Accumulation and Translocation of Heavy Metals in Soil and *Amaranthus Cruentus*

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**ABSTRACT**

A biogeochemical field study was carried out in the outskirts of Mumbai, where agricultural soils were contaminated by potentially toxic elements. *Amaranthus cruens* is an important part of the diet in the state of Maharashtra and thus it was selected as a test plant. Environmental pollution is one of the severe problems that are faced by Mumbai and agriculture is also affected due to it. Analysis of lead and cadmium, two of the most hazardous heavy metals was done from the soil samples and its accumulation patterns in the test plant were studied. Lead and cadmium analysis was done using ICP-AAS to determine if these vegetable samples are fit for human consumption.

**Keywords**


**Introduction**

Environmental pollution is the result of industrialization and urbanization. Environmental pollution is a broad spectrum terminology used for any kind of substance of any process that may cause harm or have toxic effect on the environment and the eco-system. Even though these processes have a positive effect on the daily life of individuals but its deep impact on the environment cannot be neglected.

Metals are the natural components of the soil. Some of these metals are micronutrients and are required for plant growth, such as Zn, Cu, Mn, Ni and Co, while others have unknown biological functions such as Cd, Pb and Hg (Gaur and Adholeya, 2004).

Heavy metal may interrupt normal functioning of the soil ecosystem which in turn causes negative effects on the growth of plants. The plants are dependent on the soil for their mineral nutrition as well as water. Heavy metal accumulation in soil due to various factors causes the plants to absorb water contaminated with these metals with the other vital minerals. These heavy metals do not have any specific role in the plant metabolism and thus start getting accumulated in the plants.

A biogeochemical research was done in areas located in the outskirts of Mumbai viz. Dahisar, Mira Road, Vasai and Nallasopara. The sampling sites are classic examples of areas that have contaminated soil due to the lack of proper water supply and overuse of chemical fertilizers by the cultivators have increased the level of heavy metals in the soil which can be easily absorbed by the test plant.

This project work has been done for analyzing the soil and the accumulation of heavy metals in the test plant i.e. *Amaranthus cruens*. Heavy metals can cause phytotoxic effects (Prasad and Hagemeyer, 1999) and affect human health harmfully (Reilly, 1991). Thus, this research work is a synthesis between ecotoxicology and toxicology. On the one hand, it analyzes the potential risks posed by heavy metals to the environment; on the other hand, it examines the effect on human health.

The present study focused on the content of potential toxic elements (PTE’s) in the soil and plant samples collected from the sampling sites. The main objectives of the research were:

To study the heavy metal contamination in the soil.
To evaluate if the PTE’s from the soil are transferred to the plants.
To study the accumulation pattern of heavy metals in the plants.
To determine the pollution index and to evaluate if these plants are fit for human consumption.

**Materials and Methods**

**Sampling Sites**

The study area was the northern suburbs of the city of Mumbai, India. Mumbai is the said to be the economic capital of India and has a major role in the Indian economy but like any other metropolitan city it faces several environmental problems. These problems are due to increasing population, vehicular pollution, industrial expansion, urbanization and lack of environmental policies.

The railways are one of the best modes of transport in the city and everyday millions of people use railways. Mumbai Suburban Railway consists of exclusive inner suburban railway lines augmented by commuter rail on main lines serving outlying suburbs to serve the Mumbai Metropolitan Region. Spread over 465 kilometers (289 mi), the suburban railway operates 2,342 train services. These railway lines are been used for cultivation of vegetables for many years now. The sampling sites were specific i.e. biased sampling was performed. The test plant i.e. *Amaranthus cruens* were taken from the railway tracks of Dahisar, Mira Road and Nalla Sopara. The climatic and environmental factors in all these locations were same but the method of cultivation used...
by the cultivators differed. A sample of plant was also collected from a farm in Vasai Road which used traditional methods of cultivation. The sample that was taken from all these regions was a fully grown plant of *Amaranthus cruentus* and soil. The sample was authenticated and identified by the experts from botany department of the college.

The plant from the 3 regions near the railway tracks served as samples whereas the plant from Vasai village served as a control. For the reference values the permissible limits of lead and cadmium in plants as suggested by the Ministry of Health and Welfare in 2011 was used. (Food safety and standards (Contaminants, toxins and residues) Regulations, 2011).

**Sampling Strategy**

Top soil samples (0-15cm) from all the four regions were collected during the month of May-June just before monsoon. The soil was collected for physio-chemical analysis and to analyze the concentration of heavy metals in it. *Amaranthus cruentus* was also collected during the same period. The selection of samples from these sites for soil as well as plant was random or unbiased. The pH of soil was check with pH meter.

**Sample Preparation**

Top soil samples (0-15cm) from all the four regions were collected during the month of May-June just before monsoon. The soil was collected for physio-chemical analysis and to analyze the concentration of heavy metals in it. *Amaranthus cruentus* was also collected during the same period. The selection of samples from these sites for soil as well as plant was random or unbiased. The pH of soil was check with pH meter.

**Laboratory Treatment**

The soil collected from different areas were labeled, dried at 40°C for 7-8 days and then sieved through a 2mm sieve. The plant samples that were collected were washed thoroughly first using tap water and then twice using deionized water to remove the soil and dust on it. The plant was then divided into 4 parts viz. root, stem, leaf and inflorescence. These parts were separately labeled and kept for drying at 50°C until they were completely dry. After drying, the samples were mechanically grinded using motor and pestle to particles which were <0.5mm in diameter.

**Sample Preparation**

Physiochemical Properties of Soil

The dried and sieved soil samples were taken and 5 grams of the soil was weighed accurately and 25 ml of deionized water was added to it. This solution was mechanically stirred for 30 min and then filtered. The filtrate was used as the sample for analysis of pH.

**Metal Analysis from Soil Samples**

The sample preparation for metal analysis was done as follows:-

- 0.5 gram of the dried and sieved sample was treated with 10 ml of 1:1 HCl.
- The sample was evaporated to dryness and the residue was cooled.
- To this residue, 5ml of 3:1 HNO₃ : HCl was added and the solution was heated till near dryness.
- The residue was then dissolved in 25ml 1:4 HCl and heated for 15 minutes.
- The solution was filtered into a 100 ml volumetric flask and the volume was made up to 100ml.

Soil samples preparation and the dried plant samples treated in the same manner were labeled properly and sent for analysis of lead and cadmium to Manisha Analytical Lab Pvt. Ltd, Kandivali, India using ICP-AAS.

The protocol followed by the laboratory as mentioned on the final analysis report was as follows:-

- Sample was treated with 5ml of ultrapure nitric acid.
- Digestion was carried out by heating the solution and was continued till brown fumes stopped evolving. Once the fumes ceased evolving, the sample was added to 50 ml of distilled water.

**Data Analysis**

**Transfer Factor**

Transfer factor was calculated to determine heavy metals accumulation in the edible parts of *Amaranthus cruentus* according to Zu. et. al., (2004).

\[ TF = \frac{\text{Concentration of metal in stem and leaf}}{\text{Concentration of metal in root}} \]

**Bioaccumulation Factor**

This factor (also termed Bioaccumulation Factor) is “the ratio of the concentration of a contaminant in an organism to the concentration in the ambient environment at steady state, where the organism can take in the contaminant through ingestion” (US EPA, 2011). This factor gives an estimation of the relative availability of heavy metals in soil and the plant’s ability to uptake a particular metal.

\[ BCFo-av [\%] = \frac{HMo [mg/kg, DW]}{HMav [mg/kg, DW]} \times 100 \]

Where,

- HMo : heavy metal concentration in the plant organ [mg/kg, DW]
- HMav : heavy metal concentration in the available fraction of soil [mg/kg, DW]
- o: root, stem, leaf, inflorescence

**Pollution Index**

The combined effect of heavy metals was considered by calculating the Ionic Impulsion according to Romero et. al., (1987) and the subsequent Soil Pollution Index (Plsoil) and Plant Pollution Index (Plplt) according to Romero et.al. (1989) these factors consider that the mixture and combination of HMs in soil may have an influence on the plants uptake (Hang et. al., 2010; Romero et. al., 1987, 1989).

\[ I_{I_{Ix}} = \sum (C_{m})^{1/n_{m}} \]

The soil and plant pollution in reference to unpolluted and toxic levels was calculated according to Romero et. al., (1987).

\[ PI = I_{I_{Ix}} \cdot I_{Iref} \]

where,

- \( I_{I_{Ix}} \) : Ionic Impulsion
- \( I_{Iref} \) : Ionic Impulsion of the Reference site
- \( I_{Itox} \) : Ionic Impulsion for toxic levels

**Human Hazard Factors**

The intake of contaminated food is the main exposure route to heavy metals for human beings (Peralta-Vide et. al., 2009) and could cause harm to health. Therefore, the non-cancer health risk, deriving from heavy metal intake via food, was estimated by calculating the non-cancer Hazard Quotient (ncHQ) (US EPA, 2000). The ncHQ assumes that even for sensitive receptors (young children, unborn babies) harmful health effects are unlikely to expect if the estimated exposure rate (here the Chronic Daily Intake (CDI)) does not exceed the oral Reference Dose (RfDo). The latter being a “toxicity value for evaluating non-carcinogenic effects from exposures” (US EPA,1989).

**Non-cancer Hazard Quotient**

The ncHQ was calculated separately for children and adults with the following equations:
nCHQ_{max} = CDI_{h} RfD_{h} / RfDo_{h}

Where,

- CDI – Daily Intake
- RfD_{h} – Oral reference dose
- RfDo_{h} – Oral reference dose

\[
\text{CDI}_{h} = \text{CF} \times \text{IR} \times \text{EF} \times \text{ED}_{h}
\]

\[
\text{BW}_{h} \times \text{AT}
\]

An ingestion rate of 17 g, given for leafy by Tripathi et al., (1997), was used. The RfDos are 0.001 mg/kg*d for Cd the IRIS information system. ARfDo for Pb is still in discussion [IRIS, 2011], so the concentration of 0.004 mg/kg*d (calculated from the tolerable daily intake limit recommended by the FAO/WHO found in Huang et al., (2008) was used.

### Non-cancer Hazard Index

The nCHI was calculated to explore the additive effects by more than one heavy metal. It was calculated by summing up the nCHQs of individual elements:

\[
\text{nCHI} = \sum n\text{CHQs}
\]

1. Metal Analysis Of Soil Sample

Soil is the sites from which the heavy metals get entry into the plants the results of ICP-AAS of soil samples are tabulated.

Highest concentration of lead was found in the soil sample from Mira Road and the least concentration was detected in the soil samples from Vasai. The soil sample from Mira Road has more concentration of lead than the permissible limit as defined by Chahal et al.,(2014).

All the samples were found to be contaminated with lead and exceeded the permissible limit as defined by Chahal et al.,(2014). Highest concentration of Cadmium was detected in Nallasopara and least concentration in the sample from Vasai.

### Table 2. Concentration of lead and cadmium in the soil samples.

<table>
<thead>
<tr>
<th>Sampling site</th>
<th>Concentration of lead (in ppm)</th>
<th>Concentration of cadmium (in ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NALLASOPARA</td>
<td>46.184</td>
<td>1.918</td>
</tr>
<tr>
<td>VASAI</td>
<td>8.924</td>
<td>1.421</td>
</tr>
<tr>
<td>MIRA ROAD</td>
<td>194.457</td>
<td>1.623</td>
</tr>
<tr>
<td>DAHISAR</td>
<td>26.626</td>
<td>1.621</td>
</tr>
</tbody>
</table>

2. Metal Analysis From Plant Sample

The results obtained from the analysis of the digested plant samples, indicates high accumulation of metals i.e. Lead and Cadmium in all the parts of the plants. The accumulation pattern of metals was not similar and showed varying concentrations in all the parts of the plant. The concentration of heavy metals in the edible parts of the plant was far more than the permissible limit as per Joint FAO/WHO food standards program codex committee on contamination of food 2011.

Transfer factor was one of the key aspect which helped in determining the ratio of contaminant in the root to the contaminant in the edible parts of the plant. It was calculated to be >1 in each of the sample for both lead as well as cadmium, indicating that the aerial parts of the plants are heavy accumulators of the metal and not the roots.

### Table 3. Concentration of lead in different parts of Amaranthus cruentus

<table>
<thead>
<tr>
<th>Parts of Plant</th>
<th>Site</th>
<th>ROOT</th>
<th>STEM</th>
<th>LEAF</th>
<th>INFLORO SCIENCE</th>
<th>TRANSFER FACTOR</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>NALLASOPARA</td>
<td>5.802</td>
<td>9.615</td>
<td>9.004</td>
<td>5.133</td>
<td>3.21</td>
</tr>
<tr>
<td></td>
<td>VASAI</td>
<td>2.738</td>
<td>6.763</td>
<td>0.0</td>
<td>-</td>
<td>2.47</td>
</tr>
<tr>
<td></td>
<td>MIRA ROAD</td>
<td>3.21</td>
<td>5.558</td>
<td>8.163</td>
<td>6.032</td>
<td>2.63</td>
</tr>
<tr>
<td></td>
<td>DAHISAR</td>
<td>3.788</td>
<td>6.675</td>
<td>7.055</td>
<td>-</td>
<td>3.02</td>
</tr>
</tbody>
</table>

* \( \_ \_ \_ = \text{Not performed} 

### Table 4. Concentration of cadmium in different parts of Amaranthus cruentus

<table>
<thead>
<tr>
<th>Parts of Plant</th>
<th>Site</th>
<th>ROOT</th>
<th>STEM</th>
<th>LEAF</th>
<th>INFLORO SCIENCE</th>
<th>TRANSFER FACTOR</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>NALLASOPARA</td>
<td>0.804</td>
<td>0.993</td>
<td>0.776</td>
<td>0.627</td>
<td>2.20</td>
</tr>
<tr>
<td></td>
<td>VASAI</td>
<td>0.98</td>
<td>0.762</td>
<td>0.768</td>
<td>-</td>
<td>1.56</td>
</tr>
<tr>
<td></td>
<td>MIRA ROAD</td>
<td>0.622</td>
<td>0.583</td>
<td>0.97</td>
<td>0.99</td>
<td>2.497</td>
</tr>
<tr>
<td></td>
<td>DAHISAR</td>
<td>0.618</td>
<td>0.95</td>
<td>0.661</td>
<td>-</td>
<td>2.61</td>
</tr>
</tbody>
</table>

* \( \_ \_ \_ = \text{Not performed} 

### Bioconcentration Factor

The calculated Bioconcentration Factors (BCF), which are shown in the table which can help to evaluate the incorporation of heavy metals by plants. Higher quotients reflect a greater potential of the test plants to absorb a particular heavy metal, whereas lower quotients suggest a stronger adsorption in soil.

### Table 5. Bioconcentration factor of cadmium for different parts of Amaranthus cruentus from 4 sample site

<table>
<thead>
<tr>
<th>Sampling Site</th>
<th>BCF_ROOT (%)</th>
<th>BCF STEM (%)</th>
<th>BCF LEAF (%)</th>
<th>BCF INFLORO SCIENCE (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NALLASOPARA</td>
<td>41.92</td>
<td>51.77</td>
<td>40.46</td>
<td>32.69</td>
</tr>
<tr>
<td>VASAI</td>
<td>68.97</td>
<td>53.62</td>
<td>54.04</td>
<td>-</td>
</tr>
<tr>
<td>MIRA ROAD</td>
<td>32.32</td>
<td>35.92</td>
<td>59.77</td>
<td>60.998</td>
</tr>
<tr>
<td>DAHISAR</td>
<td>38.12</td>
<td>58.61</td>
<td>40.78</td>
<td>-</td>
</tr>
</tbody>
</table>

* \( \_ \_ \_ = \text{Not performed} 

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3. Pollution Index

- For Soil Samples

Soil ionic impulsion value helps in indicating the effect of heavy metals on soil and in turn helps in determination of pollution levels using pollution index. Pollution index if above 1 indicates that the given sample is polluted while a value below 1 indicates that the soil is not polluted for the given heavy metal.

The value for Pollution index for soil samples from all sampling sites for lead is under 1 and thus it can be said that the soil has not been contaminated with lead. On the contrary, the values for pollution index for soil samples from all 4 sampling sites have been found above 1 and thus all the soil samples are said to be polluted or contaminated with cadmium.

- For Plant Samples

The ability of plant to sustain the ionic absorption of lead and cadmium can be demonstrated with the help of ionic impulsion and pollution index is calculated so as to study the extent to which the plant is contaminated and if it is fit for human consumption.

The value for pollution index (lead) for all sampling site except for Vasai is more than 1 and thus the plant sample is contaminated with lead. The values for pollution index (cadmium) for all sampling sites is found to be 4 times that of the reference value and thus it can be said the samples are contaminated with cadmium.

4. Human Hazard Quotients

The total hazard index, summing over all HAPs with non-cancer effects in an area. Each HAP contributes its single chemical hazard index to the total. Scorecard calculates a cumulative index across all health effects, and also effect-specific hazard indices (for neurotoxicity, reproductive toxicity, etc.).

If the risk indices of the non-cancer Hazard Quotient (nHQ) and Hazard Index (nHI) exceed unity, harms to human health through non-cancer effects are possible.

Table 6. Bioconcentration factor of lead for different parts of *Amaranthus cruentus* from 4 sampling sites

<table>
<thead>
<tr>
<th>Sampling Site</th>
<th>BCF (%)</th>
<th>BCF ROOT (%)</th>
<th>BCF STEM (%)</th>
<th>BCF LEAF (%)</th>
<th>BCF INFLORESCENCE (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NALASOPARA</td>
<td>12.56</td>
<td>20.82</td>
<td>19.50</td>
<td>11.11</td>
<td></td>
</tr>
<tr>
<td>VASAI</td>
<td>30.68</td>
<td>75.78</td>
<td>0.0</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>MIRA ROAD</td>
<td>2.86</td>
<td>2.86</td>
<td>4.20</td>
<td>3.10</td>
<td></td>
</tr>
<tr>
<td>DAHISAR</td>
<td>14.23</td>
<td>25.07</td>
<td>26.50</td>
<td>-</td>
<td></td>
</tr>
</tbody>
</table>

Table 7. Pollution index of the four soil samples from sampling sites

<table>
<thead>
<tr>
<th>SAMPLING SITES</th>
<th>IONIC IMPULSION FOR LEAD</th>
<th>IONIC IMPULSION FOR CADMIUM</th>
<th>POLLUTION INDEX (LEAD)</th>
<th>POLLUTION INDEX (CADMIUM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NALASOPARA</td>
<td>6.796</td>
<td>1.918</td>
<td>0.181</td>
<td>3.22</td>
</tr>
<tr>
<td>VASAI</td>
<td>2.987</td>
<td>1.192</td>
<td>-0.116</td>
<td>2.52</td>
</tr>
<tr>
<td>MIRA ROAD</td>
<td>13.946</td>
<td>1.274</td>
<td>0.737</td>
<td>2.815</td>
</tr>
<tr>
<td>DAHISAR</td>
<td>5.160</td>
<td>1.273</td>
<td>0.0535</td>
<td>2.811</td>
</tr>
</tbody>
</table>

Table 8. Pollution index for the plant samples from the 4 sampling sites

<table>
<thead>
<tr>
<th>SAMPLING SITES</th>
<th>IONIC IMPULSION FOR LEAD</th>
<th>IONIC IMPULSION FOR CADMIUM</th>
<th>POLLUTION INDEX (LEAD)</th>
<th>POLLUTION INDEX (CADMIUM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NALASOPARA</td>
<td>5.436</td>
<td>1.789</td>
<td>1.88</td>
<td>5.162</td>
</tr>
<tr>
<td>VASAI</td>
<td>3.082</td>
<td>1.584</td>
<td>0.969</td>
<td>4.373</td>
</tr>
<tr>
<td>MIRA ROAD</td>
<td>4.996</td>
<td>1.779</td>
<td>1.701</td>
<td>5.123</td>
</tr>
<tr>
<td>DAHISAR</td>
<td>4.1854</td>
<td>1.493</td>
<td>1.392</td>
<td>4.023</td>
</tr>
</tbody>
</table>

The higher the score exceeds unity, the greater is the level of concern. Indices were calculated separately for adults and children because children are seen to be more sensitive to environmental pollution by heavy metals (Lacatusu *et al.*, 1996; US EPA, 2000; Rieuwerts *et al.*, 2000). The calculated nHQ and HI are tabulated:-

The values for hazard index are found to be very high and thus it can be said that the consumption of this vegetables is not recommended. The least Hazard Index was found to be in the sample Vasai and the highest Hazard Index was found to be in the samples from Nalasopara, Mira road and Dahisar. These samples are not fit for human consumption.

Table 9. The calculated hi for adults

<table>
<thead>
<tr>
<th>SAMPLING SITE</th>
<th>nHQ (LEAD)</th>
<th>nHQ (CADMIUM)</th>
<th>HI (ADULTS)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NALASOPARA</td>
<td>14.975</td>
<td>1.2901</td>
<td>16.2651</td>
</tr>
<tr>
<td>VASAI</td>
<td>0</td>
<td>5.12</td>
<td>5.12</td>
</tr>
<tr>
<td>MIRA ROAD</td>
<td>13.57</td>
<td>6.451</td>
<td>20.021</td>
</tr>
<tr>
<td>DAHISAR</td>
<td>11.725</td>
<td>4.4</td>
<td>16.125</td>
</tr>
</tbody>
</table>

Table 10. The calculated hi for children

<table>
<thead>
<tr>
<th>SAMPLING SITE</th>
<th>nCHQ (LEAD)</th>
<th>nCHQ (CADMIUM)</th>
<th>HI (CHILDREN)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NALASOPARA</td>
<td>34.891</td>
<td>12.028</td>
<td>46.919</td>
</tr>
<tr>
<td>VASAI</td>
<td>0</td>
<td>11.9</td>
<td>11.9</td>
</tr>
<tr>
<td>MIRA ROAD</td>
<td>31.63</td>
<td>15.035</td>
<td>46.665</td>
</tr>
<tr>
<td>DAHISAR</td>
<td>27.34</td>
<td>10.2455</td>
<td>37.586</td>
</tr>
</tbody>
</table>

**Discussion**

The present study was conducted in order to assess the heavy metal contamination in soil and plants from the outskirts of Mumbai in the vicinity to the railway tracks. For the same, 4 samples of soil and *Amaranthus cruentus* were collected from 4 sampling sites. The collected samples were analyzed for the presence of heavy metals Lead and Cadmium. For the soil samples, the concentration of lead was found to be within the permissible limit except for the sample from Mira Road, which exceeded the concentration beyond the limit set for soil used for cultivation. The concentration of Cadmium was above the permissible limit in each of the 4 samples.

For the plant sample, the concentration of lead as well as cadmium was found to be beyond the permissible limit as set by FAO/WHO food standard program codex committee. It was observed that the plant absorbed lead as well as cadmium from the soil and accumulated it in different parts. The accumulation was higher in the aerial parts of the plant as compared to the roots.
The reason for such extremity values might be due to the utilization of waste water for irrigation, over-utilization of land as well as the excessive addition of chemical fertilizer. All these reasons point at the improper management of land and other resources as well as the methods of cultivation which are inappropriate.

Conclusion

The main goal of this research work was to assess the concentration of some toxic heavy metals in the soil samples collected from the vicinity of railway tracks in the outskirts of Mumbai. Also *Amaranthus cruentus* plants were analyzed for heavy metals. A total of 4 soil samples and 4 plant samples were analyzed for presence of lead and cadmium using standard procedures. The results show that the concentration of lead as well as cadmium in the soil sample as well as the plant sample was very high and exceeded the permissible limits.

Also, the pollution index and Non-Cancer Hazard Index were calculated which showed that the plant samples as well as the soil samples were polluted and the consumption of the plants from these areas is a potential risk. This risk is assessed in the Non-Cancer Hazard Indices.

Acknowledgement

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Bibliography