***Pak. J. Bot.*, 58(12): December 2026.**

ARTICLE | **DOI: http://dx.doi.org/10.30848/PJB2026-12(13)**

ASSESSMENT OF HEAVY METAL POLLUTION IN SOIL AND VEGETATION ALONG ROADSIDE OF GUJRAT, PAKISTAN

LASANIA MEHMOOD[1], KHALID NAWAZ[1*], KHALID HUSSAIN[1], NOSHIA ARSHAD[1], DURR-E-NAYAB[1], MANAL A. ALSHAQHAA[2], AROOJ KHALID[3] AND MOUNIRA MKADDEM-GUEDRI[4]

[1]Department of Botany, University of Gujrat (UOG), Gujrat-Pakistan
[2]Department of Biology, College of Science King Khalid University ABHA, Saudi Arabia
[3]Department of Chemistry, University of Gujrat (UOG), Gujrat-Pakistan
[4]Institut Supérieur des Sciences Appliquées et de Technologie de Gabès, Tunisia

*Corresponding author's email: halid.nawaz@uog.edu.pk

Abstract

Overuse of vehicles causes heavy metal pollution which lead to a negative effect on the soil and vegetation grown along roadside. Present study aims to examine the effect of vehicular metal pollution on soil and wild plants grown along roadside i.e., *Cannabis sativa* L., *Parthenium hysterophorus* L., *Cynodon dactylon* L, *Calotropis procera* (Ait.) Ait. F. and *Ricinus communis* L. Triplicates of samples were collected from roadsides and control of four sites i.e. G.T road, Jalalpur Road, Shahdiwal road, and Sargodha-road of Gujrat Pakistan. Soil and plant samples were analyzed to determine Cu2+, Cd2+, Ni2+ and Cr2+ using AAS (atomic absorption spectroscopy) after acid digestion. Results revealed that the soil and plants of roadside were contaminated with Ni as compared to the control while Cr, Cd, and Cu were absent in both soil and plant samples. The highest Ni concentration, 0.0899 ppm, was found in *Ricinus communis* at site 1, whereas no Ni was detected in the control. Metal pollution reduced photosynthetic pigments and protein in roadside plants compared to control by 29% chlorophyll a, 45% chlorophyll b, 40% carotenoids and 37.86% soluble protein content. While increase in antioxidant activity (CAT, POD, SOD) was observed in roadside plants due to metal stress in order of SOD>POD>CAT. It is concluded that vehicular metal stress shows negative correlation with photosynthetic pigments and total soluble protein content while positive correlation with antioxidant enzymes (Catalase, Peroxidase and Superoxide dismutase) in roadside plants as compared to control. The translocation and accumulation factor concluded that *Calotropis procera* and *Ricinus Communis* had high adaptability factor to grow in metal contaminated sites. It is concluded that vehicular-driven metal pollution, particularly Ni contamination, significantly ($p<0.05$) affects the physiological and biochemical attributes of roadside plants. However, *Calotropis procera* and *Ricinus communis* exhibit considerable tolerance and phytoremediation potential under metal stressed conditions.

Key words: Heavy metals (Cu^{2+}, Cd^{2+}, Ni^{2+} and Cr^{2+}); Roadside Soil; Phytoremediation**;** Pollution assessment

Introduction

Road transport plays a major role in economic and social development by transporting people and goods but in recent years overuse of vehicles has been the major contributor to exacerbating environmental pollution by releasing toxic substances like heavy metals and energy into air, land and water (Ahmed *et al.,* 2016; Jiménez *et al.,* 2018). According to the CEIC (Global Economic Data, Indicators, Charts and Forecasts) report on registered vehicles, Pakistan reported 1,411,226 registered vehicles in 1990, which increased to 7,020,803 by December 2022, marking a 10% annual increase. Due to this, environmental pollution is increasing day by day. Vehicles release pollutants by both exhausts as well as non-exhaust emissions.

Exhaust emission source of vehicle pollutant is fuel combustion which include metalloids, oxides of metals like lead oxide and smokes while the source of non-exhaust emission pollutants are released from clutch, batteries, abrasion of tyre, engine corrosion (McKenzie *et al.,* 2009; Werkenthin *et al.,* 2014; Sobhanardakani, 2019). Many heavy metals released from the wires, tyres, pipes of the vehicles are copper, lead, chromium, iron, cadmium, nickel, manganese and zinc accumulated in the roadside soil, water and vegetation (Ubwa *et al.,* 2013; Adamiec, 2017). Concentrations of heavy metal pollutants have found to be higher in soil, water and vegetation grown near roads as compared to those which are far away from the roads (Lu *et al.,* 2014; Wawer *et al.,* 2015).

One of the major harmful sources of air pollution for humans is vehicular pollution because these gaseous pollutants readily transpired while particle pollutants come in contact with humans near roads thus heavy metal pollution in soil and vegetation has sought global attention (Bernardino *et al.,* 2019).

Soil accumulates heavy metals for a long time, although many heavy metals are mobile that is why the overall pollution level of HMs is higher in soil. The retention of heavy metal pollutants in soil results in change in physio-chemical properties of soil like pH, Electrical conductivity, soil texture, Organic carbon and organic matter. Thus, roadside soil functions as an indicator of vehicular pollution.

There is a complex relationship between plants and heavy metals as plants require trace amounts of heavy metals as nutrient for their growth and physiological

Received: 29-01-2026 | Revised: 05-06-2026 | Accepted: 15-06-2026 | Online: 13-07-2026

functions (Tangahu *et al.,* 2011). Deficiency or high concentration of metals can disrupt the physiological and biochemical functions of plants thus result in stunted growth and death (Nagajyoti *et al.,* 2010). The plant mitigation potential to withstand vehicular emission is regulated by morphological, anatomical and physiological traits of plants. Plants grown along roadside have ability to absorb and accumulate metals in their leaves causing changes in their morphological, physiological and biochemical attributes of leaves (Pathak *et al.,* 2015).

Phytoremediation which is also known as green remediation (Shackira *et al.,* 2021) is the most suitable, low cost and eco-friendly method for the rehabilitation of metal polluted sites by growing different varieties of plants (Raj *et al.,* 2020). It involves the reclamation process by removing, destroying or accumulating the metal pollutants by using different techniques (Patra *et al.,* 2020). Among various remediation techniques, phytoremediation was found to be the least expensive technique for the reclamation of HMs (Ashraf *et al.,* 2019).

According to the CCI (Council of Common Interests) report population growth rate of Pakistan is 2.55% annually due to which vehicle growth rate has also increased to 7,020,803 which ultimately leads to metal pollution. The most widely used fuel in Pakistan is petrol which releases many toxic heavy metals and ultimately sinks in the roadside soil. Due to non-biodegradable nature of heavy metals, they stay in the environment for long period of time and become toxic when their concentration increases beyond a permissible limit. The plants grown along the roadside help to mitigate the metal pollution but little is known about the response of naturally growing roadside herbs and shrubs for the ecological restoration of urban areas because of heavy metal tolerance.

The ability to sequester HMs varies with the species and cultivars (Ali *et al.,* 2013; Mukhopadhyay & Maiti, 2018; Wei *et al*., 2021). Roadside wild plants show competent capability to grow for the ecological restoration of urban areas because of heavy metal tolerance (Rai, 2013). Due to relative tolerance of wild plants to heavy metals, the study was carried out to determine the heavy metal accumulation capability of roadside wild plants. Although numerous studies have reported heavy metal contamination of roadside environments due to vehicular emissions, most investigations have primarily focused on metal accumulation in soil or a limited number of plant species were examined. Furthermore, available studies are concentrated in highly industrialized regions, while limited information is available regarding heavy metal contamination and its physiological effects on naturally growing roadside vegetation in study area. The present study provides a comprehensive assessment of vehicular heavy metal pollution and its impact on naturally growing vegetation along road side. Unlike other studies this research integrates heavy metal analysis of soil and plants with physiological and biochemical indicators. The main objectives of our study were (i) To determine HM pollution in soil and vegetation in proximity to the roadside by potential pollution Indices of different roadsides of Gujrat, (ii) To explore the effect of heavy metal pollution on local vegetation grown along roadside of Gujrat (iii) To select the plant species for biomonitoring and bioremediation of heavy metal released from traffic emissions.

Materials and Methods

Study area: The research was conducted on naturally growing vegetation along roadsides of Gujrat, Pakistan spreading over an area of 3,192 km^2 with 32^0 40'N latitude and 74^0 4'E longitude. Samples were collected from 4 major roads with relatively high traffic density i.e. G.T road, Jalalpur Jattan Road, Shahdiwal road and Sargodha Road were selected. For conducting this research, two sites from each roadside were selected, one was study site that was (0-2 m) distant from road and second one was control which was 50m away from roads and outside the direct influence zone of traffic emissions. The selection of control site at a distance of 50m was based on previous studies demonstrating that concentration of traffic-driven heavy metal decreases substantially with increasing distance from roads (Werkenthin *et al.,* 2014). These sites were suitable for the research of HMs accumulated by plants because, (1) The roads occupy comparatively high traffic density (2) There is no industrial and agricultural area near sampling sites of Gujrat.

Collection and preparation of plant and soil samples: Three independent biological replicates of both soil and plants samples were collected from each sampling site to ensure statistical reliability and account for spatial variability within sites. Plant and soil samples were collected from 4 selected sites of Gujrat including both study and control sites during summer season June and July, 2023 from study site ranges from 0-2m distance from road and control site 50m away from road. Five plant species were collected from each site of Gujrat which were *Calotropis procera* L., *Parthenium hysterophorus* L., *Cynodon dactylon* L, *Cannabis sativa* L. and *Ricinus communis* L. To minimize contamination during sampling, clean stainless-steel tools were used for sample collection. The plant samples were carefully uprooted to obtain intact root and shoot tissue. Plant samples were washed and fresh leaves were stored in polythene bags and kept in freezer for the biochemical analysis while for the determination of heavy metals accumulated in plants, the plant samples were oven dried for 3 days at 70ºC (Abbas *et al.,*2020; Naveed e*t al.,* 2020). Plant roots and shoots were separated for heavy metal determination and the samples were ground with the help of pestle mortar and converted into powder form. About 500g of soil samples were collected from each site with depth of 0-10cm, stored and labeled in polythene bags. Visible plant residues, stones and other debris were removed from soil samples then, air dried, ground with the help of pestle mortar and sieved through 2mm mesh for soil analysis.

Physicochemical analysis of soil: To determine the soil pH 10 g of soil was weighed and 10ml of distilled water was gradually added to make slurry. The soil slurry was covered and kept for saturation at room temperature for 1 hour. The pH meter was calibrated with buffer solution of both pH 7 and pH 10. The pH of the soil was determined by placing the pH tester into the slurry and recorded the measurements after the stabilization of meter (Dudala *et al.,* 2020). To determine the electrical conductivity of soil samples, 10g of soil from each site was weighed and 25ml of distilled water was added. The solution was stirred for 30 seconds and kept it for saturation for about 16 hours. Filtration was done with Buchner funnel having a filter paper by the process of vacuum filtration. The solution was filtered, stirs it again and measured the EC by using EC meter. Walkley & Black (1934) method was used for the determination of soil organic carbon by using titrimetric method, Organic matter was determined by multiplying percentage of organic carbon with 1.724. Water holding capacity of soil was determined by the method described by Ibitoye (2006).

Digestion method of soil: Acid digestion was used for the analysis of heavy metals in soil. Aqua regia (HNO3: HCL=1:3) of 10ml HNO3 and 30ml of HCl was added to 1g of soil. The mixture was shaken vigorously and heated at temperature 380 ºC for 3 hours. After that, the mixture of digestion tubes was allowed to cool at room temperature and solution was made to 50ml by diluting it with distilled water. The solution was filtered by using Whatman's filter paper and filtrate was subjected to glass vials for (Atomic absorption spectrophotometry) AAS for the analysis of heavy metals (Gong *et al.,* 2020).

Determination of chlorophyll and carotenoid contents: The chlorophyll and carotenoid contents were measured by using the method of Arnon (1949). According to this method 0.5g of fresh leaves were grinded in the pestle mortar with the addition of 5ml of 80% acetone. The extract solution was filtered with the help of filter paper and kept at 4ºC for overnight. Then, the solution was centrifuged at 14000rpm for 15 minutes and supernatant was used to measure the absorbance at 645, 663, 510, and 480nm with the help of spectrophotometer:

Chl a = [12.7(OD 663)–2.69 (OD 645)] * V/1000 * W
Chl b = [22.9 (OD 645)–4.68 (OD 663)] * V/1000 * W
Total chl = [20.2 (OD 645)–8.02 (OD 663)] * V/1000 * W
Carotenoids = [7.6(OD480) 1.69(OD510)]* V/1000*W

Determination of protein content: Total soluble protein was determined according to the method given by Bradford (1976). 0.25g of fresh leaf sample was grinded with 10ml of 5mM potassium phosphate buffer of pH 7.8. The sample mixture was centrifuged at 10,000rpm for 15 mins. Then 2ml of Bradford reagent was mixed with the solution and absorbance was recorded by spectrophotometer at 595nm for total soluble proteins.

Determination of antioxidant (SOD, POD, CAT): The activity of SOD was determined by the method of Giannopolitis & Ries (1977). Catalase activity was evaluated according to the method Chance & Maechly (1955). The POD activity was determined by peroxidase guaiacol method.

Heavy metal pollution assessment in plants: For the digestion of plant samples 0.1g of powdered plant sample was taken in 250ml digestion flask followed by the addition of 6ml Nitric acid and kept at 25˚C. The solution was heated at 150˚C until acid was evaporated by placing the digestion flask on hot plate. As the fumes were started appearing then 1ml of hydrogen peroxide was added to each digestion flask. After cooling the samples were cooled down, the solution was made to 50ml by diluting it with water. Then solution was filtered and filtrate was used for the measurement of heavy metals accumulated in plants by using atomic absorption spectrophotometer (Altaf *et al.,* 2021).

To determine the plants efficiency in accumulating and transferring elements from the soil to underground and aerial tissues of plants, bioconcentration factor (BCF), bioaccumulation factor (BAF) and transfer factor (TF) were calculated.

Bioconcentration factor (BCF): The bioconcentration factor of metals in plants was calculated by bioconcentration factor which is the ratio of metals in roots to the concentration of metal in soil.

BCF = Cr (Roots)
Cs (Soil)
Cr represents the metal concentration in roots (mg kg−1), and Cs represents the concentration of metal in the soil (mg kg−1).

Bioaccumulation factor (BAF): The accumulation of metals in plants was calculated by bioaccumulation factor which is the ratio of metals in plants aerial tissue to the concentration of metal in soil.

BAF = Cx (aerial part)
Cs (soil)
Cx and Cs represent the concentrations of metals in the plant aerial tissues and the soil (mg kg−1) (Kumar & Maiti., 2015).

Translocation factor (TF): Translocation factor also known as uptake factor of plants which indicate the transfer rate of metal in plants.

TF = Cx (Shoot)
Cr (Root)
Cx indicate the concentrations of metal in the plant shoots and Cr indicate the concentrations of metal in the plant roots (mg kg^{-1}) (Gupta & Sinha, 2007). The value of translocation factor less than 1 shows plant potential to extract the metal from root into shoot, whereas the value greater than 1 indicate Phytostabilizing property of plant.

Statistical analysis

All statistical analyses were performed using Microsoft Excel. Descriptive statistics i.e. mean, standard deviation, and standard error were calculated for all measured parameters. Data were expressed as mean ± SE of three independent replicates. A three-way analysis of variance (ANOVA) was performed to evaluate the effects of sampling sides, plant species, and road location as well as their interaction, on heavy metal accumulation and other physiological attributes. Tukey's test was used for lettering the bars of the graphs. Statistical significance was accepted at $p \leq 0.05$.

Results

Soil physicochemical characteristics: Physicochemical parameters of soil collected from four roadsides and controls of Gujrat Pakistan were analyzed statistically by mean and standard deviation i.e. pH, EC, OC, OM and saturation percentage is presented in Table 1. pH of soil samples ranged from 7.2-8.2, which is neutral to alkaline. The maximum pH value (8.2) was measured at roadside of site 2 while minimum pH value (7.2) at control site 2. Electrical conductivity of soil ranges from 0.78-3.37 mS/cm. Site 3 and 4 had maximum EC value near roadside compared to control while site 1 and 2 had more EC value in control compared to RS. Roadsides of site 2, 3, 4 had minimum percentage of organic carbon and organic matter compared to control site except site 1 which had minimum percentage in control site compared to control. The O.C and O.M present in roadsides of all sites in the trend of site1>site 4>site 3> site 2. Saturation% of soil ranged from 32-50%. Control soil samples exhibited higher saturation percentage than roadside soil.

Chlorophyll "a" content (mg/g): Fig. 1 depicted that Chlorophyll a content was reduced in roadside plants compared to control due to metal stress. It was found that the chlorophyll a content of plants in every roadside was affected different under vehicular metal stress due to change in traffic density. The highest chlorophyll-a content was found in site 1 with mean value (0.0410) and lowest was found in site 2 with mean value (0.0125). The chlorophyll a concentration in control plants was higher than roadside by 44.58% in site 1, 1.68% in site 2, 65.32% in site 3 and 5.23% in site 4. Among all the plant species in control site highest (mean 0.035) chlorophyll a was found in *C. procera* while lowest (mean 0.026) in *C. sativa* similarly along roadside *P. hysterophorus* shows maximum chlorophyll a content with mean value (0.023) while lowest was recorded in *C. procera* with mean value (0.020).

Chlorophyll "b" content (mg/g): Fig. 2 depicted the impact of vehicular metal stress as chlorophyll b content was reduced in roadside plants as compared to control plants. The chlorophyll-b concentration in control plants was higher than roadside by 36.03% in site 1, 25.81% in site 2, 84.53% in site 3 and 13.31% in site 4. Among all the plant species in control site highest chlorophyll b with means value 0.079 was found in *C. dactylon* while lowest mean value 0.059 was in *R. communis* similarly along roadside *P. hysterophorus* shows maximum chlorophyll-b content with mean value 0.051 while lowest was recorded in *C. procera* with mean value 0.031.

Carotenoids (mg/g): Fig. 3 showed reduction of carotenoids in roadside plants as compared to control due to vehicular metal stress. The carotenoid content was higher in control plants as compared to roadside by 27.06% in site 1, 19.20% in site 2, 73.79% in site 3 and 1.89% in site 4. Among different sites highest carotenoid was found in site 1 with mean value (0.018) while lowest in site 2 with mean value (0.005). Thus, carotenoid content was found in order of site 1> site 3> site 4> site 2. Among all the plant species in control site highest carotenoids (mean 0.015) was found in *C. procera* while lowest (mean 0.014) in *C. dactylon* similarly along roadside *C. sativa* showed maximum carotenoids with mean value (0.010) while lowest was recorded in *C. dactylon* with mean value (0.0078).

Total Chlorophyll content (mg/g): Fig. 4 depicted plants grown along roadside showed reduction in total chlorophyll content compared to control due to vehicular metal stress. Among different sites total chlorophyll concentration in control plants was higher than roadside by 39% in site 1, 16.82% in site 2, 81% in site 3 and 10.26% in site 4. Among all the plant species in control site highest total chlorophyll (mean 0.110) was found in *C. dactylon* while lowest was (mean 0.093) in *R. communis* similarly along roadside *P. hysterophorus* shows maximum total chlorophyll content with mean value (0.075) while lowest was recorded in *C. procera* with mean value (0.051).

Protein content (mg/ml): Fig. 5 depicted that release of heavy metals from vehicular activities lowers the protein content in plants grown along roadside as compared to those growing away from roadside. Total soluble protein content in roadside plants was decreased than control by 61.13% in site 1, 6.75% in site 2, 43.62% in site 3 and 55.14% in site 4 due to metal stress. Maximum reduction in protein content was exhibited by *P. hysterophorus* (1.802) and minimum reduction by *C. sativa* (3.919) along roadside of all sites.

Catalase activity (U/g): Fig. 6 depicted impact of heavy metals from vehicular activities increased the catalase production to overcome stress in plants grown along roadside compared to those growing away from roadside. Catalase production in roadside plants was greater than control by 69.91 % in site 1, 72% in site 2, 30% in site 3 and 66.66% in site 4 due to metal stress. Among different sites maximum catalase activity was found in Site 3 with mean value (0.0306) and lowest was found in site 2 with mean value (0.0192). While site 1 and 4 exhibit minor difference with mean value (0.022) and (0.021) respectively. Among different plant species grown along roadsides maximum catalase production was observed in *R. communis* (0.036) and minimum production in *C. dactylon* (0.029).

Table 1. Means and standard deviation of physicochemical parameters of soil collected from road and control sites of Gujrat, Pakistan.

Sites	pH	EC	OC (%)	OM (%)	Saturation (%)
Site 1 (RS)	7.96 + 0.057c	2.47 + 0.026c	0.412 + 0.005a	0.71 + 0.01a	32+ 1d
Site 1 (Control)	8.16 + 0.057ab	3.37 + 0.02a	0.25 + 0.010c	0.43 + 0.017c	40+ 2c
Site 2 (RS)	8.2 + 0.1a	0.78 + 0.01h	0.197 + 0.010d	0.34 + 0.017d	50+ 2b
Site 2 (Control)	7.2 + 0.1e	1.77 + 0.02e	0.232 + 0.011c	0.4 + 0.02c	46+ 1ab
Site 3(RS)	8.06 + 0.11abc	2.07 + 0.01d	0.244 + 0.005c	0.42 + 0.01c	44+ 2bc
Site 3 (Control)	8.1 + 0abc	0.98 + 0g	0.308 + 0.015b	0.53 + 0.026b	42 + 1.73bc
Site 4 (RS)	7.73 + 0.057d	2.68 + 0.026b	0.296 + 0.005b	0.51 + 0.01b	40 + 1c
Site 4 (Control)	8 + 0bc	1.35 + 0.01f	0.389 + 0.011a	0.67 + 0.02c	46 + 2ab

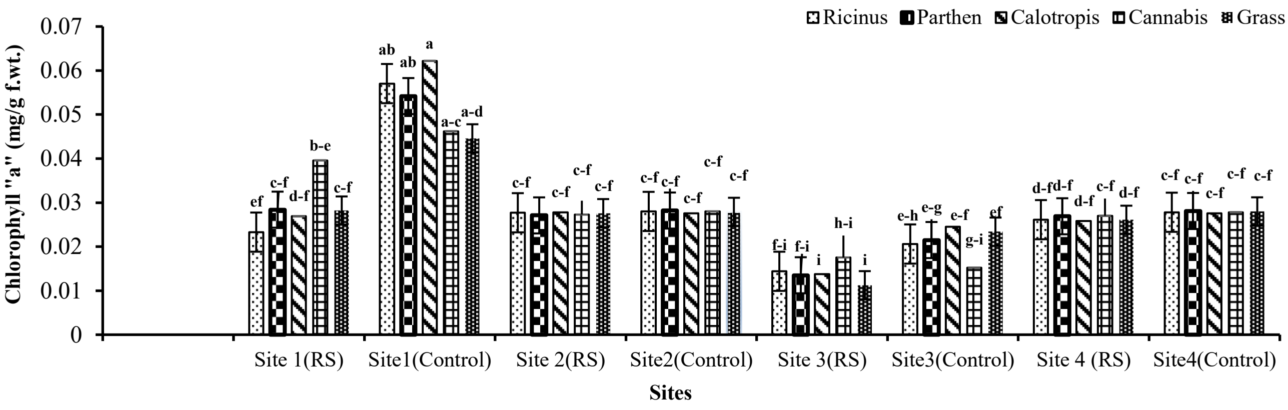


Fig. 1. Comparison of heavy metals on Chlorophyll „a” of five plant species from four sites of roadside and control.

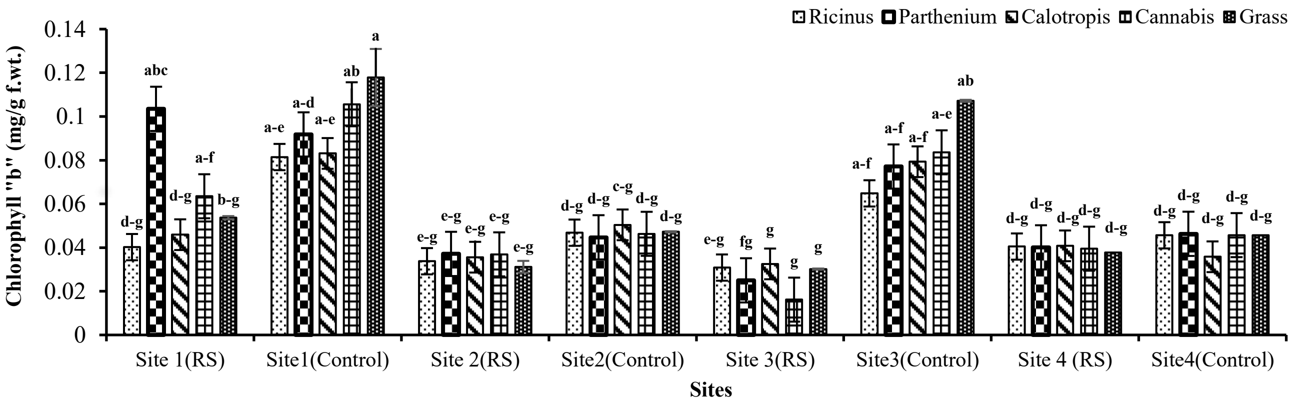


Fig. 2. Comparison of heavy metals on Chlorophyll „b” of five plant species from four sites of roadside and control.

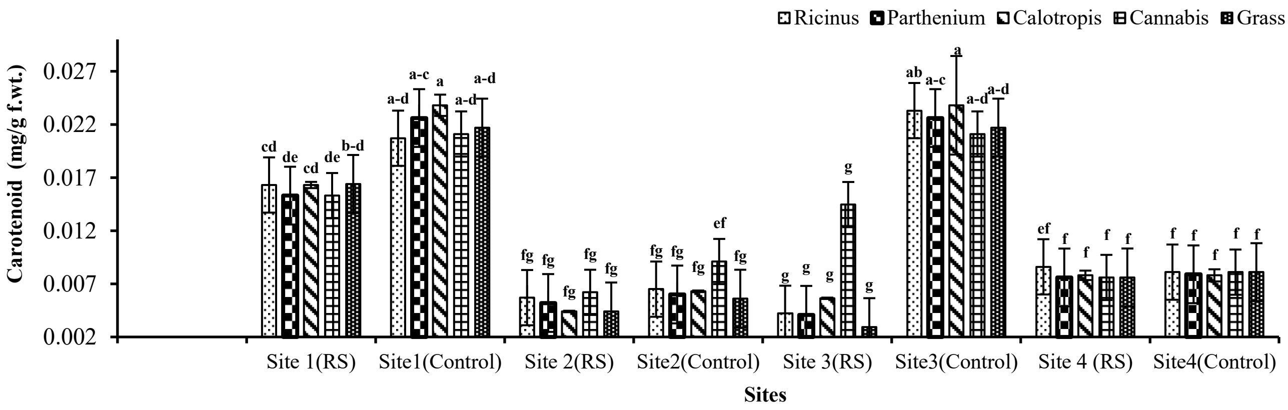


Fig. 3. Comparison of heavy metals on Carotenoids of five plant species from four sites of roadside and control.

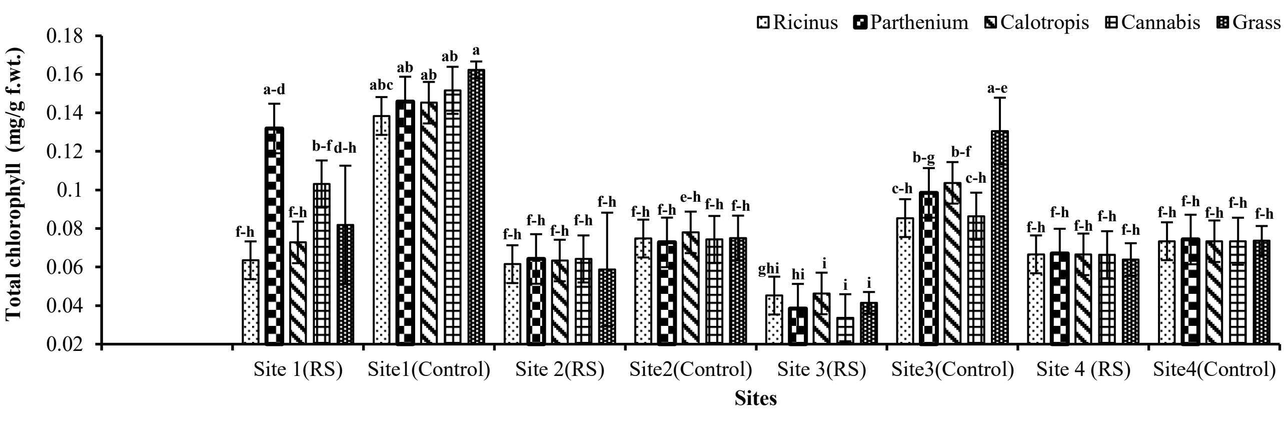


Fig. 4. Comparison of heavy metals effect on total Chlorophyll of five plant species from four sites of roadside and control.

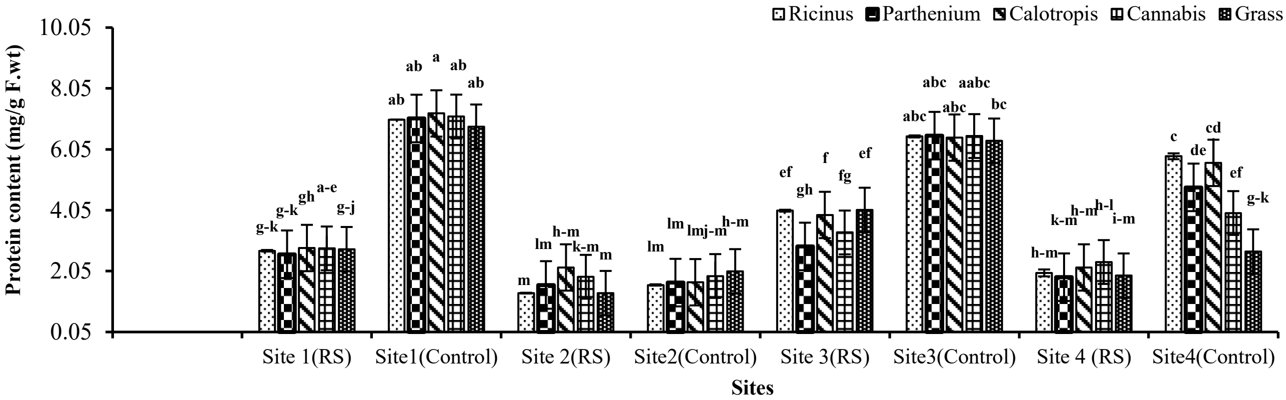


Fig. 5. Comparison of heavy metals effect on total soluble protein content of five plant species from four sites of roadside and control.

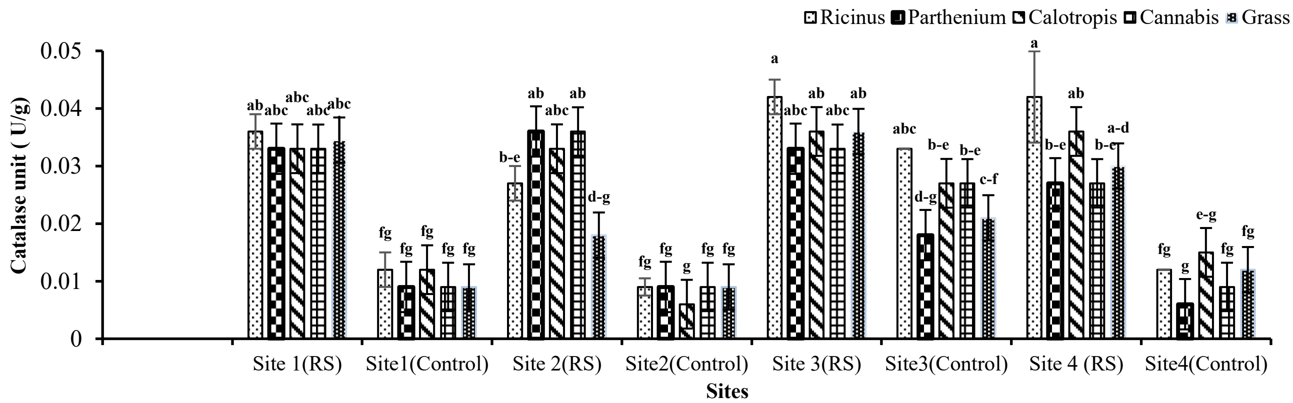


Fig. 6. Comparison of heavy metals effect on catalase activity of five plant species from four sites of roadside and control.

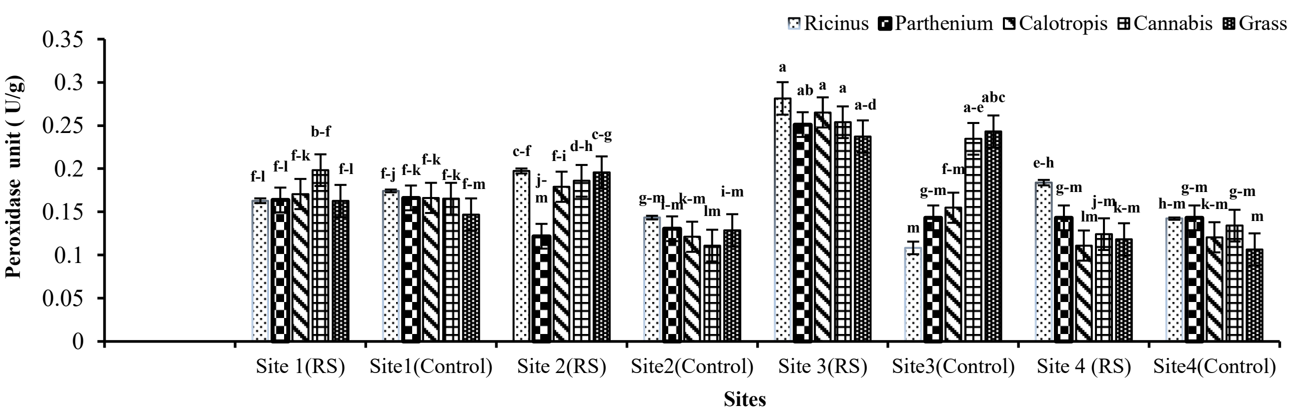


Fig. 7. Comparison of heavy metals effect on peroxidase activity of five plant species from four sites of roadside and control.

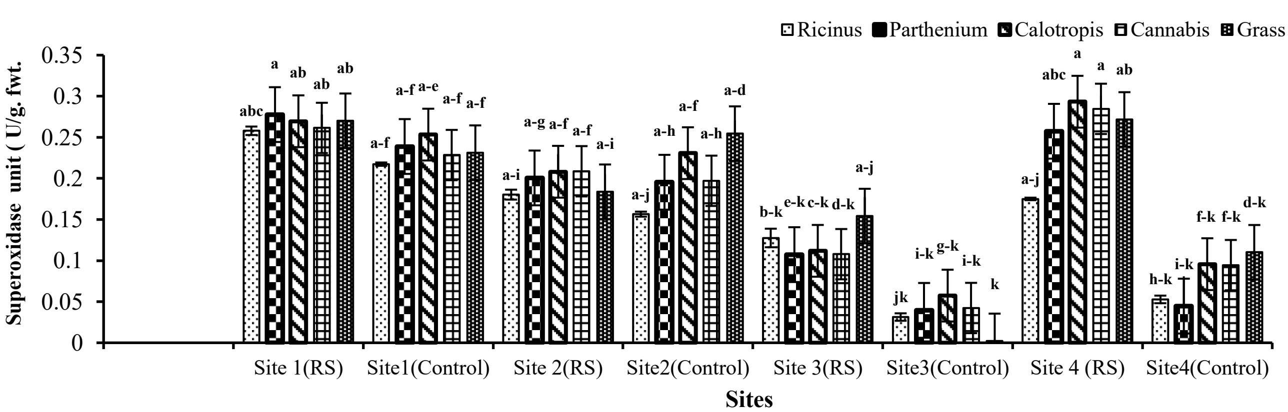


Fig. 8. Comparison of heavy metals effect on superoxide dismutase activity of five plant species from four sites of roadside and control.

Peroxidase activity (U/g): Fig. 7 depicted that release of heavy metals from vehicular activities increase the peroxidase production grown along roadside as compared to control to overcome metal stress. Peroxidase production in roadside plants was increased than control by 4.57% in site 1, 27.85 % in site 2, 31.42% in site 3, and 4.98% in site 4 due to metal stress.

Among different sites maximum peroxidase activity was found in Site 3 with mean value (0.217) and lowest was found in site 4 with mean value (0.132). While site 1 and 2 exhibited minor differences with mean value (0.167) and (0.151) respectively. Among different plants species grown along roadsides maximum peroxidase production was observed in *R. communis* (0.206) and minimum production in *P. hysterophorus* (0.169).

Superoxide dismutase activity (U/g): Fig. 8 depicted that release of heavy metals from vehicular activities increased the superoxide dismutase production in plants grown along roadside compared to control to overcome stress. SOD production in roadside plants was increased compared to control by 12.53% in site 1, 72.38% in site 3, and 68.98% in site 4 due to metal stress while in site 2 5.10% more SOD was observed in control plants compared to roadside. Among all sites maximum SOD activity was found in site 1 with mean value (0.250) and lowest in site 3 with mean value (0.077). Among all plant species maximum SOD production was observed in *C. procera* (0.220) and minimum was in *R. communis* (0.169) along roadsides of all sites.

Heavy metal concentration in soil samples: Out of four heavy metals only nickel was detected in the soil samples of roadside as well as control while non-detection of Cd, Cr, Cu showed the absence of heavy metals in the soil samples. Fig. 9 depicted the comparison of Ni mean concentration present in the soil of roadside and control of Gujrat Pakistan. Maximum nickel concentration was found in site 3 (18.875) and lowest in site 4(1.675). The concentration of Ni in roadside soil sample was higher than control by 72.98% in site 1, 103.55% in site 2, 72.89% in site 3 and 36.58% in site 4.

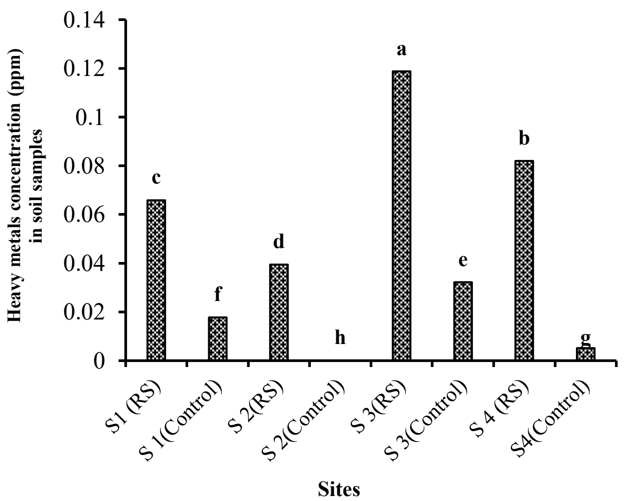
Heavy metals concentration in plant samples: Out of four heavy metals only nickel was detected in all plant samples of roadside as well as control while non-detection of Cd, Cr, Cu showed the absence of heavy metals in the plant samples. The result showed high concentration of nickel in all plant species grown in roadside compared to control due to vehicular activities (Fig. 10). The trend of Ni concentration in plant samples from roadside and control in the order of site 1(0.0408 ppm)> site 4(0.0307ppm)>site 2 (0.0226ppm)> site 3(0.02068ppm)> Control (-0.00512ppm). Highest Ni concentration was found in *R. communis* (0.0899ppm) in site 1 and also lowest in *R. communis* (-0.0231ppm) grown in control. The concentration of nickel in plant species was in the order of *P. hysterophorus*>*C. procera*>*R. communis*>*C. sativa* >*C. dactylon.* Nickel concentration in all plants was below the permissible limit (10ppm) of Ni in plants.

Heavy metal pollution indices of soil: Pollution of Ni heavy metal was calculated by pollution indices of soil. Values of Contamination factor and geo accumulation index indicated that strong contamination of Ni was observed at site 2 while moderate contamination of nickel at site 3 (Tables 2 and 3).

Table 2. Risk assessment of soil by Ni pollution indices.

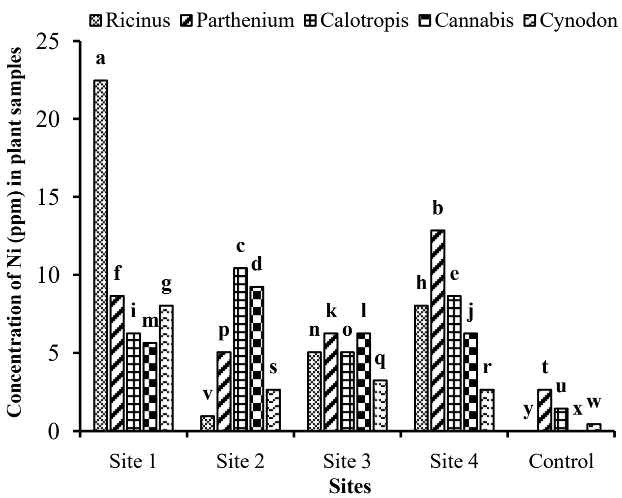
Sites	Metal	Contamination factor	Geo-accumulation index
Site 1	Ni	3.70224	1.303439
Site 2	Ni	28.14286	4.229734
Site 3	Ni	3.689441	1.29844
Site 4	Ni	1.576923	0.07215

Heavy metal pollution indices of plants: Highest Ni mobility from root to shoot was observed in *Calotropis procera* while maximum Ni accumulation among all plant was observed in *Calotropis procera* and *Ricinus communis*).



S1 = Sarghoda Road: **S2** = Jalalpur Jattan Road: **S3** = Shadiwal Road: **S4** = G.T Road

Fig. 9. Comparison of mean concentration of Ni present in soil between roadside and control of Gujrat Pakistan. Different alphabets showed significant difference and similar alphabets shows non-significant difference between roadside and control plant species.



Site 1 = Sarghoda Road: **Site 2** = Jalalpur Jattan Road: **Site 3** = Shadiwal Road: **Site 4** = G.T Road

Fig. 10. Comparison of Ni concentration in plant species of roadside and control of Gujrat Pakistan. Different alphabets showed significant difference and similar alphabets shows non-significant different between roadside and control plant species.

Table 3. Heavy metal pollution indices of plants.

Plant species	Heavy metals	BCF	TF	BAF
Ricinus communis L.	Ni	6.268293	0.626459	3.926829
Parthenium hysterophorus L.	Ni	6.268293	0.019455	0.121951
Calotropis procera L.	Ni	0.41463	10.17647	4.219512
Cannabis sativa L.	Ni	1.878049	1.623377	3.04878
Cynodon dactylon L.	Ni	0.414634	3.117647	1,292683

Discussion

The variation of physicochemical properties of roadside and control soil was due to the metal pollution released from vehicles. pH is the measure of hydrogen ion concentration which indicate the acidic or alkaline nature of soil. The pH values of roadside were found to be alkaline for retention and immobilization of metal ions in soil particles (Rees *et al.,* 2014). Increase in soil pH also increases the metal adsorption on soil due to the competition between protons and metal cation (Werkenthin *et al.,* 2014). Soil components such as clay minerals and organic matter develop more negative charges at higher pH. This increase in negative charges attract positively charged metal ions leading to greater adsorption. The electrical conductivity of three road soils was higher than the control ranging between (2.07-2.68 mS/cm), might be due to high metal retention in soil emitted from vehicles, while one site had EC value 0.78mS/cm. The variation in the soil electrical conductivity (EC) might be caused by heavy metals acting as cations in soil (Khan *et al.,* 2011). An increase in the concentration of metals in soil caused by high electrical conductivity which change the availability of ions (Altaf *et al.,* 2021). Low EC of soil indicates non saline nature of soil meaning that soil particles have poor capacity to hold metal ions (Desavathu *et al.,* 2018). The percentage of organic carbon and matter in our study was less in the road soil compared to the control except site 1 by 12.82% in site 2, 20.75% in site 3 and 23.88% in site 4. Along with other physicochemical parameters soil organic matter also play important role in metal and nutrient retention in soil which ultimately helps plant to grow (Bhatti *et al.,* 2018). The results of Wang & Zhang *et al*., (2018) showed that there was a strong correlation of pH and organic matter with the retention of heavy metals in the soil. A possesses positive correlation occurs between soil organic matter and sorption capacity of metal cation, thus presence of organic matter helps in the soil retention of metal cation in soil (Saeedi *et al.,* 2009).

The present study concluded that vehicular metal stress reduced the photosynthetic pigments (chlorophyll a, b, carotenoids and total chlorophyll) in the plants growing along roadside compared to those grown away from roadside. Many previous studies revealed that content of photosynthetic pigments in plants grown near roadside was reduced due to metal stress released from vehicles (Popova, 2017). Chlorophyll a was more significantly impacted by metal stress than chlorophyll b. Chlorophyll b content was 52% higher in site 1, 21% higher in site 2, 49% higher in site 3, and 33% higher in site 4 than chlorophyll a (mg/g). Among all the photosynthetic pigments carotenoid was more affected than chlorophyll a. The decline of carotenoid was due to presence of metal pollutants causing cellular damage to organelles of plants and production of ROS which cause reduction of light direction to photosystem (Houri *et al.,* 2020; Singh *et al.,* 2023). The order of metal stress on photosynthetic pigments was found in the order of Total chlorophyll> chlorophyll b> chlorophyll a> carotenoids. The vehicle emissions enter into the tissues of plants through stomata and cause denaturation of chloroplast, suppresses chlorophyll synthesis, change the electron transport chain and closing of stomata which results in reduction of photosynthetic pigments in roadside plants as compared to control (Akinci *et al.,* 2010; Pant & Tripathi, 2012). Thus, negative correlation was found between heavy metals and photosynthetic pigments.

Inverse correlation was found between metal stress and soluble protein content in plants growing along roadsides. Significant reduction in protein content was found under metal stress due to structural modification, limited protein biosynthesis or enhanced hydrolysis of protein (Seregin & Kozhevnikova, 2006; Kandziora-Ciupa *et al.,* 2013). Metal pollutants bind with protein synthesizing enzyme function group SH, thus cause change in protein structure (Seregin & Kozhevnikova, 2006). Maximum reduction in protein content was recorded in *P. hysterophorus* (1.802 mg/ml) and minimum reduction in *C. sativa* (3.919 mg/ml) along roadside. Maximum protein reduction was found at site 4 roadside due to heavy traffic at that site.

Present research concluded that there is a positive correlation between metal stress and antioxidant production (CAT, POD and SOD). Metal stress increases the production of catalase, peroxidase and superoxide dismutase as the defensive mechanism due to production of ROS. Catalase is universally accepted oxidoreductase which decompose hydrogen peroxide into water and release molecular oxygen (Lin and Kao, 2000) and Peroxidase enzyme is widely accepted stress enzyme (Gasper, 1991). In plants electrons transfer to chloroplast or mitochondria transfer system, when come in contact with the oxygen molecules produce reactive oxygen species (ROS) (Panda & Rai, 2015) which cause cellular destruction in plants. The plants in response to oxidative damage produce antioxidant enzymes such as catalase, peroxidase which act as scavengers of ROS (Ghorbanli *et al.,* 2007). The production of antioxidant enzymes was found in order of SOD>POD>CAT. Maximum catalase production was observed in *R. communis* (0.036 u/g) and minimum production in *C. dactylon* (0.029 u/g) while maximum peroxidase production was observed in *R. communis* (0.206 u/g) and minimum production in *P. hysterophorus* (0.169 u/g) and maximum SOD production was observed in *C. procera* (0.220 u/g) and minimum production was in *R. communis* (0.169 u/g) along roadside.

The result of this study figured out that mean concentration of nickel in roadside soil was higher than control site due to usage in chrome plating of vehicles

(Ndiokwere, 1984). Ni a micronutrient in plants involved in the enzyme formation urease and other enzymes involved in plant metabolism (Hadayat, 2018) but high concentration in plants cause phytotoxicity (Chen *et al.,* 2010). The maximum Ni concentration in soil was detected at site 3 which was 16.47 mg/kg which was below the permissible limit (50 mg/kg) of Ni (WHO 1996) which indicate the presences of Ni in the soil but not accumulated in the soil. Result of Ni accumulation in roadside soil below the permissible limit is consistent with the results of research conducted in Islamabad, Pakistan (Faiz *et al.,* 2009). The result of AAS showed the absence of Cu, Cr and Cd in the soil samples due to less traffic density in Gujrat.

The soil of roadside was moderately contaminated with Cd and Cu as their value of contamination was 1 while roadside soil was lowly contaminated with Cr as their value of contamination was 0.98. The result of this study revealed the strong contamination of roadside soil with Nickel with mean value 9.27. Geo-accumulation index of roadside soil for Cd, Cr, and Cu was in negative values which showed the absence of this metal contamination in roadside soil which was consistent with the results of Su (2014) who reviewed the metal contamination in the soil on worldwide basis. The geo-accumulation index of Ni in roadside soil was in order of site 2>site 1> site 3> site 4 with the mean values 4.229734, 1.303439, 1.29844 and 0.07215 respectively. Thus, the roadside soil was contaminated with Ni due to release of Ni from automobile chrome plating.

Results of this study figured out that roadside plants were contaminated with Ni in all sites of plants and Cr in plants of one site Gujrat while in all other sites it was not detected. Similarly, Cd, and Cu was not detected in any plant of roadsides due to absence of these metals in soil. The concentration of Ni in roadside plants was greater than control plants which were grown 50m away from roads. The range of Ni accumulated in roadside plants was in between of 0.95-22.45 mg/kg. Several other researches have reported high contamination of roadside plants with Ni in Pakistan (Nawaz *et al.,* 2022). The permissible limit of Ni in roadside plants is 10mg/kg thus Ni accumulated in some roadside plants was above the permissible limit by WHO. Many researchers concluded that exhaust fumes produced due to the combustion of oil were the sources of Ni and Pb in the environment (Johansson *et al.,* 2009; Pey *et al*., 2010; Khan *et al.,* 2011).

Among all the plant species grown along roadside *R. communis* and *P. hysterophorus* tend to accumulate large amount of heavy metals in their tissues. In some previous findings, metal deposition by *R. communis* and *P. hysterophorus* at contaminated site was also reported (Khalid *et al.,* 2023). Our result revealed that each roadside plants have different metal accumulation property from soil thus exhibited different concentrations of metals in their tissue.

Our result concluded that the plants differed in their property of metal mobility and accumulation. Highest metal transfer rate from root to shoot was observed in *Calotropis procera a*nd *Cannabis sativa* as roots possessed less amount of metal compared to shoot. Thus above grounded parts of *C. procera a*nd *C. sativa* above ground part of plant accumulate large amount of metal compared to underground parts. Bothe the plants showed significant variation of metal mobility from root to shoot due to restriction in internal transport system of plants (Chaplygin *et al.,* 2018).

Plant's ability to deposit metal in their aerial parts is clearly reflected by bioaccumulation factor (BAF). All the sites differ in their translocation and bioaccumulation of metal due to change in their physicochemical property of soil. *C. procera* dominated in accumulation of metal in their aerial parts of plants as compared to soil. Thus maximum translocation from root to shoot and accumulation in respective tissues was exhibited by *C. procera.* Bio concentration Factor (BCF) was observed highest in *R. communis* and *P. hysterophorus* as their roots have high tendency to uptake metal from the soil. High BCF at site 4 was recorded due to low pH as low pH of the soil helps in dissolution of metals and enhance their uptake. Thus, bioavailability of metals to roots increases with the decrease of soil pH (Yang *et al*., 2016). The accumulation and translocation of heavy metals varied among plant species and sampling sites, indicating that plant characteristics and soil properties influence metal uptake and distribution. The high bioaccumulation factor observed in *C. procera* suggests that this species possess efficient mechanism for transporting absorbed metals from roots to aerial parts. The highest bioconcertation factor was recorded in *R. communis* and *P. hysterophorous*, reflecting their strong ability to absorb metals from soil through their root systems. From ecological perspective. Species with high BCF and BAF values play an important role in reducing metal concentration in contaminated soils and can serve as effective bio indicators.

Conclusion

The present study demonstrates that roadside soils and naturally growing vegetation serve as important sinks for vehicular heavy metal pollution. Nickel accumulation was consistently higher in road side plants and soils than in their respective controls, indicating the role of traffic-related activities to environmental contamination. The accumulation of Nickel was associated with a significant reduction in photosynthetic pigments and total soluble protein contents, suggesting adverse effects on physiological performance of the plants. In contrast, antioxidant activities (SOD, POD and CAT) increased significantly in roadside plants, indicating the activation of defense mechanism against metal-induced oxidative stress. Among the investigated plant species, *R. communis* and *C. procera* exhibited comparatively higher accumulation and translocation capacities, together with relatively lower physiological impairment under metal stress. These characteristics indicate a greater tolerance to metal pollution and enhanced ability of surviving in polluted environments. Therefore, these species may be considered promising candidates for biomonitoring and phytoremediation of Ni-contaminated roadside ecosystems.

Acknowledgements

The authors extend their appreciation to the Deanship of Research and Graduate Studies at King Khalid University, Saudi Arabia for this work under Large Research Project Under grant number RGP2/205/47.

Author's Contribution: Lasania Mehmood and Khalid Nawaz: Designed the study and conducted field surveys. Khalid Hussain and Noshia Arshad: Co-supervised the study and helped in data collection. Durr-e-Nayab and Manal A. Alshaqhaa: Conducted data analysis and wrote the paper Arooj Khalid and Mounira Mkaddem-Guedri: Helped in graph making including tables and paper writing.

Conflict of Interest: There is no conflict of among the authors for this publication.

References

Abbas, A., M. Azeem, M. Naveed, A. Latif, S. Bashir, A. Ali, M. Bilal and L. Ali. 2020. Synergistic use of biochar and acidified manure for improving growth of maize in chromium contaminated soil. *Int. J. Phytoremed.*, 22(1): 52-61.

Adamiec, E. 2017. Chemical fractionation and mobility of traffic-related elements in road environments. *Environ. Geochem. Health*, 39: 1457-1468.

Ahmed, M.J.K. and M.J. Ahmaruzzaman. 2016. A review on potential usage of industrial waste materials for binding heavy metal ions from aqueous solutions. *J. Water Proc. Eng.*, 10: 39-47.

Akinci, I.E., S. Akinci and K. Yilmaz. 2010. Response of tomato (*Solanum lycopersicum* L.) to lead toxicity: Growth, element uptake, chlorophyll and water content. *Afr. J. Agric. Res.*, 5(6): 416-423.

Ali, H., E. Khan and M.A. Sajad. 2013. Phytoremediation of heavy metals—concepts and applications. *Chemosphere*, 91(7): 869-881.

Altaf, R., S. Altaf, M. Hussain, R.U. Shah, R. Ullah, M.I. Ullah, A. Rauf, M.J. Ansari, S.A. Alharbi, S. Alfarraj and R. Datta. 2021. Heavy metal accumulation by roadside vegetation and implications for pollution control. *PLoS One*, 16(5): e0249147.

Arnon, D.I. 1949. Copper enzymes in isolated chloroplasts. Polyphenoloxidase in *Beta vulgaris*. *Plant Physiol.*, 24(1): 1-15.

Ashraf, S., Q. Ali, Z.A. Zahir, S. Ashraf and H.N. Asghar. 2019. Phytoremediation: Environmentally sustainable way for reclamation of heavy metal polluted soils. *Ecotoxicol. Environ. Saf.*, 174: 714-727.

Bernardino, C.A., C.F. Mahler, R.E. Santelli, A.S. Freire, B.F. Braz and L.A. Novo. 2019. Metal accumulation in roadside soils of Rio de Janeiro, Brazil: Impact of traffic volume, road age and urbanization level. *Environ. Monit. Assess.*, 191: 1-14.

Bhatti, S.S., V. Kumar, A. Kumar, J. Gouzos, J. Kirby, J. Singh, V. Sambyal and A.K. Nagpal. 2018. Potential ecological risks of metal(loid)s in riverine floodplain soils. *Ecotoxicol. Environ. Saf.*, 164: 722-731.

Bradford, M.M. 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal. Biochem.*, 72(1-2): 248-254.

Chance, B. and A.C. Maehly. 1955. Assay of catalases and peroxidases. *Methods Enzymol.*, 2: 764-775.

Chaplygin, V., T. Minkina, S. Mandzhieva, M. Burachevskaya, S. Sushkova, E. Poluektov, E. Antonenko and V. Kumacheva. 2018. Effect of technogenic emissions on heavy metal accumulation by herbaceous plants. *Environ. Monit. Assess.*, 190: 1-18.

Chen, X., X. Xia, Y. Zhao and P. Zhang. 2010. Heavy metal concentrations in roadside soils and correlation with urban traffic in Beijing, China. *J. Hazard. Mater.*, 181(1-3): 640-646.

Desavathu, R.N., A.R. Nadipena and J.R. Peddada. 2018. Assessment of soil fertility status in Paderu Mandal, Visakhapatnam district of Andhra Pradesh through geospatial techniques. *Egypt. J. Remote Sens. Space Sci.*, 21(1): 73-81.

Dudala, S., S.K. Dubey and S. Goel. 2020. Microfluidic soil nutrient detection system: Integrating nitrite, pH and electrical conductivity detection. *IEEE Sensors J.*, 20(8): 4504-4511.

Faiz, M., Tufail, M.T. Javed and M.M. Chaudhry. 2009. Road dust pollution of Cd, Cu, Ni, Pb and Zn along Islamabad Expressway, Pakistan. *Microchem. J.*, 92(2): 186-192.

Gasper, T.H. 1991. Peroxidases in plant growth, differentiation and development processes. In: *Biochemical, Molecular and Physiological Aspects of Plant Peroxidases*. pp. 249-280.

Ghorbanli, M., Z. Bakand and S. Bakand. 2007. Air pollution effects on the activity of antioxidant enzymes in *Nerium oleander* and *Robinia pseudoacacia* plants in Tehran. *J. Environ. Health Sci. Eng.*, 4(3): 157-162.

Giannopolitis, C.N. and S.K. Ries. 1977. Superoxide dismutases: I. Occurrence in higher plants. *Plant Physiol.*, 59(2): 309-314.

Gong, L., D. Qiu, X. Yao and G. Yang. 2020. Determination of heavy metals in plant sample pretreatment methods. *J. Phys. Conf. Ser.*, 1549(2): 022008.

Gupta, A.K. and S. Sinha. 2007. Phytoextraction capacity of plants growing on tannery sludge dumping sites. *Bioresour. Technol.*, 98(9): 1788-1794.

Hadayat, N. 2018. Heavy metal pollution from vehicular emissions and its phytomonitoring along two roads (Pindi Bhattian–Kala Shah Kaku and Lahore–Gujranwala). Ph.D. Diss., Univ. Agric., Faisalabad, Pakistan.

Houri, T., Y. Khairallah, A. Al Zahab, B. Osta, D. Romanos and G. Haddad. 2020. Heavy metals accumulation effects on the photosynthetic performance of geophytes in Mediterranean reserve. *J. King Saud Univ. Sci.*, 32(1): 874-880.

Ibitoye, A.A. 2006. *Laboratory Manual on Basic Soil Analysis*. Foladave Nigeria Ltd., Ibadan, Nigeria, pp. 16-36.

Jiménez, M.D., R. De Torre, I. Mola, M.A. Casado and L. Balaguer. 2018. Local plant responses to global problems: *Dactylis glomerata* responses to different traffic pollutants on roadsides. *J. Environ. Manage.*, 212: 440-449.

Johansson, C., M. Norman and L. Burman. 2009. Road traffic emission factors for heavy metals. *Atmos. Environ.*, 43(31): 4681-4688.

Kandziora-Ciupa, M., R. Ciepał, A. Nadgórska-Socha and G. Barczyk. 2013. A comparative study of heavy metal accumulation and antioxidant responses in *Vaccinium myrtillus* L. leaves in polluted and non-polluted areas. *Environ. Sci. Pollut. Res.*, 20: 4920-4932.

Khalid, N., A. Noman, A. Nazir, A. Tufail, N. Hadayat, F.M. Alzuaibr, S. Ikram, N. Akhter, M. Hussain and M. Aqeel. 2023. *Nerium oleander* could be used for sustainable management of traffic-borne elemental-enriched roadside soils. *Environ. Sci. Pollut. Res.*, 30(14): 40551-40562.

Khan, M.N., A.A. Wasim, A. Sarwar and M.F. Rasheed. 2011. Assessment of heavy metal toxicants in roadside soil along the N-5 National Highway, Pakistan. *Environ. Monit. Assess.*, 182: 587-595.

Khan, S., M.A. Khan and S. Rehman. 2011. Lead and cadmium contamination of roadside soils and plants in Peshawar City, Pakistan. *Pedosphere*, 21(3): 351-357.

Kumar, A. and S.K. Maiti. 2015. Assessment of potentially toxic heavy metal contamination in agricultural fields, sediment and water from an abandoned chromite-asbestos mine waste of Roro Hill, India. *Environ. Earth Sci.*, 74: 2617-2633.

Lin, C.C. and C.H. Kao. 2000. Effect of NaCl stress on H_2O_2 metabolism in rice leaves. *Plant Growth Regul.*, 30: 151-155.

Lu, X., L. Li, N. Zhao, D. Shang and R. Li. 2014. Concentration, speciation and environmental risk of heavy metals in dusts from different functional areas of a medium-sized city in China. *Int. J. Environ. Eng.*, 1(4): 1-5.

McKenzie, E.R., J.E. Money, P.G. Green and T.M. Young. 2009. Metals associated with stormwater-relevant brake and tire samples. *Sci. Total Environ.*, 407(22): 5855-5860.

Mukhopadhyay, S. and S.K. Maiti. 2018. Techniques for quantitative evaluation of mine site reclamation success. In: *Bio-geotechnologies for Mine Site Rehabilitation*. Elsevier, pp. 415-438.

Nagajyoti, P.C., K.D. Lee and T.V.M. Sreekanth. 2010. Heavy metals, occurrence and toxicity for plants: A review. *Environ. Chem. Lett.*, 8: 199-216.

Naveed, M., H. Sajid, A. Mustafa, B. Niamat, Z. Ahmad, M. Yaseen, M. Kamran, M. Rafique, S. Ahmar and J.T. Chen. 2020. Alleviation of salinity-induced oxidative stress through calcium-fortified composted animal manure in canola. *Sustainability*, 12(3): 846.

Nawaz, R., M. Aslam, I. Nasim, M.A. Irshad, S. Ahmad, M. Latif and F. Hussain. 2022. Air pollution tolerance index and heavy metals accumulation of tree species in Lahore. *Air*, 1(1): 55-68.

Ndiokwere, C.L. 1984. Heavy metal pollution from motor vehicle emissions and its effect on roadside soil and vegetation. *Environ. Pollut. Ser. B*, 7(1): 35-42.

Panda, L. and P.K. Rai. 2015. *Roadside Plants: Study on Eco-sustainability*. LAP Lambert Academic Publishing.

Pant, P.P. and A.K. Tripathi. 2012. Effect of lead and cadmium on morphological parameters of *Syzygium cumini* seedlings. *Ind. J. Sci.*, 1(1): 29-31.

Pathak, R.K., C. Tomar and S. Mahajan. 2015. Phytomonitoring of atmospheric pollution in roadside perennial trees of Indore city. *Int. J. Adv. Eng. Technol.*, **7**(6): 1727.

Patra, D.K., C. Pradhan and H.K. Patra. 2020. Toxic metal decontamination by phytoremediation approach. *Environ. Technol. Innov.*, 18: 100672.

Pey, J., X. Querol and A. Alastuey. 2010. Discriminating regional and urban contributions in PM levels. *Atmos. Environ.*, 44(13): 1587-1595.

Popova, E. 2017. Impact of heavy metals on photosynthetic pigment content in roadside plant communities. *AIP Conf. Proc.*, 1899(1): 1-6.

Rai, P.K. 2013. Environmental magnetic studies using roadside plant leaves. *Atmos. Environ.*, 72: 113-129.

Raj, D., A. Kumar and S.K. Maiti. 2020. *Brassica juncea* for phytoremediation of mercury contaminated soils. *Int. J. Phytoremediation*, 22(7): 733-744.

Rees, F., M.O. Simonnot and J.L. Morel. 2014. Short-term effects of biochar on soil heavy metal mobility. *Eur. J. Soil Sci.*, 65(1): 149-161.

Saeedi, M., M. Hosseinzadeh, A. Jamshidi and S.P. Pajooheshfar. 2009. Assessment of heavy metal contamination in highway side soils. *Environ. Monit. Assess.*, 151: 231-241.

Seregin, I. and A.D. Kozhevnikova. 2006. Physiological role of nickel and its toxic effects on plants. *Russ. J. Plant Physiol.*, 53: 257-277.

Shackira, A.M., K. Jazeel and J.T. Puthur. 2021. Phycoremediation and phytoremediation. In: *Sustainable Environmental Clean-Up*. Elsevier, pp. 273-293.

Singh, D.V., J.I.A. Bhat, R.A. Bhat and J.A. Tali. 2023. Vehicular emission impact on heavy metal accumulation in pine needles. *Environ. Sci. Pollut. Res.*, 30(9): 23898-23907.

Sobhanardakani, S. 2019. Ecological and human health risk assessment of atmospheric heavy metals. *Biol. Trace Elem. Res.*, 187(2): 602-610.

Su, C. 2014. Heavy metal contamination in soil worldwide. *Environ. Skept. Critics*, 3(2): 24-30.

Tangahu, B.V., S.R.S. Abdullah, H. Basri, M. Idris, N. Anuar and M. Mukhlisin. 2011. Heavy metal uptake by plants through phytoremediation. *Int. J. Chem. Eng.*, 2011: 939161.

Ubwa, S.T., J. Abah, C.A. Ada and E. Alechenu. 2013. Heavy metals contamination of street dust in Jos metropolis. *J. Biodiv. Environ. Sci.*, 3(7): 13-21.

Walkley, A. and I.A. Black. 1934. Determination of soil organic matter. *Soil Sci.*, 37(1): 29-38.

Wang, M. and H. Zhang. 2018. Accumulation of heavy metals in roadside soil. *Int. J. Environ. Res. Public Health*, 15(6): 1064.

Wawer, M., T. Magiera, G. Ojha, E. Appel, G. Kusza, S. Hu and N. Basavaiah. 2015. Traffic-related pollutants in roadside soils. *Water Air Soil Pollut.*, 226: 1-14.

Wei, Z., Q. Van Le, W. Peng, Y. Yang, H. Yang, H. Gu, S.S. Lam and C. Sonne. 2021. Phytoremediation of contaminants in air, water and soil. *J. Hazard. Mater.*, 403: 123658.

Werkenthin, M., B. Kluge and G. Wessolek. 2014. Metals in European roadside soils. *Environ. Pollut.*, 189: 98-110.

Yang, R., F. Guo, J. Li, N. Su, Z. Shao and S. Zan. 2016. Effect of copper tolerant *Elsholtzia splendens* on bacterial community. *J. Environ. Sci.*, 46: 165-173.