egypt (30° 11’ 33.6” N; 31° 02’ 38.4” E) in a contaminated soil irrigated for 80 years with low quality water composed of agricultural drainage water mixed with sewage and industrial effluents. Field pots with an area of 3×3 m² were treated for in three replications for each treatment, including:

- Uncultivated soil as control 1 (UC),
- Untreated cultivated soil as control 2 (C),
- Soil treated with elemental sulfur at the rate of 1.25 ton fed⁻¹ (T₁)
- Soil treated with elemental sulfur at the rate of 1.25 ton fed⁻¹ and inoculated with a mixture of *Thiobacillus thiooxidans*, *Thiobacillus ferrooxidans* at the rate of ten-liter of a microbial suspension (10⁶ CFU) (T₂)
- Soil treated with rock phosphate at the rate of 1.25 ton fed⁻¹ and inoculated with *phosphate dissolving bacteria* (PDB) at the rate of ten-liter fed⁻¹ of a microbial suspension (10⁶ CFU), (T₃),
- Soil treated with bentonite at the rate of 1.25 ton fed⁻¹ and inoculated with *Pseudomonas fluorescence* at the rate of ten-liter fed⁻¹ of a microbial suspension (10⁶ CFU), (T₄),
- Soil treated with kaolinite at the rate of 1.25 ton fed⁻¹ and inoculated with *Pseudomonas fluorescence* at the rate of 10 liter fed⁻¹ of a microbial suspension (10⁶ CFU), (T₅),
- Soil treated with elemental sulfur and rock phosphate each at the rate of 1.25 ton fed⁻¹ and inoculated with a mixture of *Thiobacillus thiooxidans*, *Thiobacillus ferrooxidans* and phosphate dissolving bacteria (PDB) at the rate of ten-liter fed⁻¹ of a microbial suspension (10⁶ CFU), (T₆),
- Soil treated with a combined mixture of bentonite and kaolinite at the rate of 1.25 ton fed⁻¹ for each type and inoculated with *pseudomonas fluorescens* at the rate of ten-liter fed⁻¹ of a microbial suspension (10⁶ CFU), (T₇),
- Soil treated with a combined mixture of bentonite, kaolinite and rock phosphate each at the rate of 1.25 ton fed⁻¹ and inoculated with *Pseudomonas fluorescence* and phosphate dissolving bacteria (PDB) at the rate of ten-liter fed⁻¹ of a microbial suspension (10⁶ CFU), (T₈).

Each plot was sown with radish (*Raphanus sativus*) or turnip (*Brassica rapa*) seeds as hyper accumulator plants. At the maturity stage of both radish and turnip plants were sampled in three replicates, shredded to roots and shoots, air-dried, oven dried at 70°C, ground and their PTEs contents were determined.

Microbiological analyses

*Pseudomonas fluorescence*, *Acidithiobacillus ferrooxidans* and *Bacillus megatherium var. phosphaticum: Phosphate dissolving bacteria* (PDB) were isolated and grown in nutrient media according to (Atlas, 2005). *Acidithiobacillus thiooxidans* was isolated and grown according to (Ryu et al., 2003). All trailed...
microbial suspensions were initially mixed with the soil before sowing either solely or in combination with other remediation amendments.

**Soil analyses**

- Soil characters, *i.e.*, (pH, electrical conductivity EC, organic matter OM, cation exchange capacity CEC and soil texture) were determined as given in (Sparks et al., 2020) while the plant analysis was done according to Paech and Tracey (2013) using A Perkin–Elmer flame atomic absorption spectrometer (FAAS).

**Plant analysis**

All treatments were replicated three times. After harvest, Radish and Turnip plants collected from the field experiments were dried at 70 °C to a constant weight, grounded to fine powder, and dried in a muffle furnace at 500°C for 6 hr. the ash was dissolved in a mixture of 2 M HCL and 1M HNO3 (Nanda Kumar et al., 1995). The concentrations of the heavy metals in the clear-acid extracts were measured using A Perkin–Elmer flame atomic absorption spectrometer (FAAS).

**Distribution analysis of PTEs**

Studied PTEs distribution in trailed soil was analyzed as mentioned by Ma and Rao (1997). One gram of each soil sample was weighted into a 40-ml polycarbonate centrifuge tube and the following fractionating items were estimated:

- **Water-soluble form (F1):** One gram soil sample extracted with 15 ml of de-ionized water for 2 h.
- **Exchangeable form (F2):** The residue from water-soluble fraction is extracted with 8 ml of 1M magnesium chloride MgCl₂ (pH 7.0) for 1h.
- **Carbonate form (F3):** The residue from exchangeable fraction is extracted with 8 ml of 1 M sodium acetate NaOAc (adjusted to pH 5.0 with acetic acid HOAc) for 5h.
- **Fe-Mn oxides form (F4):** The residue from carbonate fraction is extracted with 0.04 M hydroxyl amin hydrochloride NH₂OH.HCL in 25% (v/v) acetic acid HOAc at 96° C with occasional agitation for 6h.
- **Organic form (F5):** The residue from Fe-Mn oxide fraction is extracted with 3 ml of 0.02 M nitric acid HNO₃ and 5 ml of 30% hydrogen peroxide H₂O₂ (adjusted to pH 2 with nitric acid HNO₃). The mixture is heated to 85°C for 2h, with occasional agitation. A second 3 ml of 30% H₂O₂ (pH 2 with HNO₃) is added and the mixture heated again to 85°C for 3h with intermittent agitation. After cooling, 5 ml of 3.2 M ammonium acetate NH₄OAc in 20% (v/v) HNO₃ is added and the samples diluted to 20 ml and agitated continuously for 30 min.
- **Residual form (F6):** The residues from organic fraction are digested using HF-HCl/HNO₃.

**Phytoremediation:** Two types of water ferns, *i.e.*, Turnip (*Brassica rapa*) and Radish (*Raphanus sativus*) were separately used to phytoremediation contaminated soil samples.

**Bioavailability index (BI) Determination**
BI index for different treatments applied was calculated by using the model:

\[\text{BI} = \frac{(F1+F2+F3)}{(F1+F2+F3+F4+F5+F6)} \times 100\]

Where \(F\) represents the fraction type as mentioned in distribution study. Decreasing the value of BI indication to the succession of remediation material(s) to minimizing the hazards of contaminants studied.

**Statistical analysis**

Regression’s analysis, discriminate analysis and the fitting of curves to the data obtained were performed using separate two-way ANOVAs. The data of biomass PTEs were analyzed by discriminate analysis. Statistical analysis aimed to examine the succession of the applied remediation amendments in returning the studied contaminated soil ecosystem to its normal settings. SAS software was used to evaluate the different obtained results *(SAS Institute, 1996)*.

**Results and Discussions**

**Bioavailability of PTEs in remediated soil ecosystems as affected by trailed remediation amendments used**

Results given in Table (2) confirmed that both radish and turnip plants were efficient in the uptake of PTEs from the contaminated soil with preferability of Radish compared to Turnip. In addition, in the untreated cultivated soil, all pollutants except for arsenic (As) accumulated by the turnip plant were concentrated in the root part. For example, the zinc concentration in radish sprouts was 33.6 ppm, while in roots it was 55.2 ppm. The corresponding values were 8.3 and 9 for copper, 22.5 and 28.6 for nickel; 31.4 and 79.6 for Cr. Although the same trend was observed in radish, arsenic again showed a reverse trend by having numerical values 127 and 103 in shoot and root respectively.
Table 2 PTEs concentrations in radish and turnip plants grown in contaminated soil as affected by various treatments (mg kg⁻¹)

<table>
<thead>
<tr>
<th>Plant sample</th>
<th>Radish (C)</th>
<th>Turnip (C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shoot</td>
<td>Zn: 33.6±1.8 Cu: 8.3±0.1 Ni: 22.5±0.4 Cd: 193.9±2.9</td>
<td>Zn: 18.5±1.3 Cu: 2.2±0.2 Ni: 2.2±0.2 Cd: 18.4±0.9</td>
</tr>
<tr>
<td>Root</td>
<td>Zn: 55.2±1.5 Cu: 9.0±0.2 Ni: 28.6±0.8 Cd: 219.0±2.0</td>
<td>Zn: 58.4±0.8 Cu: 4.3±0.2 Ni: 4.3±0.2 Cd: 38.6±0.8</td>
</tr>
<tr>
<td>T1</td>
<td>Shoot: 36.1±0.8 Root: 57.6±1.4</td>
<td>Shoot: 22.9±1.2 Root: 62.9±1.2</td>
</tr>
<tr>
<td>T2</td>
<td>Shoot: 38.9±0.5 Root: 62.9±0.7</td>
<td>Shoot: 25.1±0.6 Root: 65.1±0.6</td>
</tr>
<tr>
<td>T3</td>
<td>Shoot: 15.5±0.5 Root: 35.3±0.7</td>
<td>Shoot: 16.8±0.8 Root: 38.7±1.4</td>
</tr>
<tr>
<td>T4</td>
<td>Shoot: 12.3±1.2 Root: 35.3±0.7</td>
<td>Shoot: 11.6±1.4 Root: 38.7±1.4</td>
</tr>
<tr>
<td>T5</td>
<td>Shoot: 18.8±0.4 Root: 35.3±0.7</td>
<td>Shoot: 28.2±0.8 Root: 61.0±1.5</td>
</tr>
<tr>
<td>T6</td>
<td>Shoot: 9.2±0.7 Root: 34.9±0.5</td>
<td>Shoot: 16.2±1.0 Root: 31.4±1.4</td>
</tr>
<tr>
<td>T7</td>
<td>Shoot: 3.4±1.1 Root: 20.3±0.3</td>
<td>Shoot: 18.3±1.2 Root: 40.8±2.0</td>
</tr>
<tr>
<td>T8</td>
<td>Shoot: 1.0±0.5 Root: 8.7±0.9</td>
<td>Shoot: 10.4±0.9 Root: 28.7±2.2</td>
</tr>
</tbody>
</table>

Starting from T3 to T8, remediation techniques applied to contaminated soil based on stabilization the rest of inorganic contaminants didn’t uptake by hyperaccumulated plants cultivated in contaminated soil ecosystem. Phosphate chemistry could be applied in developing as environmentally friendly remediation techniques for PTEs clean-up and valorization of contaminated soil ecosystems (Andrunik et al., 2020; Bolan et al., 2014; Nzhio et al., 2020). This result again indicated that kaolinite had high specification in retaining Ni compared to bentonite.

Remediation with elemental sulfur incorporated with rock phosphate and inoculated with mixture of Thiobacillus thiooxidans, Thiobacillus ferrooxidans and phosphate dissolving bacteria (PDB) (T6) applied in contaminated soil showed a significant decrease in concentrations of all PTEs compared to T4 and T5 in cultivated plants. Results indicated that the uptake of As in radish was 126.9 and 157.3 ppm in shoots and roots of radish cultivated in soil remediated with T4, the corresponding values were 42.6 and 123.1 ppm by remediation with T6, although the same trend was found in turnip, the capacity of radish to absorb As and other
PTEs from the soil ecosystem was higher than that of turnip. The double effect of both modified natural clay minerals with microorganisms and phosphate added to polluted soils enhanced contaminants fixation in soil which decrease the uptake by growing hyper accumulated plants. Kaolinite and bentonite clay minerals were observed to be more powerful for the removal of Fe, Zn, Ni and Cr from wastewater (Ahmed et al., 2021; Jock et al., 2021; Hussain and Ali, 2021; Mustapha et al., 2020; Szewczuk-Karpisz et al., 2020), Mansour et al., 2019a-e, Hu et al., 2019, Abdalla et al, 2019, Jiandong, et al, 2019, Mansour and Pibars 2019, Pibars and Mansour, 2019.

As shown in Table (2), results emphasized that treatments T7 and T8 were the highest in decreasing PTEs uptake by hyperaccumulated plants. In other words, application of bentonite, kaolinite, and to a less extent rock P (T6 and T7) or even a mixture of all remediation amendments (T8) retained most of PTEs in the soil ecosystem and their reserves were translocate to the hyper accumulated plants.

**Inorganic contaminants status in remediated soils through the distribution study**

Before discussing the significance of PTEs fractionation in remediated soil in more details, it should be mentioned that the time of using wastewater in irrigation of the studied soils is more than 80 years. This residence time get the opportunities of PTEs to accumulate in the soil through main three categories i.e. readily available form (fraction 1 and 2), moderately available form (fraction 3 and 4) and hardly available form (fraction 5 and 6) Ma and Rao (1997).

Existing guidelines or regulations for the assessment of PTEs contamination of soil ecosystems are based on the total soil PTEs concentrations. Yet, it is generally recognized that total concentrations don’t necessarily provide good information on the potential bioavailability or mobility of PTEs in soil ecosystems (Mansour et al., 2019; Yang et al., 2018). Sequential fractionation techniques are being used increasingly to provide more useful assessments of soil contamination status before and after remediation (Wang et al., 2021; Farrag, 2020; Saleem et al., 2018; Soliman et al., 2018). In addition, fractionation of inorganic contaminants is a fairly used technique to understand the mechanism(s) of PTEs distribution in soil ecosystems and to help assess bioavailability of trace metals in soil ecosystems.
Fig. 1 PTEs distribution in contaminated soil as affected by various bio-chemical treatments cultivated with radish plant
Fig. 2 PTEs distribution in contaminated soil as affected by various bio-chemical treatments cultivated with turnip plant.
Hardly available forms

Residual fraction RF

The distribution of Zn represented in table 1 showed that the residual fraction gave the highest value compared to other fractions (Figures 1 and 2), residual fraction values were 53 and 51% of total Zn found in untreated soil UC cultivated with radish and turnip, while the non-residual forms were 47 and 79% respectively. Except sulfur treatment T1 and T2, the % of residual form were increased and reached the maximum value in T8 regardless the type of hyper accumulated plant cultivated. In radish, the RF values increased from 51% in UC to 66% in T3 and reached 79% in T8. The corresponding values of Turnip were 53, 70 and 86 % respectively. The residual form of Cu and Ni gave almost the same trend, however, RF values of Cr were higher than Zn and reached to 84 and 96% of total Cr in the soils cultivated with Radish and Turnip and treated with T8, increased to 85 and 99% in As pollutant for the same plants and treatment applied in contaminated soil.

Organic fraction OF

Organic fraction as mentioned before is the most serious fraction influences human health. The ranges of this fraction were 0.34 and 30% for Turnip and from 0.07 and 30% for all contaminants found in contaminated soils and cultivated with Radish. The highest values was observed in UT and decreased till reached T8 except treatments contain individual S (T1 and T2) treatments for both cultivated plants used. The clearer example could be seen in Zn pollutant. Results showed that the percentage of Zn bond to organic matter in UT treatment cultivated with Turnip was 30% decreased to 14% in T4 and 8% in T8, the same values were 30, 18 and 12% in Radish. It should be mention that the same trend was observed in other contaminants studied.

Moderately available form

Moderately available form represents about from 1-26% of total percentage of contaminants distribution varied according to type of pollutant studied, hyperaccumulated plant and treatments applied.

Fe-Mn oxide fraction

Results showed that Fe-Mn oxide fraction ranged between about 0.02 to 15% and from 1-30% in Turnip and Radish respectively, these variations mainly due to remediation treatment applied. The highest values were observed in UC, CC, T1 and T2, meanwhile, the low values were mainly observed in treatments contain modified clay minerals. In treated soil cultivated with radish as an example, Ni distribution in Fe-Mn oxide fraction value was about 23% in UC treatment, decreased to 12.5% in T4 which contains bentonite and to 6.5% in T8 which having both bentonite, kaolinite beside PR. The same trend was also observed in the contaminated soil cultivated with Turnip previously amended with the same treatments.
Carbonate bound contaminants fraction

The percentage of carbonate-bound fraction for contaminants values was less than the Fe-Mn oxide fraction for all contaminants in both plants. In addition, the values of Turnip were higher than radish, the variations are only observed according to treatments applied. The percentage values were ranged between 0.5 and 4% and 4-11% for Radish and Turnip in all treatments applied. In Radish cultivated plants, results showed that carbonated pound Zn was 4.4% in UC treatment, this value decreased to 1.8% in T8 and all other treatments have modified clay mineral(s), the corresponding values for Turnip were 11.5 and 4.7%. In treatments contain S with a mixture of *Thiobacillus thiooxidans*, *Thiobacillus ferrooxidans* T2, results showed increase of this fraction could be mainly due to decreasing in soil pH. In Cr, for example, in Radish cultivated in remediated soil the numerical value of Cu percentage bound to carbonate bound Cu was 4.41% for UC treatment, application of T2 increased this value to about 7%. The same trend was also observed in other plants and contaminants studied.

Readily available forms

These forms i.e. water soluble and exchangeable fractions represent less than 4% for all treatments applied. In most cases in the treatments contain modified clay minerals, the sum of both fractions reached 0, in other words all available forms were sorbed on modified clay minerals and become not available for both cultivated plants like T7 or T8 for As in Radish or Ni and Cu in Turnip remediated soils.

Bioavailability Index (BI) as an indicator of management of PTEs remediation

The bioavailability index (BI) is defined as the proportion of reduction in a plant’s accumulation of inorganic contaminants, caused by the removal of the available fraction(s) of the hazardous contaminants found in soil ecosystem. The ratios of PTEs concentrations in plants to total their concentrations in the soil ecosystem have been used as a bioavailability index of PTEs in soil ecosystems (Wang et al., 2021; Saleem et al., 2018).

The effects of T8 (Soil treated with a combined mixture of bentonite, kaolinite and rock phosphate each and inoculated with *Pseudomonas fluorescense* and phosphate dissolving bacteria (PDB)) was shown through the having the lowest value in BI for all contaminants studied, this treatment was the best treatment in minimizing the uptake of PTEs by both grown hyper accumulated plants. The numerical BI value of Cu uptake in cultivated control soil ecosystem was 5.84, T8 decreased this value to 1.85 under radiash. In some cases, results showed that some contaminants completely sorbed in soil by having zero value bioavailability index in both cultivated plants. This result may explain decreasing the concentrations of all contaminants uptake by plants like Zn in the soil cultivated with Radish, the uptake of Zn was decreased from 89 to 10 ppm in control and T8 treatment.
Fig. 3 Bio availability index of contaminants in treated soil as affected by various bio-chemical treatments cultivated with turnip plants.

Fig. 4 Bio-availability index of contaminants in treated soil as affected by various bio-chemical treatments cultivated with turnip plant
The application of clay or modified clay minerals proved to be friendly and promising technique for environmental ecosystem in remediation of contaminated soil (Wahba and Zaghloul, 2007). Gained results having these types of amendments were the most treatments significantly decrease PTE’s availability in contaminated soil. In the studied contaminant, Arsenic and cadmium had the lowest numerical bioavailability index values in untreated soil ecosystems cultivated with either radish or turnip plants; such values reached 0.70 and 1.96 for As; 0.85 and 0.46 for Cd, respectively which means that As and Cd had the lowest risk or ability to move to biotas.


The geochemical behavior of PTEs indicates that application of PDB enhanced phosphate release at sufficient amounts which significantly reduced the availability index of PTEs (Mansour et al., 2019). Both rock phosphate and hydroxyapatite have been used as the primary P sources in the current study and both effectively reduced PTEs solubility. Thus, phosphate minerals have the potential to immobilize PTEs in contaminated soil ecosystems (Andrunik et al., 2020).

**Conclusions**

Potential toxic elements contamination of ecosystem is a major environmental concern. Bioavailability index was taken in this study to express the best manage practices showed be applied in sol polluted with inorganic contaminants. In this work, several remediation technologies have been implemented represented by addition of biochemical amendments and phytoremediation Turnip and Radish plants to minimize the hazards of Ni, Cu, Cr, As, Cd and Zn. Through the sequential extraction procedure and BI, results imply the combination of phytoremediation and chemo-biological techniques represented in soil treated with a combined mixture of bentonite, kaolinite and rock phosphate and
inoculated with *Pseudomonas fluorescence* and phosphate dissolving bacteria (PDB), (T8), was the best treatment in minimizing the hazards of PTEs according to their chemical distribution and bioavailability index. Under our experimental conditions, radish cultivated in contaminated soil treated with T8 and for less extent turnip cultivated in the contaminated soil amended with the same treatment, both showed priority in remediation of El-Rahway alluvial contaminated soil and are highly recommended to be applied to cleanup polluted soil ecosystems.

**Declarations**

**Ethics approval and consent to participate**
Not applicable.

**Consent for publication**
Not applicable.

**Availability of data and materials**

The data used for this study was obtained from determining the heavy metals and making a spot light on the chemical distribution and bioavailability index as indicators of phytoremediation studies in soil samples collected from El-Rahawy site (Giza Governorate), Egypt, which varied in the source of soil contaminants. The data are available from the corresponding author upon reasonable request.

**Competing interests**
The authors declare that they have no competing interests.

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**Authors Contributions**

HM helped in planting experiments, collecting the soil and plant samples from the site, determining PTE contaminants in soils, making the chemical fractionation to the soil ecosystems and writing the manuscript. AZ helped in finding the relationship between the fates of contaminants in relation to soil properties, discussing the obtained results and revising the manuscript. MS helped in clearing the objective of the study, writing the manuscript and revising the whole manuscript after it was written by Hesham. All authors contributed equally in the all-article steps, writing the manuscript, and approved the final manuscript.

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