Comparative Physiological and Biochemical Responses of Spinach and Tomato to Roadside Environmental Stress in Sulaimani

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Abstract

This study examined the effects of roadside heavy-metal contamination on the physiological and biochemical responses of spinach (Spinacia oleracea L.) and tomato (Solanum lycopersicum L.) in Sulaimani, Kurdistan Region, Iraq. the former a roadside habitat that has been highly exposed to vehicular fumes and the latter a farmland that is quite remote to the vehicular emissions and therefore has minimal contamination.

Inductively Coupled PlasmaOptical Emission Spectrometry (ICPOES) was used to determine the lead (Pb) and cadmium (Cd) in soil and leaf samples. Measures of physiological parameters such as chlorophyll content in terms of SPAD, chlorophyll fluorescence (Fv/Fm), antioxidant enzyme superoxide dismutase (SOD) activity, measurement of lipid peroxidation through malondialdehyde (MDA) levels and soluble sugar content were measured to determine the response of plants to stress.

The findings found that there were considerably greater levels of Pb and Cd in the soils and plant tissues at the roadside location as compared to the remote agricultural location. Increased levels of metal were correlated with significant decreases in SPAD and Fv/Fm values which are indicative of a photosynthetic inhibition. At the same time the SOD activity and MDA levels were elevated which indicates the activation of antioxidants defenses and oxidative stress. Under roadside conditions, soluble sugar concentrations were reduced significantly, which supported the fact of metabolic disruption.

The accumulation of Pb and Cd and oxidative response towards heavy metal were higher in spinach than in tomato, indicating spinach was more sensitive to heavy metal stress and a better bioindicator species. The results highlight the relevance of environmental monitoring program that includes physiological parameters and suggest avoiding the cultivation of leafy vegetables near high-traffic roads.

Keywords : Heavy metals; Spinach; Tomato; Lead; Cadmium; SOD; MDA.

Introduction

One of the most nagging environmental challenges related to the sustainability of agriculture and human health is the contamination of the environment with heavy metals. The lack of heavy metals in arable soils around the world has been a result of the increasing urbanization, vehicular emissions, industrial effluents, and the use of untreated wastewater to irrigate arable soils **[1], [2]**. Toxic

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elements, especially the lead (Pb) and cadmium (Cd), are not biodegradable and may be used by plants, thus, entering the food chain and causing ecological risks in the long term **[3]**.

The most sensitive agricultural commodities related to the uptake of heavy-metals are vegetable crops because of high growth rate and a large area of surface cover. The concentration of metals in the edible parts of leafy vegetables is higher than in fruiting or root crops **[4], [5]**. Spinach (Spinacia Spinacea L.) has been considered as a sensitive species to monitor heavy-metal contamination due to its high rates of transpiration and due to the fact that it directly releases foliar deposition of the expelled particles of the air, making tomato (Solanum lycopersicum L.) a moderately tolerant fruiting crop that can be used as a comparative physiological variable **[6], [7]**.

Pb and Cd exposure trigger numerous physiological and biochemical disruptions in plants, such as the inhibition of chlorophyll biosynthesis, decreased the efficiency of photosystem II, and the increased production of reactive oxygen species (ROS) **[8]**. Plants counteract such stress by the activation of antioxidant enzymes, including superoxide dismutase (SOD), catalase (CAT), and peroxidase (POD) and by accumulation of osmolytes and their membrane repair mechanism **[9], [4]**. The indicators of these changes are physiological parameters, which include (SPAD) chlorophyll content, (Fv/Fm) chlorophyll fluorescence, (MDA) lipid peroxidation, and (soluble sugar content), which can be used to obtain the information on the stress tolerance of plants **[10]**.The Sulaimani area of the Kurdistan Region in Iraq is a good example of agro-urban landscape whereby agricultural fields are often situated along major roads. The constant traffic emissions and deposition of the dust exposes the soil and crops to a higher risk of accumulating Pb and Cd. Nevertheless, there is less evidence on the physiological effects of such contamination on the locally grown vegetables. The scientific basis of this study is also supported by the recent studies that have determined the effect of heavy metals and oxidative stress in different biological systems. They include the influence of lead and molybdenum on promoting oxidative stress **[11]**, the comprehensive profiling of active compounds and antioxidant abilities in medicinal plants **[12]**, and that effects of selenium supplementation on fundamental and detrimental components and their relationships with the environment **[13]**. Together, these works provide more evidence, which is used to explain physiological and biochemical stress responses that can be seen in plants that are exposed to lead and cadmium.

In this regard, the paper examines the correlation between roadside heavy-metal pollution and the physiological activity of spinach and tomato that were planted in two opposite locations; a roadside location and a remote farmland. The relevant objectives are to:

1. Establish the concentrations of Pb and cadmium (Cd) in soils, as well as in plant leaves in the two locations.
2. Assess the impact of the exposure to heavy-metals on chlorophyll content, photosynthetic efficiency, indicators of oxidative stress, and soluble sugars.
3. Compare spinach and tomato response to determine species-specific tolerance as well as whether they can be used as bioindicators to monitor the environment.

The research findings are likely to add to the knowledge of the physiology of heavy-metal stress and also will provide the baseline of information to justify safer urban agriculture operation in Sulaimani area.

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Material and Methods

Selection of Study Area and Site.

In the current field study, the researcher conducted a field study in the Sulaimani Governorate, Kurdistan Region of Iraq (35°33′N, 45°25′E). The region has a semi-arid Mediterranean climate, and the average annual temperature is about 17 o C and the yearly rainfall is about 600 mm. Two arable sites (different in their distance to major thoroughfares) were chosen to model the opposite levels of exposure to vehicular emissions and the related heavy-metal deposition. The roadside site was located approximately 5–10 m from a major paved highway, whereas the distant site was located approximately 500 m from the road, so it could be used as a reference with minimal anthropogenic contamination. The sites were similar in terms of their soil texture, irrigation and history of crop cultivation which were carefully selected to reduce confounding effects of the environment. The sampling was performed in the spring growing period (April May 2025) when the meteorological conditions were similar. Geographic data and elevation were taken with a garmin eTrex 32x GPS gadget.

Plant Material

Two economically valuable vegetable species that grow in Sulaimani area were chosen according to the following criteria: (1) Spinacia oleracea L. (spinach), a leafy vegetable, and (2) Solanum lycopersicum L. (tomato), which is a fruiting crop. The selection of these crops was based on the fact that they are commonly cultivated in the study area and have a clear physiological response to the exposure of heavy-metals.

Sampling Design and Replication.

The study followed a 2 × 2 factorial arrangement within a completely randomized design, with Site (roadside and distant) and Species (spinach and tomato) as the two experimental factors, Five independent biological replicates were collected for each Site × Species combination, resulting in a total of 20 plant samples. which were Site (RS, DS) and Species (spinach, tomato). Five sample plants (n = 5) of each species were picked up in each site and each sample plant was obtained in a different subplot of 10x10m to capture the within-field variation. All the physiological and biochemical variables were triated in all the biological replicates and the mean value thus used in analysis. The overall sample size of the plants was hence 1 species 2 sites 5 replications = 20.

Sampling and Preparation of Soils.

Out of every subplot, 5 subsamples of rhizosphere soil (0 20 cm deep) were taken with an auger made of stainless steel. The subsamples were mixed, dried at ambient temperature, ground gently, sieved with mesh diameter of 2mm, and kept in closed polyethylene bags. Physicochemical investigation of the soil followed the following: pH was measured using a digital pH meter (Hanna instruments) in a 1:2.5 (soil:water) suspension, electrical conductivity (EC) was measured using an EC meter (HANNA HI 993310) and organic matter (OM) content was determined using the WalkleyBlack method.

Heavy Metal Concentrations Determinations.

The concentration of lead (Pb) and cadmium (Cd) in soils and plant leaves was measured by Inductively Coupled Plasma–Optical Emission Spectrometry (ICP–OES) (PerkinElmer Avio⁻ 550) after acid digestion. The procedure to digest soil entailed the 1.0g of soil was

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added with 10mL of HNO3:HClO4 (3:1) and the mixture was heated to 150 o C until a clear solution was obtained, then it was filtered through Whatman No. 42 filter paper and diluted to 25mL. A 0.5 g of dried leaf powder had to be used in leaf digestion, after which the solution was which was mixed with HNO 3:H 2 O 2 (4:1) was subjected to low temperature until it was completely digested, filtered and subsequently diluted to 25 mL. Certified multi-element standards (Merck, Germany) were used as a calibration. Analytical precision and accuracy were provided by procedural blanks, duplicates and reference materials (NIST SRM 2711a).

Physiological and Biochemical Measures.

Chlorophyll Content (SPAD)
The youngest fully expanded leaf was used to assess the in situ leaf chlorophyll content using a SPAD-502 Plus chlorophyll meter (Konica Minolta, Japan). Each replicate was measured three times on each leaf.

Phytoplankton Chlorophyll Fluorescence (Fv/F m)
Photosystem II (Fv/Fm) was measured using a PAM -2500 fluorometer (Heinz - Walz, Germany) after 20min dark adaptation.

Antioxidant Enzyme Activity (SOD)
The activity of the superoxide dismutase (SOD) was determined according to [14] depending on the reduction of nitroblue tetrazolium (NBT) photoreduction. The expression of activity was described as U g -1FW with one unit representing 50 percent inhibition of NBT reduction. One unit of SOD activity was defined as the amount of enzyme required to cause 50% inhibition of NBT photoreduction. SOD inhibition was calculated as: Inhibition (%) = [(Acontrol − Asample)/Acontrol] × 100.

Lipid Peroxidation (MDA)
The thiobarbituric acid (TBA) technique was used to measure the level of malondialdehyde (MDA) content, which is a sign of lipid peroxidation of the membrane. The absorbance was measured at 532nm (corrected at 600nm) using a UV-Vis spectrophotometer (Shimadzu UV -1900). MDA concentration was calculated from the difference between absorbance at 532 and 600 nm using an extinction coefficient of 155 mM−1 cm−1.

Soluble Sugar Content
The soluble sugars were measured using the anthrone sulfuric acid method at 625nm, which involved the use of 80 percent ethanol to extract the soluble sugars and measured in mg gram per 1 gram of the fresh weight. A glucose standard curve was prepared, and soluble sugar concentration was calculated from the regression equation and expressed as mg g−1 fresh weight.

Quality Control and Assurance.
Glasses were acid-washed (10 per cent HNO 3) and washed with deionised water followed by drying in an oven. Calibration curves were found to have an R 2 of not less than 0.999. There were 10 per cent analytical blanks and duplicates. The standard deviation was threefold the mean of the blank which was called method detection limits (MDLs). Replicate variability was less than 5 percent coefficient of variation (CV).

Statistical Analysis
Analysis of data was done using XLSTAT 2026 and Microsoft Excel 2026. The replication plan consisted of five biological replications, and three technical replications of each parameter. The descriptive statistics were in the form of mean which implied standard deviation (SD). Normality and homogeneity of

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variance were tested by use of Shapiro-Wilk and Levene test respectively. The effects of Species (spinach, tomato), Site (RS, DS) and the interaction between them (Species × Site) were analyzed using a two-way analysis of variance (ANOVA). The p-value used was 0.05 where the honestly significant difference (HSD) test was used. Effect sizes were quantified using partial η 2. Pearson correlation coefficients were determined to determine relationships between physiological parameters and heavy- metals concentrations. After the z -score standardisation of variables, principal component analysis (PCA) was performed to visually depict multivariate tendencies between variables. Tukey's HSD test was used for multiple mean comparisons because it controls the overall Type I error among pairwise comparisons.

Health and Safety and Ethical Conformity.

Prior consent of landowners was obtained in all the sampling procedures. The tasks in the laboratory, which included the use of concentrated acids, were done under the vents in the fume hood and with proper personal protective equipment (PPE). Acyclovite was neutralised using waste acids before disposing it in accordance with the institutional environmental safety requirements.

Results and Discussion

Heavy Metal in Soils and Leaf of Plants.

The levels of lead (Pb) and cadmium (Cd) had strong spatial difference between the two study sites (Figure 1, 2). Pb and Cd showed significant concentrations in soils and leaf samples gathered in the roadside location (RS) than in the remote agricultural location (DS). Spinach (Spinacia oleracea) had more accumulation of the two metals than the tomato (Solanum lycopersicum) due to its leafy structure and higher rate of transpiration which allows the metals to be deposited to the atmosphere and uptaken by soil. On average, spinach leaves on the roadside location contained about two times the amount of Pb compared to tomato and Cd was also the same. On the other hand, the two metals were kept close to the background levels at the remote location. These results support the idea that the major contributors to metal contamination of soil and plants along the roadside in the Sulaimani region are vehicular emissions and dust deposition.

Chlorophyll Content and Efficiency of Photosynthesis.

Site and species affected photosynthetic performances (physiological indicators such as chlorophyll content (SPAD values) and maximum quantum yield of PSII (Fv/Fm) significantly (Figure 3, 4). The two parameters reduced significantly in the plants sampled at the roadside location than at the remote farmland. The average SPAD values in both spinach and tomato leaf declined by about 25-30 percent during roadside exposure which also means the loss of chlorophyll pigments as well as the effect of Pb and Cd on the magnesium and iron metabolism in the chloroplasts. Equally, Fv/Fm ratio decreased atypical optimal values (0.83) down to below 0.70 in plants in the roadside, which showed photoinhibition and loss of photosystemII efficacy to oxidative stress. Despite the fact that the two crops had been affected, tomato had higher SPAD and Fv/Fm values in comparison to spinach indicating a rather more robust photosynthetic stamina under the heavy-metal stress.

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Antioxidant Enzyme Activity (SOD)

Superoxide dismutase (SOD) is a major antioxidant enzyme and its activity also significantly increased ($p < 0.01$) in both the species that were cultivated close to the road (Figure 5). SOD activity increase is also an adaptive mechanism to increase the levels of reactive oxygen species (ROS) produced during the presence of heavy-metal stress. Spinach leaves had better SOD activity compared to tomato which showed that it has a stronger biochemical defense mechanism. Nevertheless, this did not result in complete mitigation of oxidative damage effect, as indicated by the elevated levels of lipid peroxidation.

Lipid Peroxidation (M.D.A Content)

The level of malondialdehyde (MDA) which is a marker of lipid peroxidation and damage to the membrane was significantly increased in plants that were subjected to roadside environments (Figure 6). In spinach and tomato, the MDA grew at least two times compared with the far control site ($p < 0.01$). Such increase implies that Pb and Cd accretion caused significant oxidative stress, which disrupted cellular membranes and caused metabolic imbalance. Spinach showed a high response in terms of MDA as compared to tomato, which justifies the identification of spinach as more sensitive to the vehicular heavy-metal pollution.

Soluble Sugar Content

There was the opposite trend in the levels of soluble sugars as compared with the oxidative-stress indicators (Figure 7). The soluble sugar content in both spinach and tomatoes grown along the road were much lower than the soluble sugar content of spinach and tomatoes at the remote location ($p < 0.05$). Photosynthetic activity is attenuated, and this is accompanied by the possible repositioning of carbohydrate metabolism with stress-response mechanisms, reflected in this degradation. Tomato retained more sugar content than spinach in both conditions that could be a factor in its relatively higher tolerance and physiological stability.

Correlations and Multivariate Tests.

The correlation analysis performed by Pearson showed that there were strong negative correlations between the concentrations of Pb and Cd and photosynthetic parameters (SPAD, Fv/Fm), and positive correlations between the concentrations of SOD and MDA. The soluble sugars were inversely linked with the two heavy metals ($r= -0.82$ -0.89, $p=0.01$). These correlations demonstrate that there is a distinct trade-off between oxidative stress and photosynthetic activity in the presence of heavy-metal. The Principal Component Analysis (PCA) indicated that there was a clear division between samples of roadside and distant locations along the first principal component (PC1), which accounted over 90 per cent of the entire variance. The clustering of roadside samples was high in terms of high Pb, Cd, SOD as well as MDA and far samples were linked to high SPAD, Fv/Fm as well as sugar content. The biplot also indicated the differentiation of species where tomato was placed between the two clusters, hence it is semi-tolerant.

Summary of Findings

In general, the results show that:

Soils and plants surrounding the roadways have significantly increased levels of lead and cadmium.

Increased levels of heavy-metals were also found to suppress the production of chlorophyll and lower the activity of photosystem II.

Compensatory responses such as oxidative stress indicators such as superoxide dismutases activity and malondialdehyde were induced.

The spinach was found to be more sensitive and tomato rather more physiologically resilient.

These results prove that the distance to the roads is a significant factor in the physiological and biochemical reaction of the vegetable crops that grow in the Sulaimani region and may be used as a bioindicator of heavymetal pollution at urbanagricultural interface.

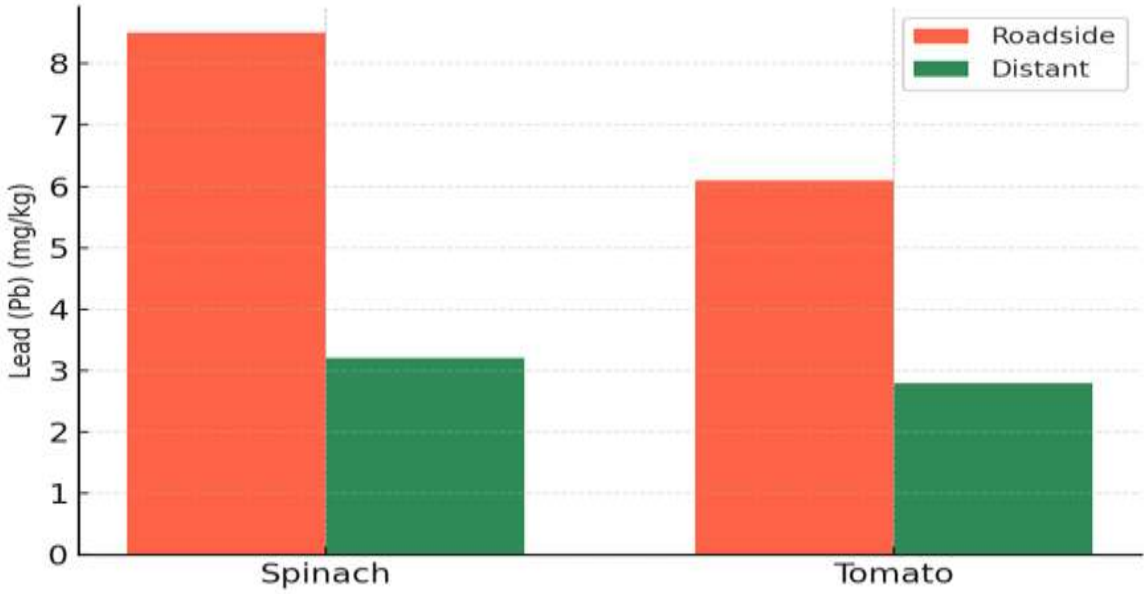


Figure 1. Lead (Pb) concentrations in leaves of spinach and tomato at roadside and distant sites.

Values are presented as mean ± SD (n = 5). Different letters indicate significant differences according to Tukey's HSD test at $p < 0.05$.

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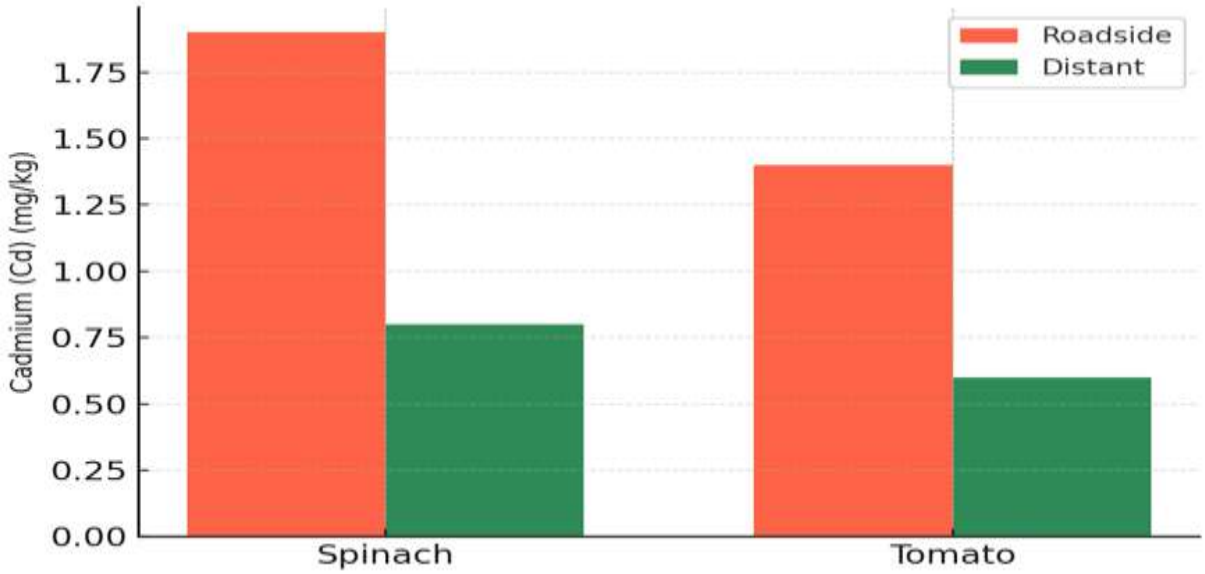


Figure 2. Cadmium (Cd) concentrations in leaves of spinach and tomato at roadside and distant sites.

Values are presented as mean ± SD (n = 5). Different letters indicate significant differences according to Tukey's HSD test at $p < 0.05$.



Figure 3. Chlorophyll content (SPAD values) of spinach and tomato under roadside and distant conditions.

Values are presented as mean ± SD (n = 5). Different letters indicate significant differences according to Tukey's HSD test at $p < 0.05$.

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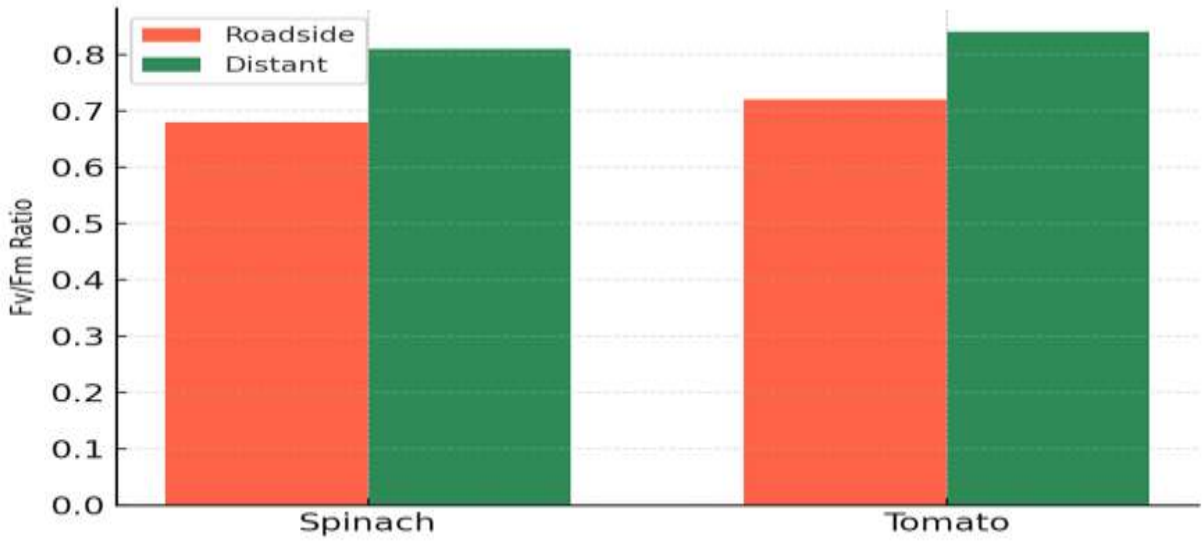


Figure 4. Photosystem II efficiency (Fv/Fm ratio) of spinach and tomato grown at different distances from the road.

Values are presented as mean ± SD (n = 5). Different letters indicate significant differences according to Tukey's HSD test at $p < 0.05$.

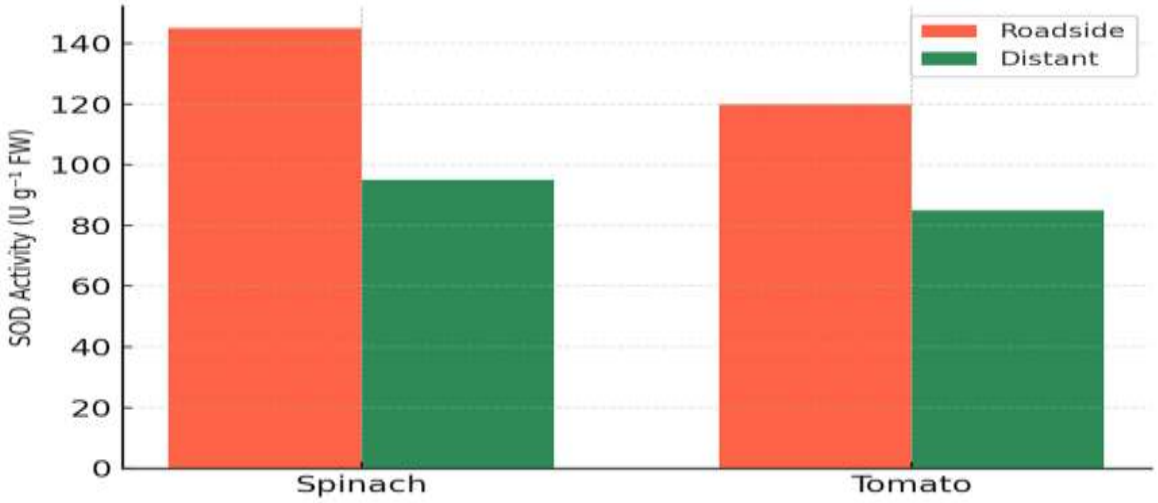


Figure 5. Superoxide dismutase (SOD) activity in leaves of spinach and tomato at roadside and distant sites.

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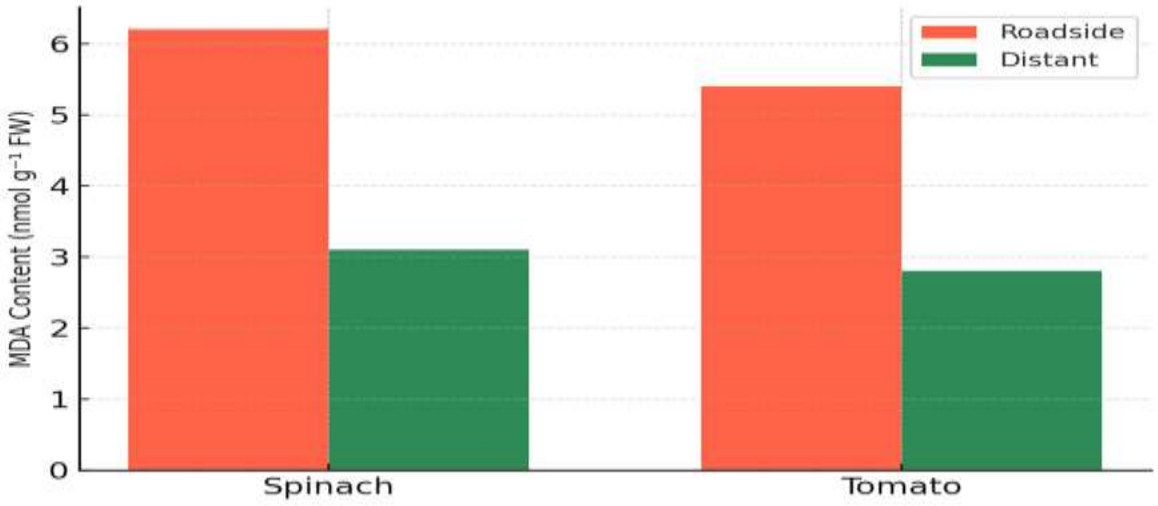


Figure 6. Lipid peroxidation (MDA content) in spinach and tomato leaves under roadside and distant soil conditions.

Values are presented as mean ± SD (n = 5). Different letters indicate significant differences according to Tukey's HSD test at $p < 0.05$.

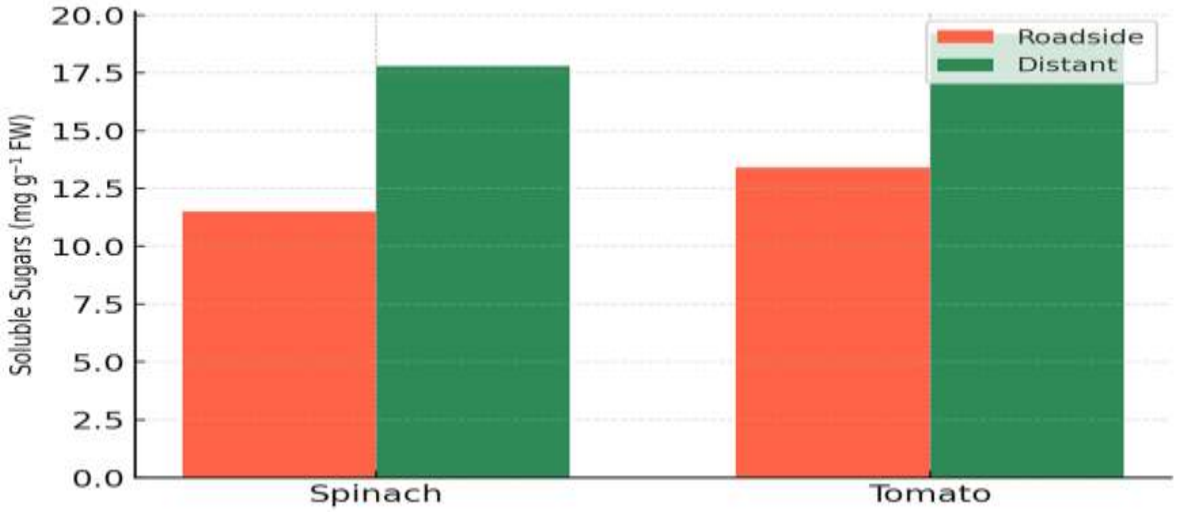


Figure 7. Soluble sugar content in leaves of spinach and tomato grown near and away from roads.

Values are presented as mean ± SD (n = 5). Different letters indicate significant differences according to Tukey's HSD test at $p < 0.05$.

Discussion

Roadside proximity as a source of Pb and Cd contamination

The significantly high lead (Pb) and cadmium (Cd) levels in urban soils and foliage support the hypothesis that vehicular exhausts and the settlement of atmospheric particulates are the major

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contributors to enrichment of heavy-metals in urban places of agriculture.

Similar findings were measured in a comparative study in Sulaimani, where Sürücü et al. [15] indicated that the loads of metals in the soils located along three main roadways were significantly higher than the loads in the soils of more remote agricultural areas, which is consistent with the current findings. The same accumulative patterns are reported in different geographic locations, which highlights the role of pollutants caused by traffic in the deposition of metals [15], [16].

The twofold average percentage increase in Pb and Cd concentrations of the plants located at roadside areas, as opposed to the distant control vegetation, supports [17] and [18] results, thus, proving that the distance to main roads is an attractive tool of the degree of anthropogenic contamination intensity.

Species-specific accumulation: fruiting tomato and leafy spinach.

The concentrations of Pb and Cd were statistically higher in spinach compared to tomato, which is expected based on the previous literature showing that the leafy vegetables accumulate and accumulate higher fractions of metals due to the high surface area of the leaves and their direct exposure to the contaminated airborne dust [19], [20].

In its turn, being a fruiting crop, tomato is likely to inhibit migration of heavy metals between roots and edible tissues, which can be explained by its relatively thick cuticle and lower rate of transpiration [21], [22].

This observed dissimilarity justifies the usefulness of spinach to be a bioindicator species of heavy-metal fire in peri-urban farming sites, a finding that is reproduced by [23] study conducted in Sulaimani.

Impacts on photosynthesis: decreases in SPAD and Fv/Fm

The measured decrease in chlorophyll content (SPAD) and maximum quantum yield of PSII (Fv/Fm) in vegetation in the roadside are evidence of direct damage to the activity of chlorophyll biosynthesis and photosystem 2.

Exposure to heavy metal interferes with chlorophyll biosynthesis by replacing either Mg^{2+} or Fe^{2+} in the rings of porphyrins and disrupting the integrity of thylakoid membranes [24], [19].

These findings are supported by the earlier studies [25] and [26] that proved that chlorophyll fluorescence parameters are the most sensitive bioindicators of environmental stress.

According to [27], the decreases of Fv/Fm were closely correlated with the photoinhibition of PSII in the case of cadmium-induced toxicity.

It is important to note that spinach rather than tomato was more severely affected implying an increased susceptibility of spinach to oxidative photodamage under metal stress.

Oxidative stress markers: elevated SOD and MDA

Activation of oxidative stress pathways is manifested by the concomitant increase of the superoxide dismutase (SOD) activity and malondialdehyde (MDA) levels in

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roadside vegetation. SOD catalyzes the dismutation process of superoxide anions which serve as the major enzymatic defense in relation to reactive oxygen species [28]. However, the fact that the levels of MDA increased proves that the antioxidant defense was not sufficient to prevent lipid peroxidation and the consequent damage of the membrane. Similar reactions have been observed in spinach and tomato that are put under the stress of lead (Pb) and cadmium (Cd) [29], [30], [31]. High levels of SOD activity before MDA formation have also been identified as one of the indications of oxidative stress in the plants subjected to heavy metals [32].

Carbohydrate metabolism: reduction in soluble sugars

The soluble sugars concentrations showed a statistically significant reduction in plant populations along road sides which indicated a diminished photosynthetic carbon assimilation and the possible reallocation of carbohydrates to antioxidant defense mechanisms.

Similar decreases in non-structural carbohydrate pools under abiotic stress conditions were recorded by the research [33] and were explained by the metabolic realignments caused by oxidative stress.

[34] also gave additional confirmation that carbon reprogramming to metal stress serves as a mechanism that can aid the preservation of cellular homeostasis, but at the expense of growth-associated physiological status.

Analytical validation and methodological reliability

The analytical procedure used in the current research, which is the acid digestion as per EPA Method 3050B and the subsequent quantification of the analysis by the use of ICP-OES, is in compliance with the international standards of analyzing soils and plant samples [35], [36].

It was done by the inclusion of certified reference material, procedural blanks, and systematic calibration verification to guarantee the accuracy and the reproducibility of the final measurements.

The following methodological decisions are consistent with the recommendations issued by [37] and [38], thus confirming the validity of the data.

Food safety and environmental implications

Taking into consideration that spinach and tomato are the most consumed vegetables in Sulaimani, the high amounts of lead (Pb) and cadmium (Cd) in the areas of road networks are genuine reasons of concern as far as food safety is concerned.

Pb and Cd concentration in spinach were found to exceed the allowed limit of the food and agriculture organization/world health organization [39] in leafy vegetables.

Similar studies in the area have reported that there are some heavy metal remnants in products that are irrigated with wastewater or planted in peri-urban soils [18], [23].

A combination of physiological indices, such as the chlorophyll fluorescence values (SPAD and Fv/Fm) and superoxide dismutase (SOD) activity and malondialdehyde (MDA) were combined with chemical measurements is a cost-effective method of environment

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monitoring at the longitudinal scale and the timely detection of the signs of stress in agricultural systems under the influence of the pollution.

Spinach and tomato as bioindicators of agro-urban pollution

The responses that have been compared show that spinach has a greater sensitivity and accumulative capacity, but tomato has partial physiological tolerance presumably due to morphological variations and antioxidative capacity.

The spinach and tomato are a bioindicator species and comparative reference tolerant species, respectively, in evaluating metal stress at pollution gradient levels based on this distinction [40], [41].

The integration of these complementary species has the prospective of supplementing biomonitoring networks in fast urbanizing areas as is the case with the Sulaimani area.

Conclusion

The current study conclusively proves that the nearness to roadways significantly increases the lead (Pb) and cadmium (Cd) pollution in the soil matrices and the plants in the Sulaimani area.

In both cases, namely, in spinacia oleracea and in Solanum lycopersicum, there were observable physiological and biochemical perturbations in the exposure to high levels of heavy metals.

Plants which had been taken at roadside locations had a decrease in chlorophyll content (SPAD), a decrease in photosynthetic quantum efficiency (Fv/Fm), an increase in antioxidant enzyme activity (superoxide dismutase, SOD), and an increase in lipid peroxidation (malondialdehyde, MDA) which were accompanied by a decrease in the levels of soluble carbohydrates.

Spinacia oleracea had greater levels of Pb and Cd and expressed stronger stress responses as compared to Solanum lycopersicum hence demonstrating greater sensitivity and suitability as a bioindicator species to measure heavy-metal pollution.

The findings support the urgency to avoid planting leafy vegetables near heavy traffic routes and consider integrating the physiological stress biomarkers with the traditional chemical methods of environmental evaluation into the environmental monitoring models.

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