Chemical and Biological Properties of Agricultural Soils Located along Communication Routes

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Abstract: The aim of the study was to assess the quality of agricultural soils, which accumulate additional amounts of heavy metals from fertilization and modify their bioavailability, with the use of interdependencies between their biological and chemical properties conditioned by the distance from communication routes. Our results indicated that heavy metals had an impact on enzyme activity in soils and their accumulation was significantly related to the distance from the edge of the road, location of sampling sites, date of soil sampling, and years of research. It was found that the greatest amounts of zinc, cadmium, lead, and copper were accumulated at a distance of 5–20 m from the edge of the road. The highest enrichment factor and geoaccumulation index values were recorded for Pb, followed by Zn, Cu, and Cd. Principal component analysis and regression models showed that the activities of phosphatase and dehydrogenases seem to be the best bioindicators of contamination of roadside soils used for agricultural purposes. Since their activity is related to soil’s organic carbon content, inputs of organic fertilizers and crop residues should be ensured in the agroecosystems along roadsides.

Keywords: agroecosystems; zinc; copper; lead; cadmium; enzymatic activity; dehydrogenases; neutral phosphatase; urease; proteases; organic carbon; pH; enrichment factor; geoaccumulation index

1. Introduction

The increasing road infrastructure in agricultural areas, although vital for the socioeconomic development of countries in the world, causes specific negative environmental effects, including the accumulation of potentially toxic elements such as heavy metals (HMs) [1–8].

Heavy metals (HMs) generally include both biologically essential (e.g., Cu, Fe, Ni, and Zn) and non-essential elements (Cd, Pb, Hg, and As) with relatively high atomic weights (63.5–200.6 g mol⁻¹) and densities greater than 4–6 g cm⁻³ (with the exception of As, B, and Se). At higher concentration levels, these elements pollute the environment and have deleterious health implications for plants, animals, and humans. Their nonbiodegradability, bioaccumulative potential along the food chain, high biotoxicity, prevalence, and persistence in the environment pose a global threat to food safety and human health [9,10].

HMs of vehicular origin are mainly emitted from incomplete fuel combustion, oil leaking from the engine and hydraulic systems (e.g., Cd, Zn, Ni, and Cr), wear of individual components such as brake lining materials and disc/drum, the car body, clutch discs, motor parts, and/or tires (e.g., Zn, Cd, and Cu). HMs are also released by road abrasion, traffic control device corrosion, and road maintenance [1,2,6,8]. It is estimated that 90% of vehicle-emitted heavy metals end up in the soil, with depositions via aerial transport or the
infiltration of runoff and spray water usually up to a distance of 100 m from the road edge, which is strongly related to traffic density [2,8].

Fertilizing products applied to agricultural soils along roadsides supply additional amounts of heavy metals, limited by EU regulation [11], and modifying their bioavailability. This should be monitored in order to prevent HMs’ bioaccumulation, biotransfer, and biomagnification in the food chains [12–15].

Since determining the content of HMs in the soil does not always fully reflect the real ecotoxic risk associated with their presence in the environment, the activities of certain soil enzymes, e.g., dehydrogenases, urease, or phosphatase, have been proposed as indicators of heavy metal contamination in soil ecosystems [16,17]. Enzymes have a close relation with nutrient cycles and exhibit quick responses and high sensitivity to temporary changes in toxic soil elements. Thus, the analysis of microbial enzyme activities together with the total content of heavy metals provides information on soil quality and ecotoxicology [1,16,18].

Most studies on soil contamination with HMs along roadsides concentrate on areas with high anthropogenic pressure and/or with a subtropical climate [2,6,19,20], while contemporary multi-season data from agroecosystems’ temperate zones, combining chemical and biological properties of the soil, are limited.

The aim of the study was to assess the quality of agricultural soils with the use of interdependencies between their biological and chemical properties, conditioned by their distance from communication routes. The three-year environmental research conducted enabled the assessment of the intensity and direction of changes in potentially toxic HM-concentration in agroecosystems located along roads, using statistical analysis and identifying the relationship between chemical and biological parameters. These were in line with the objectives of the EU Soil Strategy for 2030, reaping the benefits of healthy soils for people, food, nature, and climate.

2. Materials and Methods
2.1. Study Area and Soil Sampling

Environmental research was conducted in 2017–2019 in six agroecosystems located along roads with different average daily annual traffic in the Kielce, Lublin, Lęczna, and Opatów counties (Figure 1). The annual average daily traffic of vehicles (AADT) was presented on the basis of data from the last General Traffic Survey. The average annual temperature ranged between 8.4 °C (2017)–9.7 °C (2019). The mean annual precipitation varied from 486.7 mm (2018) to 711.8 mm (2017). All plots were situated on soils with a particle size composition of silts.

Soil samples from the cultivated fields were collected twice each year: in early spring (March) and summer (August), from the top layer of selected plots, located 5, 20, 50, and 100 m from the edge of the road.
2.2. Determination of Soil Properties

In air-dried and sieved soil samples, the following parameters were determined: total heavy metal content (Zn, Pb, Cu, and Cd) using the ICP-AES method, pH in 1 mol KCl dm$^{-3}$ (ISO 10390) [21], total organic carbon (ISO 14235) [22], and total nitrogen (ISO 13878) [23].

The activity of selected soil enzymes was determined: dehydrogenases (Dh) [24], neutral phosphatase (Ph) [25], urease (U) [26], and proteases (P) [27].

Enrichment factor (EF) and the geoaccumulation index ($I_{geo}$) were used to assess soil contamination by heavy metals:

1. $EF = (\text{Metal}/\text{RE})_{\text{soil}}/(\text{Metal}/\text{RE})_{\text{background}}$, where RE is the value of metal, adopted as Reference Element;

2. $I_{geo} = \log_2 (Cn/1.5Bn)$, where $Cn$ is the measured concentration of HM in soil, $Bn$ represents its geochemical background value, and 1.5 is the background matrix correlation factor [28–30].
2.3. Statistical Analysis

An analysis of variance (ANOVA) with Tukey’s studentized range test was performed. Regression analysis was performed for two models—linear and second-degree polynomial. The models that best describe the changes of the variables along with the distance were presented. The analysis of Pearson’s linear correlation coefficients and principal component analysis (PCA) were also performed using Statistica, ver. 13.3 (StatSoft, Inc., Tulsa, OK, USA).

3. Results and Discussion

The average total concentrations of heavy metals in roadside agricultural soils, presented in Figures 2–5, occurred in the following order: Zn (44.36 mg kg$^{-1}$) > Pb (24.19 mg kg$^{-1}$) > Cu (6.52 mg kg$^{-1}$) > Cd (0.39 mg kg$^{-1}$). Their accumulation was significantly related to the distance from the edge of the road, location of sampling sites, date of soil sampling, and years of research (Figures 2–5).

![Figure 2](image1.png)

**Figure 2.** Mean total concentrations of Zn, Cu, Cd, and Pb at the different distances (5–100 m) from the road edge. The same letter indicates not significantly different.

![Figure 3](image2.png)

**Figure 3.** Mean total concentrations of Zn, Cu, Cd, and Pb at the different locations of sampling sites (I–VI). The same letter indicates not significantly different.
3.1. The Distance from the Edge of the Road

The concentrations of Zn, Pb, Cu, and Cd decreased significantly with increasing distance from the communication routes, from 53.07, 30.44, 7.61, and 0.45 mg kg\(^{-1}\) (5 m from the edge of the road) to 36.10, 19.10, 5.68, and 0.34 mg kg\(^{-1}\) (100 m from the edge of the road), respectively (Figure 2).

The results were in agreement with other research [4,7,8]. According to Krailertratanachai et al. [2] there was a negative correlation between the content of Zn (\(r = -0.359\)), Pb (\(r = -0.323\)), Cu (\(r = -0.220\)), and Cd (\(r = -0.176\)) and the increase in the distance from the edge of the road. Models that best describe changes in the content of zinc, lead, copper, and cadmium with increasing distance in the range of 5–100 m explained 56.3% (Zn), 55.7% (Pb), 17.9% (Cd), and 13.8% (Cu) of the observed variability of these parameters in the present study (Table 1). Undoubtedly, this was related to the structure of automotive emissions [31]. Zinc is mainly emitted from brake and tire wear in vehicles, whereas high levels of Pb are likely to be primarily related to its historical use in leaded petrol and

Figure 4. Mean total concentrations of Zn, Cu, Cd, and Pb in spring (I) and summer (II). The same letter indicates not significantly different.

Figure 5. Mean total concentrations of Zn, Cu, Cd, and Pb in 2017–2019. The same letter indicates not significantly different.
persistence in soils [1]. Lead introduced into the soil accumulates mainly on its surface layer, which is the reason for its significant concentrations, due to both a single large input and long-term low-level exposure [1]. It was shown that the area influenced by runoff water from roads was within a range of 0–5 m, while the zone beyond 10 m was generally affected by wind and airflow [7,8], which depended on the surrounding topography, traffic volumes, and wind direction [1]. In the present study, a significant positive correlation was found between the average daily annual traffic, especially of passenger cars and buses, and the content of copper (r = 0.669), cadmium (r = 0.554), and zinc (r = 0.431). No such relationship was observed with regard to lead or meteorological conditions.

Table 1. Regression model for parameters versus distance.

| Parameter | Regression Equation | R² |
|-----------|---------------------|----|
| Zn        | y = 56.871 − 0.501x + 0.0029x² | R² = 0.563 * |
| Pb        | y = 32.737 − 0.359x + 0.0023x² | R² = 0.557 * |
| Cu        | y = 7.913 − 0.059x + 0.0004x² | R² = 0.138 * |
| Cd        | y = 0.462 − 0.0027x + 1.4127 × 10⁻⁵x² | R² = 0.179 * |
| Dh        | y = 0.763 + 0.081x + 0.0005x² | R² = 0.726 * |
| Ph        | y = 6.698 + 0.114x + 0.0006x² | R² = 0.655 * |
| U         | y = 14.28 − 0.143x + 0.001x² | R² = 0.456 * |
| P         | y = 6.148 + 0.079x + 0.0004x² | R² = 0.442 * |

* Significant at p < 0.0001.

Heavy metals’ accumulation in road dust and/or transfer to agroecosystems are extremely dangerous due to the possibility of their re-mobilization and occurrence in the living zone of humans, plants, and animals. It has been also shown that trace elements of vehicular origin are more mobile in the roadside zone mainly due to their interaction with road salts and activation of mechanisms related to ion exchange, formation of chloride complexes, and colloid dispersion [32].

3.2. Location of Sampling Sites

The highest contents of zinc, copper, and cadmium were found in the soils in Łuszczów (III) (52.32 mg kg⁻¹, 8.64 mg kg⁻¹, and 0.59 mg kg⁻¹, respectively), and the lowest in Albertów (I) (Cd, Cu) and Piekoszów (V) (Zn) (Figure 3). The soils in Skorzeszyce (VI) contained statistically larger amounts of lead, and in Giełczew (II), smaller amounts of lead, compared to other locations of sampling sites (Figure 3).

Apparently, in the present study, the annual average daily traffic of vehicles (AADT), measured in vehicles per day, was a dominant factor influencing the content of Cd, Zn, and Cu. The number of vehicles per day was 9441 in Łuszczów (III), 4192 in Albertów (I), and 3462 in Piekoszów (V). The Pearson correlations coefficient confirmed these relationships. According to Hwang et al. [31], road dust falling in the vicinity of communication routes with a high vehicle traffic volume is considered one of the most important carriers of Zn, Cd, and Cu. In the case of lead, the soil reaction appears to be the main determinant contributing to the reduction of its content in the second location of sampling sites (Giełczew). The lowest pH values (4.65) enable Pb to be easily transformed into mobile forms susceptible to leaching and/or uptake by cultivated plants.

It should be underlined that the concentrations of heavy metals in all of the six locations of sampling sites were below the maximum possible safe limit for soils [13,33]. The obtained data are consistent with the results of the arable land monitoring study in which only 3 out of every 216 monitored profiles in Poland were found to exceed the threshold content of Cd, and one of Cu, Zn, and Pb [34].
3.3. Seasonal and Yearly Fluctuations in Heavy Metal Contents

In the conducted studies, soils accumulated more heavy metals in spring rather than in summer (Figure 4). Statistically significant differences in their contents were the highest in the case of lead (8.9%) and the lowest in zinc (0.88%). Their values for cadmium and copper were 5% and 3%, respectively. Some authors explain the variability of the heavy metal contents in soils along roadides before and after vegetation season by their uptake by plants and/or seasonal fluctuations in road traffic volume [5].

In 2017–2019, there was an accumulation of cadmium, zinc, and copper in soils by 3.63 mg kg⁻¹ (Zn), 0.58 mg kg⁻¹ (Cu), and 0.04 mg kg⁻¹ (Cd) (Figure 5), respectively, which can be considered as one of the characteristics of the roadside area’s ecology, evidence of the local pressure exerted by the anthropogenic factor.

In the conditions of a decreasing impact of anthropopressure, which is reflected by the reduction in automotive pollutants containing lead compounds, a statistically significant decrease in the lead content (on average by 4.38 mg Pb/kg soil/2 years) was observed (Figure 5).

3.4. Heavy Metal Contents and Enzymatic Activity

Heavy metals have a significant impact on enzyme activity in soils [1, 18]. In the present study, significant negative correlations were obtained between the activity of dehydrogenases and the content of lead (r = -0.608), zinc (r = –0.587), cadmium (r = –0.348), and copper (r = –0.199) (Table 2). As in the case of Dh, neutral phosphatase correlated negatively with Pb (r = -0.720), Zn (r = –0.4812), and Cd (r = –0.402). Proteases showed relationships with these parameters that were weaker than Dh and Ph, but statistically significant, as follows: Zn (r = -0.610), Pb (r = -0.486), Cd (r = –0.469), Cu (r = –0.264) (Table 2). It was found that the regression model that best describes the changes in the activity of Dh, Pb, Ph, and U with increasing distance in the range of 5–100 m from the road edge explained 72.6% (Dh), 65.5% (Ph) 44.2% (P), and 45.6 (U), respectively (Table 1).

Table 2. Correlation coefficients between investigated soils’ properties.

| Parameter | Pb   | Cu   | Cd   | Zn   | Dh   | Pb   | U   | P   | C   | N   | pH<sub>EC</sub> | CN   |
|-----------|------|------|------|------|------|------|-----|-----|-----|-----|---------------|------|
| Pb        | 1.000| 0.144| 0.108| 0.432| -0.608| -0.720| 0.374| -0.486| -0.299| -0.521| 0.314| 0.604         |
| Cu        | 0.144| 1.000| 0.600| 0.788| -0.199| -0.091| 0.453| -0.264| 0.051| -0.089| 0.080| 0.227         |
| Cd        | 0.108| 0.600| 1.000| 0.678| -0.348| -0.402| 0.581| -0.469| -0.249| -0.302| 0.306| 0.360         |
| Zn        | 0.432| 0.788| 0.678| 1.000| -0.587| -0.481| 0.585| -0.610| -0.290| -0.491| 0.382| 0.616         |
| Dh        | -0.608| -0.199| 0.798| 0.704| -0.586| -0.302| 0.718| -0.306| 0.306| 0.373| 0.306| 0.277         |
| Pb        | -0.720| -0.091| 0.402| 0.481| -0.798| 1.000| -0.423| 0.684| 0.560| 0.724| 0.428| -0.752        |
| U         | 0.374| 0.453| 0.581| 0.585| -0.348| -0.423| 1.000| -0.396| -0.360| -0.488| 0.274| 0.515         |
| P         | -0.486| -0.264| -0.469| -0.610| 0.719| 0.684| -0.398| 1.000| 0.254| 0.416| -0.494| -0.508        |
| C         | -0.299| 0.051| -0.249| -0.280| 0.718| 0.560| -0.380| 0.254| 1.000| 0.921| -0.665| -0.699        |
| N         | -0.521| -0.089| -0.302| -0.491| 0.837| 0.724| -0.488| 0.416| 0.921| 1.000| -0.615| -0.902        |
| pH<sub>EC</sub> | 0.314| 0.080| 0.306| 0.382| -0.625| -0.428| 0.274| -0.494| -0.466| -0.665| 1.000| 0.473         |
| CN        | 0.604| 0.227| 0.360| 0.616| -0.783| -0.752| 0.515| -0.508| -0.699| -0.902| 0.473| 1.000         |

The data presented by many authors confirmed the particular sensitivity of dehydrogenases and phosphatases to pollution of soils with heavy metals [1]. Wyszkowska and Kucharski [35] showed that the activity of dehydrogenases and phosphatases can be inhibited from 10% to as high as 90%, depending on the concentration of heavy metals in the soil environment. In the case of urease, positive correlations between its activity and the content of heavy metals were noted, which were as follows: Pb (r = 0.374), Zn (r = 0.585), Cd (r = 0.581), Cu (0.453) (Table 2). Urease is resistant to external factors and an increase in its activity is observed even in extreme conditions. Since this extracellular enzyme is synthesized only in the presence of urea, the only factor that limits its activity is...
the availability of the substrate. The sources of urea in the roadside agricultural soils are primarily mineral and organic fertilizers, waste, and postharvest residues [36]. The toxic effect of heavy metals is caused by destroying the spatial structure of the active groups of the enzyme, as well as displacing cations essential for the functioning of the cells. Moreover, the growth and reproduction of microorganisms are inhibited by HMs, which reduce the synthesis and metabolism of microbial enzymes [16,37].

The harmful effect of heavy metals on soil enzymatic activity is mitigated by the high content of organic matter and neutral pH [38]. Interestingly, there was no strong influence of pH and organic carbon on the content of heavy metals ($r = 0.051–0.382$) in the present study (Table 2). A negative correlation was found between the activity of Dh ($r = -0.625$), P ($r = -0.494$), and Pb ($r = -0.428$) and pH, and a positive with the content of organic carbon, $r = 0.718$ (Dh), $r = 0.254$ (P), and $r = 0.560$ (Ph) (Table 2). The vital role of organic matter in the detoxification of soils contaminated with heavy metals is also indicated by other authors [39,40].

Principal component analysis (PCA) showed that the activities of phosphatase and proteases were most strongly correlated with the content of zinc and lead, while dehydrogenases and ureases were most strongly correlated with the content of Cu and Cd (Table 3). In the case of Ph and D, more than 65% of the variability in their activity could be explained by the regression equation in which the explanatory variable is the distance of 5–100 m (Table 1). Hence, they seem to be the best bioindicators of roadside soil contamination.

| Variable | PC1  | PC2  |
|----------|------|------|
| Dh       | 0.470| -0.540|
| Ph       | 0.840| -0.326|
| U        | -0.447| -0.589|
| P        | 0.839| -0.130|
| Zn       | -0.808| -0.502|
| Pb       | -0.809| 0.253|
| Cu       | -0.420| -0.874|
| Cd       | -0.500| -0.509|
| N        | 0.829| -0.205|
| pH       | -0.590| 0.216|
| C:N      | -0.422| 0.837|

3.5. Enrichment Factor (EF) and Geoaccumulation Index (Igeo)

The enrichment factor in metals and the geoaccumulation index (Figure 6) are used to evaluate the presence and intensity of anthropogenic deposition on the soil surface [28–30]. In the present study, roadside agricultural soils had EFs in heavy metals below 2, i.e., showed deficiency to minimal enrichment. Average enrichment factors ranked as follows: Pb (1.59) > Zn (1.47) > Cu (1.34) > Cd (1.32); indicating that Pb made the largest contribution to soil pollution 5 m from the edge of the roads. Abderrahmane et al. [28] also suggested that Pb originating from traffic activities greatly influences pollution in soils along the roads. EF decreased to a value <1, indicating a low contamination degree [41] at a distance above 20 m from communication routes.
suggested that Pb originating from traffic activities were classified in Class 0 (uncontaminated), except for Pb at the distance of 5 m, which was placed in Class 1 (uncontaminated to moderately contaminated) [29]. Thankfully, there is broad agreement in the scientific literature that lead contents in soil started to decline once its use in petrol was stopped in a region [1].

The geoaccumulation index has been commonly used as a geochemical criterion to assess the pollution degree of a single element in soils [30,41,42]. The I\textsubscript{geo}, as in the case of EF, reached the highest values for Pb and decreased in the following order in analyzed soils: Pb \geq Zn > Cu > Cd (Figure 7). According to the I\textsubscript{geo} classification's criterion, analyzed sites were classified in Class 0 (uncontaminated), except for Pb at the distance of 5 m, which was placed in Class 1 (uncontaminated to moderately contaminated) [29]. Thankfully, there is broad agreement in the scientific literature that lead contents in soil started to decline once its use in petrol was stopped in a region [1].

The greatest amounts of zinc, cadmium, lead, and copper were accumulated at a distance of 5–20 m from the edge of the road; above this range, EF decreased to a value <1, indicating a low contamination. This indicates the need to monitor and protect this area of agroecosystems against the potential negative impact of communication routes. Agricultural soils along roadsides were characterized by the largest zinc content and the lowest cadmium. Importantly, no exceedances of the permissible concentrations of Cd, Zn, Pb, and Cu were found. The highest EF and I\textsubscript{geo} values were recorded for Pb, followed by Zn.

4. Conclusions

The geoaccumulation index has been commonly used as a geochemical criterion to assess the pollution degree of a single element in soils [30,41,42]. The I\textsubscript{geo}, as in the case of EF, reached the highest values for Pb and decreased in the following order in analyzed soils: Pb \geq Zn > Cu > Cd (Figure 7). According to the I\textsubscript{geo} classification's criterion, analyzed sites were classified in Class 0 (uncontaminated), except for Pb at the distance of 5 m, which was placed in Class 1 (uncontaminated to moderately contaminated) [29]. Thankfully, there is broad agreement in the scientific literature that lead contents in soil started to decline once its use in petrol was stopped in a region [1].
Cu, and Cd, indicating that the enrichment caused by lead in analyzed agroecosystems was more severe than for the other HMs.

Principal component analysis and regression models ($R^2 > 65\%$) showed that the activities of phosphatase and dehydrogenases seem to be the best bioindicators of contamination of roadside soils used for agricultural purposes. Since their activity is related to soil organic carbon content ($r = 0.718$, $p < 0.001$ for Dh and $r = 0.560$, $p < 0.0001$ for Ph), inputs of organic fertilizers and crop residues should be ensured in the agroecosystems along roadsides.

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