

# POTENTIALITY OF WEED PLANTS FOR PHYTOREMEDIATION OF HEAVY METAL POLLUTED SOIL

## ABSTRACT

Phytoremediation, which utilizes the remarkable ability of plants to concentrate elements and compounds from the environment and metabolize various molecules in their tissues, appears very promising for the removal of pollutants from the environment. In this study, the soil was altered by continuously adding industrial waste, affecting factors like pH, electrical conductivity, soil organic carbon, and metal concentrations. The Contamination factor for metals in the Metoda soil sample was Cu (1.98), Mn (1.58), Cr (1.58), Cd (7.67), and Pb (166.72), while the Pollution load index was 397.51. In comparison, the Shapar soil sample had a Contamination factor of Cu (2.24), Mn (1.58), Cr (1.23), Cd (12.67), and Pb (167.44), with a Pollution load index of 480.49. The contaminated sites showed higher micronutrient and trace element concentrations in weed plants compared to non-contaminated sites. *Withania somnifera* L. and *Calotropis gigantea* L. had the highest concentrations of Cd  $2.08 \pm 0.05$  and  $2.60 \pm 0.05$ , respectively. *Amaranthus spinosus* L. accumulated the most Pb ( $36.34 \pm 0.2$ ) and Cr ( $64.30 \pm 0.50$ ), while *Withania somnifera* L. accumulated the most Mn ( $28.08 \pm 0.43$ ), and *Ipomoea aquatica* Forssk accumulated the most Cu ( $43.68 \pm 0.37$ ). This article addresses key research, potential benefits and the potential future needs for phytoremediation. Results indicated that uptake patterns of heavy metals by weed plant tissues were more or less related with nature of metals and species specifications. All the studies suggested that each weed plant could be used as a modern tool as a biomonitor.

**Keywords:** Biomonitors, Industrial waste, Heavy metals, Plant tissues, Phytoremediation

## 1 INTRODUCTION

“Phytoremediation is combined multidisciplinary approach to the cleanup of contaminated soils, which combines the disciplines of plant physiology, soil chemistry, and soil microbiology. Phytoremediation has been applied to a number of contaminants in small scale field and laboratory studies. These contaminants include heavy metals, radionuclides, chlorinated solvents, petroleum hydrocarbons, polychlorinated biphenyls (PCBs), (polycyclic aromatic hydrocarbons) PAHs, organophosphate insecticides, explosives, and surfactants” [1]. “Certain species of higher plants can accumulate very high concentrations of metals in their tissues without showing toxicity” [2]. Such plants can be used successfully to clean up heavy metal polluted soils if their biomass and metal content are large enough to complete remediation within a reasonable period.

“Phytoremediation has secured popularity with government agencies and industry in the past ten years. This popularity is based in part on the relatively low cost of phytoremediation, combined with the limited funds available for environmental cleanup. The costs associated with environmental remediation are staggering. Currently, \$6-8 billion per year is spent for environmental cleanup in the United States, and \$25-50 billion per year worldwide” [3, 51]. Because biological methods are eventually solar-driven, phytoremediation is on average tenfold low-cost than engineering-based remediation methods. The way that phytoremediation is normally completed in situ adds to its expense adequacy and may decrease openness of the contaminated substrate to people, natural life, and the climate. As a result, environmental consulting firms are increasingly including phytoremediation in their package of offered technologies, corporations like the electric power, oil, and chemical industries like to advertise their involvement with this environmentally friendly technology, and government agencies like to include phytoremediation in their cleanup strategies to stretch available funds.

“In general, either in situ or ex situ, remediation technologies accomplish one of two things: They either remove the contaminants from the substratum (site decontamination or clean-up techniques) or lessen the risk of exposure (site stabilization techniques) to the contaminants. One “gentle” plant-based site stabilization approach, suitable for heavily contaminated sites, is phytostabilization aimed to decrease soil metal bioavailability using a combination of plants and soil amendments” [4]. “Another approach directed

towards real decontamination is trace element phytoextraction, representing use of plants for trace element removal from the soil by concentrating them in the harvestable parts" [5]. "An opinion exists that trace element phytoextraction will be more economically feasible if, in addition to metal removal, plants produce biomass with an added economical value" [2].

"Recently, many plants have been reported to accumulate high level of the toxic metals in their sinks in the aquatic and terrestrial ecosystems. Such plants can remove the pollutant metals from soil and water" [6, 7, 52]. This potential approach to clean the environment, termed phytoremediation, draws on our centuries of experience in cultivating crops and is emerging as a low cost treatment technology for the contaminated environment.

In our study we used native plants for remediation of polluted soils while most of the researchers were using well known metals accumulators. Schickler H. used the most promising terrestrial plant species include Indian mustard (*Brassica juncea* L), Italian serpentine plant (*Alyssum bertolonii*), and *Thlaspi caerulescens* to extract heavy metals from the soil [8]. Ye used *Typha latifolia* and *Phragmites australis* for phytoremediation of polluted industrial soil [9]. As a result, we conclude that native plants are more efficient than other plant species. We can also accept that the characteristics of the metal play a role in the selection of plant species. Phytoremediation, or the use of a potential plant species or cultivar to remove heavy metals from contaminated soils, sludges, and sediments near industrial towns with particular metal contaminants, appears to be a challenge. In order to select a large number of potential phytoremediators, an area-by-area and metal-by-metal screening must take into account Indian cultivars that have adapted to the particular agroclimatic conditions. A focus on finding a local cultivar of commercially viable phytoremediator plants aimed at cleaning specific sites in heavy metal-contaminated cities, towns, and metropolitan areas.

## 2 MATERIALS AND METHODS

### 2.1 Field survey

In India, Rajkot is most developing city of Gujarat and epicenter of the Saurashtra region. Four different industrial sites are as bellow:

Shapar A

Shapar B

Metoda A

Metoda B

As per industrialization, there are more than 500 foundry units in Rajkot region. Most of foundry units in Rajkot produce grey iron castings for domestic market and also export to worldwide. Industrial products include bearings, diesel engines, kitchen knives, watch part, automotive parts etc. there are so many forging industries, casting industries and machine tools production units. Industrial waste water is disposed of through sewer lines, which at certain points are mixed with agricultural land or this water is lifted by diesel pumps etc. to irrigate nearby crop fields.

## 2.2 Collection of soil samples

Soil samples were collected from four different industrial fields. After removing grasses and other vegetation from the surface, samples were taken from 0-15 cm depth from 5 to 8 different points in the same location using Auger. All the samples were mixed together and almost 5 kg taken out of it. Half of the soil samples were dried in shade and then stored in clean autoclaved polythene bag. These samples were then carried to the laboratory. There is no difference in soil profile for all collected soil samples. At each site 2-3 composite samples were taken from different horizons viz., 0-15 cm, 15-30 cm and 30-45 cm segments. Oven dried Soil samples were crushed in grinder, mixed well together and passed through a sieve with round holes of 2 mm diameter. One kg of soil sample was taken as representative of the original material [10].

## 2.3 Analysis of Soil

At the outset, a comprehensive survey was conducted in order to evaluate soil from different locations of industrial areas located in Rajkot for assessment of heavy metals contamination (Cu, Cr, Mn, Pb and Cd).

### 2.3.1 Physico-chemical properties

**Table 1. Methods for Physico-chemical properties**

<b>Properties</b>	<b>Methods</b>
<b>Soil texture</b>	Hydrometric method [11].
<b>pH</b>	Digital pH meter [12].
<b>Electrical conductivity</b>	Digital EC meter [12].
<b>Bulk Density</b>	By lab method for disturbed soil. [13]
<b>Soil Organic Carbon</b>	Walkley and Black method [14]
<b>Soil Organic Matter</b>	Van Bemmelen,[15]
<b>Heavy Metals</b>	Atomic Absorption Spectrophotometer (AAS), DTPA method [16]

### 2.3.2 Contamination factor (CF)

The CF is the ratio obtained from heavy metal concentrations in polluted and background sites [17].

$$CF = C_{mSample} / C_{mBackground}$$

Where  $C_m$  represents the concentrations of metals in contaminated and background sites.

### 2.3.3 Pollution load index (PLI)

PLI was calculated by the following formula adopted from Muhammad et al. [18].

$$PLI = \sqrt[n]{CF_1 \times CF_2 \times CF_3 \dots \dots \dots CF_n}$$

Where  $n$  is the number of metals and  $CF$  is the contamination factor value.

## 2.4 Collection of weed plants

### 2.4.1 Survey the flora of contaminated sites

An ecological survey was done from March to March for two years continuously. Community analysis was carried out during rainy season when majority of the plants were at the peak of their growth. In every study sites, 30 quadrats of 10 m X 10 m (100 sq m) size were randomly laid to study plant species. The herbaceous species was studied by laying 50 quadrats of 1m X 1m (1sq m) size randomly in each study site.

#### **2.4.1.1 Quantitative analysis**

The important quantitative analysis such as density and frequency of tree species, shrubs and herbs species were determined as per Curtis and McIntosh [19].

##### **(a) Density**

Density is an expression of the numerical strength of a species where the total number of individuals of each species in all the quadrats is divided by the total number of quadrats studied.

Density is calculated by the equation:

$$\text{Density} = \frac{\text{Total number of individuals of a species in all quadrats}}{\text{Total number of quadrats studied}}$$

##### **(b) Frequency (%)**

This term refers to the degree of dispersion of individual species in an area and usually expressed in terms of percentage occurrence. It was studied by sampling the study area at several places at random and recorded the name of the species that occurred in each sampling units.

It is calculated by the equation:

$$\text{Frequency (\%)} = \frac{\text{Number of quadrats in which the species occurred} \times 100}{\text{Total number of quadrats studied}}$$

Due to the heterogeneity of the characteristics of the soil, observed in the first sampling, each subplot was divided equally, establishing 20 different vegetation sampling sites. Surveys were conducted in

October-November 2021. For each survey, a 30 X 30 cm quadrat was used [20]. The quadrat was randomly placed three times within each sampling site. Plant species were listed and vegetation cover estimated. Plant species were determined and named following the keys and nomenclature proposed by Valde's et al. [21]. Then the most frequent species were collected for metal analysis.

## **2.5 Analysis of Weed plants**

### **2.5.1 Metal analysis of plants of contaminated sites.**

Plant samples were collected from the same sites as the soil samples. Plant identification was confirmed by the Forest Research of India, Dehradun. At least three to five individuals of all plant species were randomly collected within the sampling areas. Fresh plant materials were washed thoroughly with the tap water, washed for at least 15 seconds with a 0.1 N HCl solution and cleaned with distilled water and then separated into leaf, root and shoot. All plant parts were oven dried at 72°C for 72 h and then ground and passed through a 500-µm stainless-steel sieve to powders. For total metal concentrations in the plant components 0.5 g of plant samples were digested. Heavy metal concentrations in plants were also determined by acid digestion method by Gupta [22]. The total metal concentrations were measured by an atomic absorption spectrophotometry (AAS A-700, Perkin-Elmer) [16]. To assess the analytical precision, three analytical replicates of each sample, an appropriate standard reference material (from Sigma-Aldrich Company) and a reagent blank were performed in each analytical batch.

### **2.5.2 Bio-accumulation factor (BAF)**

Bio-accumulation factor is used to explain the transport of trace elements (heavy metals) from complex soil mixture to plants. The bioaccumulation factor of a metal can be calculated as the ratio between the amount of metal in the plant (dry weight) to same in the corresponding soil [23, 24]. The BAF for different heavy metals was calculated as follows:

$$BAF = C_{Plant} / C_{Soil}$$

$C_{Plant}$  is concentration of heavy metal in plant (dry weight) and  $C_{Soil}$  is concentration of heavy metal in corresponding soil under plant.  $BAF = 1$  means plant only absorbed the heavy metal but no accumulation while  $BAF > 1$  denotes absorption and accumulation of heavy metal by plant [25].

## 2.6 Data analysis

Statistical analysis was done by an analysis of variance (ANOVA). Data were considered to be significant at  $p \leq 0.05$  and highly significant at  $p \leq 0.005$ , level of significance.

## 3 RESULTS AND DISCUSSION

### 3.1 Analysis of Soil

#### 3.1.1 Physico-chemical properties of Soil

As per table 2, texture of contaminated soil was the class sandy clay loamy which was in accord with the report of Carter and Gregorich [26]. In table 2 texture reflects the particle size distribution of the soil and thus the content of fine particles like oxides and clay. These compounds are important adsorption media for heavy metals in soils. Results showed that soil samples had sand particle between 68 to 70 % where silt particles were 10 to 12%. Clay particles were 19 to 21% in soil samples. The results obtained indicate that the dumpsite soils were highly permeable which are not suitable for waste disposal due to its high leaching capability contaminating groundwater resource in and around the waste dumpsite.

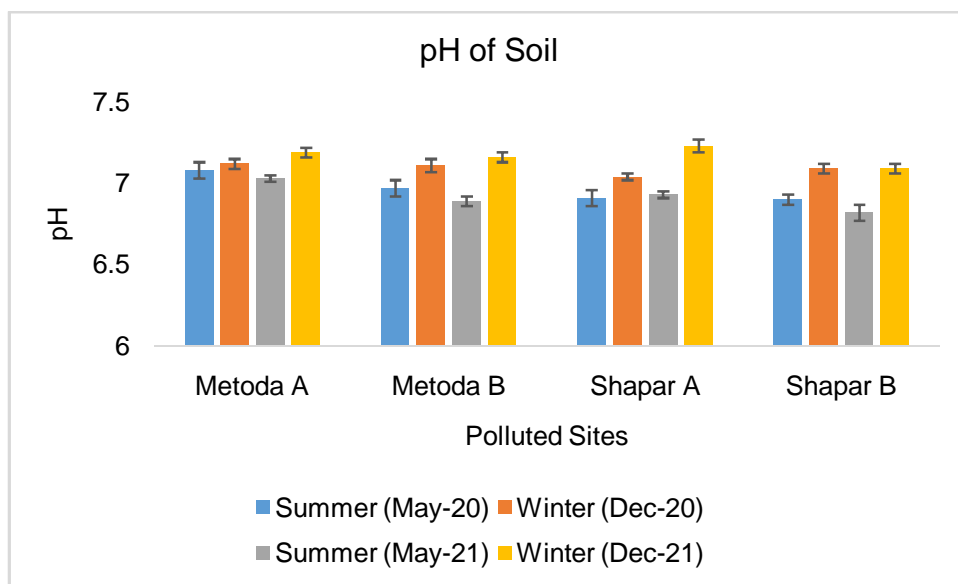
**Table 2. Soil Texture**

Textural composition (%)	Different soil profiles (0-15)cm soil depth			
	Metoda A	Metoda B	Shapar A	Shapar B
Sand (0.05– 2.0 mm)	68.75±0.85	67.84±0.85	69.27±0.85	68.9±0.85
Silt (0.002– 0.05 mm)	11.99±0.45	12.03±0.43	10.15±0.45	10.06±0.45
Clay (<0.002 mm)	19.26±0.51	20.13±0.55	20.58±0.52	21.04±0.53

*Mean values ± Standard deviation of means of three replicates*

pH is one of the most significant parameter which influences the availability of most of the elements in the soil to both plants and microbes. The soil when continuously amended with industrial waste alters the pH

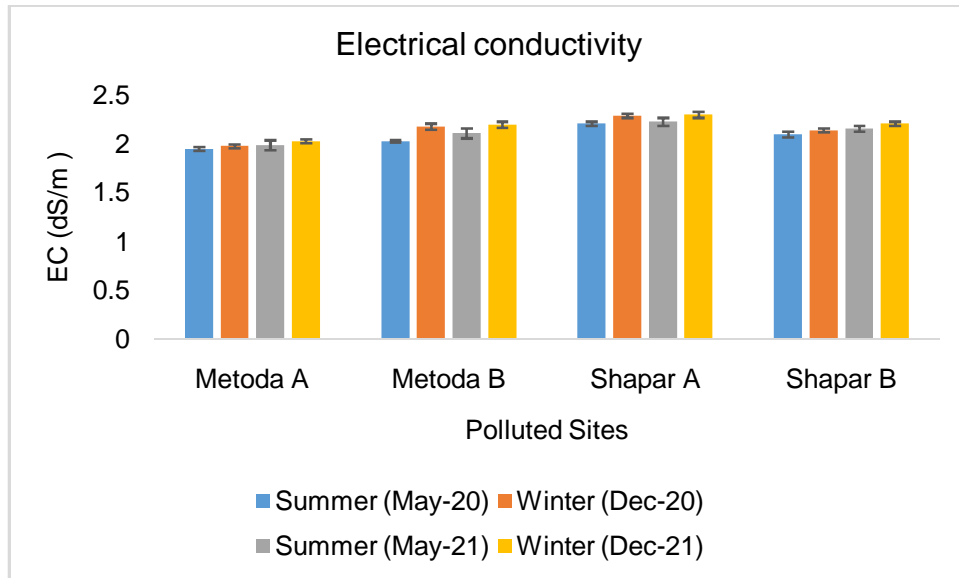
to a certain extent and thus one crucial aspect in pollution studies of soils [27]. The physico – chemical analysis of the dumpsite soil samples investigated in the present study revealed pH value was varied from 6.8 to 7.2 in summer and 6.9 to 7.3 in winter (Figure 1) with a mean pH of 6.98 in summer and 7.12 in winter [28] which were in accord with the ranges reported by [29] in their earlier studies.



**Figure 1. pH of Soil samples**

*Mean values ± Standard deviation of means of three replicates*

The electrical conductivity recorded for the soil samples in the polluted sites ranged from 1.95 dS/m to 2.23 dS/m in summer and 1.98 dS/m to 2.3 dS/m in winter (Figure 2) with a mean EC of 2.09 dS/m in summer and 2.18 dS/m in winter. The results were exactly followed the results those reported by Uba et al., [30]. The EC recorded for the polluted site soils were slightly above the prescribed ranges for agricultural soils which might hinder the uptake of water by the plants from the soils [31].



**Figure 2. EC of Soil samples**

*Mean values  $\pm$  Standard deviation of means of three replicates*

Bulk density is an indicator of soil suitability for plant growth. In table 3, bulk density recorded for the soil samples in the polluted sites were ranged from 1.29 mg/m<sup>3</sup> to 1.32 mg/m<sup>3</sup> in summer and 1.25 mg/m<sup>3</sup> to 1.3 mg/m<sup>3</sup> in winter with a mean bulk density of 1.31 mg/m<sup>3</sup> in summer and 1.28 mg/m<sup>3</sup> in winter. Mathur [32] observed that bulk density of soil treated with organic manure decreased from 1.46 to 1.40 mg/m<sup>3</sup> which confirms the present findings. Similar results were also reported by Chaudhury [33] in a 31 years long-term field experiment for assessing soil quality under rice-based cropping system in Indo-Gangetic alluvial soils of India. Bulk density was found slightly higher in summer compare to winter because metal concentration also changed in winter.

**Table 3. Bulk Density of Soil**

Bulk Density (mg/m <sup>3</sup> )				
Site	Metoda A	Metoda B	Shapar A	Shapar B
Summer (May-20)	1.31 $\pm$ 0.03	1.3 $\pm$ 0.02	1.29 $\pm$ 0.01	1.29 $\pm$ 0.01

Winter (Dec-20)	1.29±0.02	1.28±0.02	1.26±0.01	1.25±0.02
Summer (May-21)	1.32±0.01	1.32±0.01	1.31±0.02	1.3±0.01
Winter (Dec-21)	1.29±0.02	1.3±0.01	1.3±0.02	1.29±0.02

*Mean values ± Standard deviation of means of three replicates*

As per table 4, soil organic carbon in the polluted site soils ranged from 0.78% to 1.02% in summer and 0.8% to 0.96% in winter with a mean of 0.9% in summer and 0.88% in winter. The high soil organic carbon of the dumpsite soils is due to the high organic and compostable matter degradation [34] and also silt content in the soils which might have contributed to the excessive accumulation of SOC [35].

**Table 4. Organic Carbon of Soil**

<b>Organic Carbon %</b>				
<b>Site</b>	<b>Metoda A</b>	<b>Metoda B</b>	<b>Shapar A</b>	<b>Shapar B</b>
Summer (May-20)	0.9±0.02	0.96±0.02	0.78±0.02	0.81±0.02
Winter (Dec-20)	0.89±0.03	0.92±0.02	0.81±0.02	0.8±0.03
Summer (May-21)	0.98±0.02	1.02±0.03	0.87±0.02	0.9±0.02
Winter (Dec-21)	0.91±0.03	0.96±0.03	0.81±0.03	0.86±0.02

*Mean values ± Standard deviation of means of three replicates*

The SOM (Soil Organic Matter) of soil samples in the polluted site soil samples were ranged from 1.34% to 1.76% in summer and 1.38% to 1.66% in post - monsoon with a mean of 1.55% in summer and 1.52%

in winter. However as per WHO standard Agricultural soil recorded lower SOM percentages compared to the polluted site soils which was in accord with the previous studies of [36]. Soil organic matter is thus one crucial characteristics impacting soil physical properties.

**Table 5. Soil Organic Carbon**

Soil Organic Matter %				
	Metoda A	Metoda B	Shapar A	Shapar B
Summer (May-20)	1.55±0.05	1.66±0.06	1.34±0.07	1.40±0.06
Winter (Dec-20)	1.53±0.05	1.59±0.06	1.40±0.07	1.38±0.05
Summer (May-21)	1.69±0.06	1.76±0.05	1.50±0.07	1.56±0.07
Winter (Dec-21)	1.57±0.05	1.66±0.04	1.40±0.06	1.48±0.06

*Mean values ± Standard deviation of means of three replicates*

Copper content in the soils of the polluted site soil samples were ranged from 3.9 mg/kg to 6.83 mg/kg in summer and 6.38 mg/kg to 9.01 mg/kg in winter with a mean of 5.37 mg/kg in summer and 7.7 mg/kg in winter. The results recorded for polluted site soils might be due to the introduction of the industrial wastes which might increase the soil copper levels [37]. Among all the samples analyzed polluted site soils recorded lower copper levels beyond the International Standards for Agricultural soils.

The amount of manganese varies greatly and is generally present in the form of manganese associated with organic matter, exchangeable manganese or manganese oxides. The mean Mn content in the soils of the polluted site ranged from 23.5 mg/kg to 25.6 mg/kg in summer and 24.4 mg/kg to 29.41 mg/kg in winter with a mean of 24.55 mg/kg in summer and 28.71 mg/kg in winter. The results recorded for the polluted site soil samples were lower than the ranges reported in a similar study by Udeme [38]. Among all the soil samples analyzed polluted site soils recorded higher Mn content due to the solubilizing effect

of organic matter on Mn. The high Mn levels in polluted site may be due to the discarded battery materials and other metallic discards [39].

The mean Cr content in the soils of the polluted site samples were ranged from 0.21 mg/kg to 0.4 mg/kg in summer and 0.21 mg/kg to 1.02 mg/kg in winter with a mean of 0.31 mg/kg in summer and 0.62 mg/kg in winter. Similar study by Nwajei [40] also reported lower levels of Cr in the surface soils of a waste polluted site.

The mean Pb content in the soil samples from the polluted site ranged from 2.68 mg/kg to 19.21 mg/kg in summer and 24.6 mg/kg to 41.02 mg/kg in winter with a mean of 10.95mg/kg in summer and 32.81 mg/kg in winter presented. Among all the soil samples analyzed polluted site soils recorded higher Pb content which was similar to the previous findings of Ogunyemi [41]. The high Pb levels in polluted site soils may be due to the discarded plastics, glasses, paints, ceramics, batteries etc; The overall mean Pb levels recorded for the dumpsite soils samples in both the seasons were beyond the WHO standards for soil and below the International Standards for Agricultural soils.

The mean Cd content in the soils of the polluted site samples were ranged between 0.18 mg/kg to 0.25 mg/kg in summer and 0.23 mg/kg to 0.38 mg/kg in winter with a mean of 0.22 mg/kg in summer and 0.31 mg/kg in winter. Among all the samples analyzed polluted site soils recorded higher Cd levels than the control soil samples. The overall mean Cd levels in polluted site soils were within the permissible ranges of the International Standards for Agricultural soils and WHO. The results obtained were contrary to a similar study of Awokunmi [42] who reported very high levels of Cd levels in the surface soils of the polluted site. Many industrial processes required cadmium content raw materials and many industrial operations required cadmium as a catalyst; hence these applications are responsible for increase the lead content [43].

**Table 6. Heavy metals of Soil (in mg/kg)**

	Elements	Cadmium	Lead	Chromium	Manganese	Copper
<b>Metoda</b>	Summer (May-20)	0.2±0.01	2.68±0.08	0.4±0.01	25.5±0.62	6.68±0.32
<b>A</b>	Winter (Dec-20)	0.23±0.06	30.01±0.42	0.41±0.05	26.01±0.63	6.92±0.52

	Summer (May-21)	0.2±0.01	18.4±0.51	0.4±0.01	25.6±0.52	6.83±0.41
	Winter (Dec-21)	0.31±0.01	36.31±1.02	1.02±0.04	29.41±0.87	8.34±0.69
<b>Metoda B</b>	Summer (May-20)	0.21±0.01	2.69±0.12	0.31±0.02	25.00±0.28	6.05±0.5
	Winter (Dec-20)	0.27±0.01	24.6±0.78	0.38±0.04	25.87±0.81	6.38±0.25
	Summer (May-21)	0.2±0.05	14.51±0.58	0.35±0.03	25.21±0.42	6.12±0.21
	Winter (Dec-21)	0.32±0.01	28.06±1.25	0.98±0.01	28.06±0.85	7.59±0.22
<b>Shapar A</b>	Summer (May-20)	0.18±0.01	2.68±0.05	0.32±0.01	24.00±0.96	3.9±0.05
	Winter (Dec-20)	0.32±0.02	32.78±1.04	0.21±0.01	24.4±0.84	6.57±0.52
	Summer (May-21)	0.25±0.02	19.21±1.02	0.21±0.02	23.9±0.74	5.28±0.5
	Winter (Dec-21)	0.37±0.02	41.02±0.21	0.29±0.02	26.54±1.05	7.91±0.5
<b>Shapar B</b>	Summer (May-20)	0.21±0.01	2.68±0.05	0.40±0.01	23.5±0.88	4.09±0.05
	Winter (Dec-20)	0.38±0.02	30.14±0.85	0.32±0.01	25.93±1.04	7.82±0.14
	Summer (May-21)	0.23±0.02	15.67±0.52	0.30±0.01	23.53±0.99	5.42±0.5
	Winter (Dec-21)	0.3±0.01	39.10±0.75	0.40±0.01	26.88±1.05	9.01±0.6

*Mean values ± Standard deviation of means of three replicates*

### 3.1.2 Contamination factor and Pollution load index

Heavy metal concentrations in soil of Rajkot industrial zones are mentioned in table 1 & 2. With the help of background value (agricultural soil) of the heavy metals the contamination factor and pollution load index (PLI) was calculated which reflects the pollution of metals in soil. Contamination factor of metals in Metoda soil sample was Cu (1.98), Mn (1.58), Cr (1.58), Cd (7.67) and Pb (166.72). The PLI for this site was 397.51. Contamination factor of Shapar soil sample was Cu (2.24), Mn (1.58), Cr (1.23), Cd (12.67) and Pb (167.44). The PLI value for this site was 480.49. The Shapar site was highly contaminated when compared to the Metoda sites. These high values of PLI in the study area could be due to open dumping of industrial waste that may cause a potential health risk to the local community as well to grazing animals. In this study, the PLI values were greater than those reported by Muhammad et al. [18].

Accumulation of lead in soil was significantly high compare to other metals found in their respective presence in soil during the study period. The intensity of adverse effects of several heavy metals depends upon the form and percentage distribution in soil. The soil parameters such as soil texture, organic matter content, pH, redox potential will affect the mobility of metal and its translocation [44]. Most of the trace metals are found in crystalline state and are immobile. Oxides of iron and manganese are generally coated on organic matted present in soil and fine particles of clay along with other colloidal material which are generally active provide mobility platform to trace metal. Several human interventions in environment geochemical cycle of trace metals, resulting in soil and water contamination which finally enters in food chain [44].

The CF values at the Metoda site were observed to be in the order of  $Pb > Cd > Cu > Mn \geq Cr$ . According to the Muller classification, Pb and Cd were present at very highly polluted levels; others were highly polluted. The CF values at the Shapar site were observed to be in the order of  $Pb > Cd > Cu > Mn > Cr$ . The concentrations of Pb, Cd and Cu were present at very highly polluted levels; others were highly polluted level, as suggested by Muller. These results revealed that the Metoda and Shapar sites showed higher CF values for Pb, Cd, Cr, Mn, and Cu. The Shapar site showed higher CF values for Cd and Cu.

### **3.2 Survey the flora of contaminated sites**

In the surveys of the polluted sites, more than 20 vascular plant species, representing 16 genera and 11 families were found. Most of these plants were annual or biannual and perennials were less represented. **Table 7 shows** the species that were present in more than 55% frequency and density of the sampling sites. Moreover, under a Mediterranean condition, greatest development of annuals occurs in spring, coinciding with the March and June surveys. In general, species richness, vegetation cover and biomass production were significantly higher in the industrial zones in each survey. This pointed to a positive effect of the metal concentration in enhancing plant colonization and plant development in spite of the chemical heterogeneity revealed by soil analyses. The results obtained from these areas clearly indicate the adverse conditions (very high metal contamination) of the polluted soil. The positive effects of the pollution might be related to an increase in some species density. There were strong positive correlations

between soil metal concentration and the number of species, vegetation cover and biomass production. The second thing was the increase in soil pH reduced trace element solubility and thus potential toxicity to plants and microorganisms. Therefore the increase in soil pH seems to be the most important heavy metals effect in reducing trace element solubility. Moreover, the nutrients added through the pollution could also contribute to improving soil fertility in this soil.

**Table 7. Dominant plant species of polluted sites**

NO	Plant Name	Family
1	<i>Alternanthera caracasana</i> Kunth.	Amaranthaceae
2	<i>Amaranthus spinosus</i> L.	Amaranthaceae
3	<i>Calotropis gigantea</i> L.	Apocynaceae
4	<i>Cyperus haspan</i> L.	Cyperaceae
5	<i>Datura stramonium</i> L.	Solanaceae
6	<i>Ipomoea aquatica</i> Forssk.	Convolvullaceae
7	<i>Phyllanthus nirudi</i> L.	Phyllanthaceae
8	<i>Withania somnifera</i> L. (Dunal)	Solanaceae

### 3.3 Metal analysis of plants of contaminated sites.

In general, plants growing in the contaminated soils had higher concentrations of micronutrients and trace elements than control plants.

#### Cadmium

The highest Cd concentration was measured 2.60 mg/kg in *C. gigantea* at shapar which is higher than the permissible limit set by WHO [45]. The result of Cd concentration of the plants species from both polluted sites were grouped together with Cd concentration of the Soil from both sites and correlated to find the correlation coefficient between them.

## Lead

The highest Pb concentration was measured 36.34 mg/kg in *A. spinosus* at shapar which is higher than the permissible limit set by WHO [45] and the lowest of 0.0 mg/kg in *I. aquatica* at shapar as shown in table. The result of Pb concentration of the plants species from both polluted sites were grouped together with Pb concentration of the Soil from both sites and correlated to find the correlation coefficient between them.

## Chromium

The highest Cr concentration was measured 64.30 mg/kg in *A. spinosus* at shapar which is higher than the permissible limit set by WHO [45]. The result of Cr concentration of the plants species from both polluted sites were grouped together with Cr concentration of the Soil from both sites and correlated to find the correlation coefficient between them.

## Manganese

The highest Mn concentration was measured 28.08 mg/kg in *W. somnifera* at metoda which is higher than the permissible limit set by WHO [45]. The result of Mn concentration of the plants species from both polluted sites were grouped together with Mn concentration of the Soil from both sites and correlated to find the correlation coefficient between them.

## Copper

The highest Cu concentration was measured 43.68 mg/kg in *I. aquatica* at metoda which is higher than the permissible limit set by WHO [45]. The result of Cu concentration of the plants species from both polluted sites were grouped together with Cu concentration of the Soil from both sites and correlated to find the correlation coefficient between them.

**Table 8. Metal analysis of weed plants of Metoda site (in mg/kg)**

Plant Name	Cd	Pb	Cr	Mn	Cu
<i>Alternanthera caracasana</i> Kunth.	1.48±0.05	11.23±0.5	4.09±0.04	18.04±0.05	2.03±0.05

<i>Amaranthus spinosus</i> L.	0.59±0.02	21.32±0.11	36.40±0.13	12.03±0.8	3.09±0.04
<i>Calotropis gigantea</i> L.	1.87±0.06	3.22±0.12	2.35±0.12	8.90±0.11	9.19±0.12
<i>Cyprus haspan</i> L.	0.98±0.01	1.79±0.01	4.36±0.11	9.65±0.11	11.64±0.11
<i>Datura stramonium</i> L.	0.12±0.01	0.43±0.01	2.70±0.05	5.96±0.05	1.22±0.04
<i>Ipomoea aquatic</i> Forssk.	0.26±0.01	0.07±0.01	6.09±0.05	18.93±0.22	43.68±0.37
<i>Phyllanthus nirudi</i> L.	0.12±0.02	1.09±0.05	3.06±0.05	12.37±0.05	11.71±0.12
<i>Withania somnifera</i> L. (Dunal)	2.08±0.05	4.75±0.07	4.09±0.05	28.08±0.43	12.44±0.17

Mean values ± Standard deviation of means of three replicates

**Table 9. Metal analysis of weed plant of Shapar site (in mg/kg)**

Plant Name	Cd	Pb	Cr	Mn	Cu
<i>Alternanthera caracasana</i> Kunth.	1.02±0.05	17.38±0.25	5.78±0.15	12.21±0.14	0.10±0.04
<i>Amaranthus spinosus</i> L.	0.45±0.05	36.34±0.2	64.30±0.50	3.06±0.12	2.01±0.05
<i>Calotropis gigantea</i> L.	2.60±0.05	10.50±0.12	4.30±0.10	7.74±0.11	18.20±0.24
<i>Cyprus haspan</i> L.	1.09±0.1	2.38±0.11	3.09±0.10	5.97±0.21	22.36±0.51
<i>Datura stramonium</i> L.	0.05±0.01	0.18±0.01	0.30±0.01	1.10±0.05	0.70±0.01
<i>Ipomoea aquatica</i> Forssk.	0.10±0.01	0.00±0.00	4.55±0.09	16.74±0.24	32.41±0.65
<i>Phyllanthus nirudi</i> L.	0.09±0.01	0.93±0.08	2.13±0.05	7.27±0.21	9.91±0.20
<i>Withania somnifera</i> L. (Dunal)	1.69±0.08	5.34±0.11	3.69±0.10	25.66±0.50	16.03±0.50

Mean values ± Standard deviation of means of three replicates

### 3.4 Bioaccumulation factor (BAF)

Ability of a plant to accumulate metals from contaminated soils was evaluated by the BAF, according to studies of Fayiga [46] and Yoon [47]. This study assumed that plants with BAF values > 1 are accumulators, while plants with BAF values < 1 are excluders [48]. Metoda and Shapar both polluted sites results showed that most of weed plant species had BAF values > 1 which indicating that they had the potential for use as accumulators of heavy metals. The success of the phytoremediation process depends on heavy metal removal by the plants [49]. Conversely, the accumulation of heavy metals from

soil to plant parts were an extremely multifaceted process which affected by several factors, which exert different influences on the process by means of various mechanisms. There were so many influencing factors include the chemical forms of the heavy metals, pH of the soil, conductivity of the soil, organic carbon, organic matter content, plant species, climatic conditions, and irrigation with polluted water [50].

**Table 10. BAF value in weed plants of Metoda site**

Plant name	Metoda soil sample (BAF value)				
	Cd	Pb	Cr	Mn	Cu
<i>Alternanthera caracasana</i> Kunth.	7.40	4.19	10.23	0.71	0.30
<i>Amaranthus spinosus</i> L.	2.96	7.96	91.00	0.47	0.46
<i>Calotropis gigantea</i> L.	9.35	1.20	5.88	0.35	1.38
<i>Cyprus haspan</i> L.	4.90	0.67	10.90	0.38	1.74
<i>Datura stramonium</i> L.	0.60	0.16	6.75	0.23	0.18
<i>Ipomoea aquatica</i> Forssk.	1.30	0.03	15.23	0.74	6.54
<i>Phyllanthus nirudi</i> L.	0.62	0.41	7.65	0.49	1.75
<i>Withania somnifera</i> L. (Dunal)	10.40	1.77	10.23	1.10	1.86

**Table 11. BAF value in weed plants of Shapar site**

Plant name	Shapar soil sample				
	Cd	Pb	Cr	Mn	Cu
<i>Alternanthera caracasana</i> Kunth.	4.87	6.49	14.45	0.52	0.02
<i>Amaranthus spinosus</i> L.	2.12	13.56	160.75	0.13	0.49
<i>Calotropis gigantea</i> L.	12.38	3.92	10.75	0.33	4.45
<i>Cyprus haspan</i> L.	5.19	0.89	7.73	0.25	5.47
<i>Datura stramonium</i> L.	0.24	0.07	0.75	0.05	0.17
<i>Ipomoea aquatica</i> Forssk.	0.47	0.00	11.38	0.71	7.92
<i>Phyllanthus nirudi</i> L.	0.44	0.35	5.33	0.31	2.42

<b><i>Withania somnifera</i> L. (Dunal)</b>	8.05	1.99	9.23	1.09	3.92
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#### 4 CONCLUSION

High concentrations of metals can accumulate in the tissues of some higher plant species without causing harm. *Withania somnifera* L. and *Calotropis gigantea* L. had the highest concentrations of Cd  $2.08 \pm 0.05$  ppm and  $2.60 \pm 0.05$  ppm, respectively. *Amaranthus spinosus* L. accumulated the most Pb ( $36.34 \pm 0.2$  ppm) and Cr ( $64.30 \pm 0.50$  ppm), while *Withania somnifera* L. accumulated the most Mn ( $28.08 \pm 0.43$  ppm), and *Ipomoea aquatica* Forssk accumulated the most Cu ( $43.68 \pm 0.37$  ppm). However, the potential of phytoremediation steps that make use of hyper accumulators has been the subject of some concerns regarding the invasiveness and disruption of indigenous ecosystems. This is due to the fact that the introduction of alien plants may alter the way that ecosystems work. Therefore, using native hyper accumulator plants from nearby regions for soil remediation is the best option.

#### 5 Competing Interests

Authors have declared that no competing interests exist.

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