THESES OF DOCTORAL (PHD) DISSERTATION

Dávid Mónok

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Physical, chemical, microbiological and ecotoxicological assessment of soils from the transitional and suburban zone of Budapest

Dávid Mónok

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Name of Doctoral School: Doctoral School of Environmental Sciences

Branch of Science: Environmental Sciences

Head of Doctoral School: Dr. Erika Csákiné Michéli
professor
Corresponding member of the Hungarian Academy of Sciences
Hungarian University of Agriculture and Life Sciences
Institute of Environmental Sciences

Supervisors: Dr. György Végvári
professor
Candidate of the Hungarian Academy of Sciences
Hungarian University of Agriculture and Life Sciences
Doctoral School of Environmental Sciences

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   Approval of School Leader   Approval of Supervisors

1. Scientific background and aims of the study

In cities, the environment-modifying, environmentally damaging effects of humans are concentrated. For this reason, the study of the state of the urban environment has become one of the main topics of environmental research in recent decades. Soils are significantly modified by urban activities, thus in many cases, they have only a limited ability to perform the functions typical of natural soils. Contamination of urban soils with potentially toxic elements (PTEs) is a major problem, as it can pose a threat to both the environment and human health.

Although the condition of local soils has been assessed in an increasing number of cities in the last 10–15 years, studies have been carried out very rarely in areas located in the outer parts of the city (e.g., in a transitional or suburban zone). However, these areas are also key, as the large areas of green space found here contribute to the regulation of the urban climate (e.g. water and temperature control, air purification) as well as providing habitat for different terrestrial communities.

During my research, I assessed the complex (physical, chemical, biological, ecotoxicological) effects of urban activities on the soils of grasslands located in the transition and suburban zones of Budapest. This included the study of the general physical and chemical properties of the soils, the assessment of PTE contamination, and the use of various bioassays to study these soils.

The research can be considered novel not only because soils that have rarely been the focus of other research were examined, but also because of the methods used. A bioassay system representing several trophic levels used to assess the biological and ecotoxicological status of soils has never been applied in urban soils.

The main objectives of the doctoral dissertation were:

- Assessing the impact of urban activities on the general physical and chemical properties of the soils of the studied sampling sites.

- Investigation of the relationships between the general physical and chemical parameters of soils to determine which soil properties play a key role in the modification of these soils.

- Assessment of PTE contamination of soils for the following six elements: Co, Cr, Cu, Ni, Pb, Zn. Comparison of PTE concentrations in soils with limit values and concentrations measured in other major European cities.

- Quantification and evaluation of soil biological status and potential ecotoxicity by various laboratory bioassays. Assessing the adverse biological effects of urban activities at study sites.

- Investigation of the relationship between soil PTE concentrations and the results of bioassays to determine whether soil PTE contamination can be associated with adverse biological effects.

- Determining how and to what extent the general physical and chemical characteristics of soils affect the results of bioassays.

- Finally, identification of sampling sites where the environmental risk may be significant: the soil is critically contaminated with PTEs and/or has significant ecotoxicity.

At the beginning of the research, the following hypotheses were formulated:

1. Urban activities modify the general physical and chemical characteristics of soils in the study area.

2. Urban activities contribute to the increase of concentrations of potentially toxic elements in the soils (Co, Cr, Cu, Ni, Pb, Zn), thus to soil contamination in the study area.

3. The potentially toxic element contamination of the studied soils is similar to the values measured in other European cities.

4. Urban activities have an impact on the microbiological status of soils in the study area.

5. Soils modified by urban activities are harmful (toxic) to various terrestrial organisms.

2. Materials and methods

During the research, soil samples were collected from three different types of areas, all of which were covered with grass vegetation: urban sites (n=6, marked as V_1, V_2…V_6), near-natural urban sites (n=6, marked as T_1, T_2…T_6) and non-urban sites (n=4, marked as K_1, K_2…K_4). The last group was used as control sites. All sampling sites were located in the transitional or suburban zone of Budapest, or east of the city (Figure 1).

A site was chosen as an urban site if the following criteria are present:

1. located near built-up areas, and
2. potentially affected by one or more of the following urban activities:
    - former use of the site;
    - heavy traffic or industrial activity within 100 m;
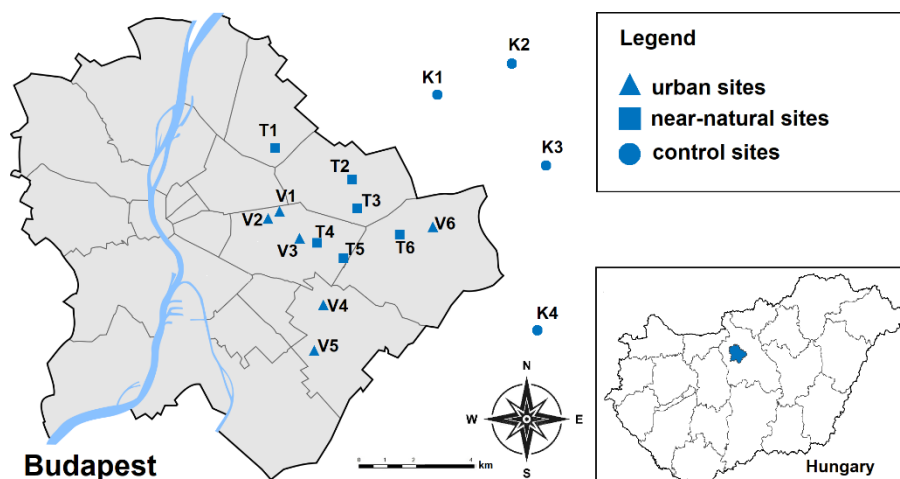   - a residential area located right next to the site.

Figure 1: Location of the sampling sites

In contrast to urban sites, near-natural urban sites were located near natural areas and had no heavy traffic or industrial activity in their immediate vicinity (within about 200-250 meters). However, local residents may occasionally visit these sites for various recreational purposes (e.g., for dog walking, sports, or recreation). For this reason, soils are not completely free from human disturbance here either, although the extent of disturbance is undoubtedly much lower than in urban areas.

Non-urban sites (control sites) are located outside the administrative border of Budapest, more than 19 km east of the city center. In their surrounding area, there are mainly natural meadows, forests and, to a lesser extent, agricultural land. Since these sampling sites are geographically close to other sites but relatively far from different urban activities, they are suitable for control in my study.

Soil samples were collected during September–October 2018. At each site, four composite topsoil samples (0–20 cm) were taken, consisting of 10 point samples. Sampling points were randomly selected at the sites. After collection, the composite samples were homogenized.

After the appropriate sample preparation, the general physical and chemical properties of the soils were determined based on Hungarian standards. The

following parameters were examined: artefacts content, plasticity limit according to Arany (K_A), pH (pH_{H2O}), calcium-carbonate content ($CaCO_3\%$), and humus content (H%). In addition, the nutrient supply of the soils was also examined for the three most important nutrients: total nitrogen content (N%) and ammonium-lactate soluble phosphorus and potassium content (AL-P, AL-K) were also measured.

The PTE concentrations of the soil samples were determined according to the Hungarian standard (MSZ 21470-50:2006). The total PTE contents were determined after $HNO_3 + H_2O_2$ digestion, while the soluble ("bioavailable") PTE contents were determined after Lakanen-Erviö digestion. The PTE concentrations of the extracts were measured with an atomic absorption spectrophotometer. All measured element concentrations were compared with the "B" contamination limits and the "A" natural background concentrations. In addition, it was also examined how the total concentrations of PTEs in the soils of Budapest relates to the values measured in other European cities with a similar population and/or area. To characterize the PTE contamination of the soils, the integrated pollution load index (PLI) was used, based on which I classified the sampling sites into contamination categories.

For the biological and ecotoxicological examination of the soils, two types of bioassay methods were used: microbiological tests and contact bioassays (using a test organism). During the microbiological tests, the dehydrogenase enzyme activity (DHA) of the soils was measured, and the number of aerobic mesophilic bacteria and the number of fungi were determined by the most probable living cell count (MPN) method. In addition, I also performed a microscopic bacterial count. With the contact bioassays, the potential ecotoxicity of the samples was assessed. These types of tests were conducted at several trophic levels. Among the bacteria, *Azomonas agilis* and *Pseudomonas fluorescens* test organisms were used, during which the inhibition of the dehydrogenase enzyme activity of the bacteria was measured. Among the plants, a seedling test was performed using

white mustard (*Sinapis alba*) and lettuce (*Lactuca sativa*) as test plants, while ryegrass (*Lolium perenne*) was used for a longer-term bioassay. Among the soil animals, bioassays were performed using the *Folsomia candida* collembolan species and the *Eisenia fetida* earthworm. In these tests, the mortality and reproduction of the animals were the endpoints of the study, and in the case of earthworms, the weight change was also measured. Based on the results of the bioassays using the test organism, I categorized the soil samples based on their toxicity.

The results obtained during the different examinations were compared by type of site (urban, near-natural and control sites) as well as by sampling site, for which the necessary statistical analyzes were performed with SPSS Statistics 26 software. *Tukey*, *Dunnett*, or *Games-Howell* tests were used for comparison (p <0.05). *Pearson*'s correlation analysis was also used to explore the relationships between the different data (p <0.01).

3. Results and discussion

3.1. General physical and chemical properties of soils

In urban areas, the soil contained significantly more artefacts than in the other two types of sites. Here, the artefacts content ranged from 11.2 % to 23.4 %. This clearly indicates that urban activities have a strong impact on the soil at all urban sampling sites. In contrast, values were between 2.2 % and 6.8 % at near-natural sites and only between 1.4 % to 3.4 % at control sites.

K_A was the lowest at control sites, while it was significantly higher at urban sites. Based on the results, the texture of most of the soil samples was sand, sandy loam, or loam. The exact value of K_A varied between 28.5 and 37.8 at control sites, between 28.5 and 43.3 at near-natural sites, and between 31.0 and 43.0 at urban sites.

The pH of the soils differed significantly by site type. The lowest pH was found at control sites (pH_{H2O} = 6.7–7.1), while the highest was found at urban sites (pH_{H2O} = 7.3–7.9). Thus, neutral, slightly alkaline pH was measured in soils more disturbed by man, while slightly acidic soils were also found in areas outside the city.

The $CaCO_3$ content of soils was significantly higher at urban sites than in the other two site types, exceeding 10 % in several sites. In contrast, at near-natural sites weakly and moderately calcareous soils ($CaCO_3$ = 0.5–8.7 %) were found, while in the control areas only weakly calcareous soils ($CaCO_3$ = 0.8–3.3 %) were found.

The humus content of the soils was significantly lower at urban sites, with very low values of only around 1 % were measured at some sampling sites. At near-natural sites, the humus content varied between 3.26 % and 6.38 %, while at control sites it ranged from 2.24 % to 4.64 %. The same was found in the case of macronutrients: the content of soils N, AL-P, AL-K was higher at control sites and significantly lower at urban sites.

Based on correlation analyzes, it can be emphasized that the artefacts content in the soils was positively correlated with soil pH (r = 0.51) and $CaCO_3$ content (r = 0.72). The more alkaline pH and higher $CaCO_3$ content of urban soils are thus partly explained by the higher amount of artefacts. This may be due to the large amount of construction debris found in urban soils, which usually has a very high lime content and is thus often very alkaline. The artefacts content was negatively correlated with the humus content of the soil (r = -0.59). This may be since most artefacts contain little organic matter and thus have an adverse effect on the humus formation process.

3.2. PTE concentrations and contamination of soils

Mean (total) element concentrations (excluding Co and Zn) measured at urban sites were significantly higher than those obtained at control sites. Among the

examined elements, Cr should be highlighted, the total concentration of which in the soil at urban sites was on average more than three times the concentration at control sites. Cu, Ni and Pb are typical urban pollutants, which was also supported by the results of the research. Cu concentrations were remarkably high at the V_5 sampling site (97.58 mg kg^{-1}), this ranged from 30.16 to 65.55 mg kg^{-1} at other urban sites. A similar finding was observed for Pb, the concentration of which at the V_5 sampling site (533.67 mg kg^{-1}) was more than one and a half times the second-highest measured concentration (326.27 mg kg^{-1}). Ni concentrations ranged from 18.58 to 37.48 mg kg^{-1} at urban sites, which is also higher than at control sites.

Based on my results, urban activities also affect the total PTE content of soils at near-natural sites, as the concentrations of Cr and Ni in the soils were significantly higher here than at control sites. Moreover, the highest Cr concentrations were measured at near-natural sites: the soil concentration of Cr was 248.94 mg kg^{-1} at the T_3 sampling site and 220.88 mg kg^{-1} at the T_1 site. These results mean that the deposition from the air may play a greater role in the accumulation of Cr and Ni than point and linear emission sources (e.g. industrial facilities, high-traffic roads).

Comparing the total element concentrations with the contamination limit, it can be concluded that all soil samples are contaminated with Pb, as its concentration exceeded the contamination limit (100 mg kg^{-1}) at all sampling sites (several times at urban sites). In addition, Cr concentrations at urban and near-natural sites (excluding T_6 and V_6 sites) also exceeded the contamination limit (75 mg kg^{-1}). Of the soil samples, only the samples taken at the V_5 site can be considered as contaminated with Cu, while the concentrations of Co, Ni and Zn did not reach the contamination limit at any of the sampling sites. Considering the natural background concentrations, it was found that in the urban and near-natural areas, in addition to Cr and Pb, the concentrations of Cu and Ni also exceeded it at most of the sites.

Based on the PLI values, most of the sampling sites were not classified as contaminated (PLI <1.00). However, at urban and near-natural sites, higher PLI values were found at all sampling sites than at control sites, which also confirms that urban activities contribute to soil contamination with PTEs. Three sampling sites were slightly contaminated based on the assessment, but two of them (sites V_1 and V_2) were just above the lower limit of this category (PLI = 1.02). Samples collected from the V_5 site were the most contaminated with the PTEs with a PLI value of 1.32.

The mean concentrations of PTEs in control soils were lower than in the soils of the 11 European cities used for comparison (Athens, Belgrade, Berlin, Vienna, Glasgow, Copenhagen, Krakow, Lisbon, Naples, Stockholm, Turin). However, the concentrations of Cr and Pb in the soils of urban and near-natural sites were higher than the average of European cities (62.4 and 201.8 mg kg^{-1}). In contrast, the measured concentrations of Co, Cu, Ni and Zn in the soil at all site types were lower than the average of European cities (12.8; 66.3; 57.4; and 188.5 mg kg^{-1}, respectively). The Zn content of soils in Budapest was particularly low, proving to be the lowest of all cities. Based on the PLI value, the mean value of PTE contamination of the soils from urban sites (PLI = 0.97) is lower than the average of the other 11 European cities. At control sites, the mean PLI value (PLI = 0.48) is lower than in any other city.

Soluble (more bioavailable) element concentrations in soils were also measured to understand the results of the bioassays. These concentrations were typically 5–15-fold lower than total elemental concentrations. Based on the statistical analysis, the mean soluble concentration of PTEs measured at urban sites (excluding Co) were significantly higher than those measured at control sites, suggesting that urban activities affect not only the total but also the soluble PTE content of soils.

3.3. Results of bioassays

According to microbiological tests, DHA was nearly 50% lower in urban soils (0.77 TPFµg g^{-1}) than at control sites (1.60 TPFµg g^{-1}). The lowest DHA was measured in the soils of the sampling sites V_1 and V_2 (0.23 and 0.22 lgMPN g^{-1}), which values are about 85 % lower than the average of the control sites. No differences were found in the total number of bacteria between the site types, the results ranged from 1.97 to 2.41 lgMPN g^{-1}. In contrast, the microscopic bacterial count showed significantly lower total cell numbers in urban and even near-natural soil samples than in soils from control sites. 8.97 and 9.54 lg pcs cm^{-3} were calculated at control sites, 8.80 and 9.14 lg pcs cm^{-3} at near-natural sites, and 8.78 and 8.95 lg pcs cm^{-3} at urban sites. There was also a significant difference in the number of fungi between the soils of the control and urban areas, which means that the number of fungi in the soil also decreased as a result of urban activities. Values ranged from 0.33 to 0.38 lgMPN g^{-1} at control sites and only from 0.28 to 0.37 lgMPN g^{-1} at urban sites.

Among the bacterial bioassays, according to the *A. agilis* test ED_{50} values were between 1.03 and 1.43 at control sites and between 0.78 and 1.37 at near-natural sites. In the latter, the soils at sampling sites T_1, T_2 and T_3 were found to be mildly toxic. Soil samples from urban sites were even more toxic to the test bacterium. ED_{50} values ranging from 0.34 to 0.59 at urban sites and all urban soil samples (except soil from the V_6 site) were found to be moderately toxic. The most toxic soils were from sampling sites V_1 ($ED_{50} = 0.34$) and V_2 ($ED_{50} = 0.34$). According to the *P. fluorescens* test, soils from urban and near-natural sites were more toxic to the test bacterium than those from control sites. ED_{50} values ranged from 0.60 to 0.63 at control sites, 0.20 to 0.62 at near-natural sites, and 0.13 to 0.41 at urban sites. Among the near-natural soils, the soil at the T_1 sampling site was found to be highly toxic, while samples from the T_2, T_4 and T_5 sites were found to be moderately toxic. Among the urban soils, the soil from the V_5 site proved to be

the most toxic ($ED_{50} = 0.13$), but the soils from the V_1, V_2 and V_4 sites were also highly toxic, while the other sites showed moderate toxicity.

According to the seedling test, none of the soil samples collected from the control sites were toxic to the test plants. In contrast, all samples from near-natural and urban sites (excluding V_3 and V_6 sampling sites) significantly reduced the values of at least 1 examined plant parameter (germination, root length or shoot length). Of the three parameters examined, plant germination was the most sensitive. The most toxic soils were from sites V_1 and V_2, these samples reduced plant germination by about 70 % compared to the control. In addition, samples from sites T_1, T_5 and V_5 were also highly toxic, with a reduction of more than 50 %. Samples from sites T_2, T_4, T_6 and V_6 were characterized by moderate toxicity for plant germination. Root and shoot growth were less damaged by the samples, however, the highest toxicity was observed at the same sites for these parameters as well. In the ryegrass bioassay, much less toxicity was observed than in the seedling test. Samples from control sites were not toxic in this plant test either, however, samples from some near-natural sites (T_2 and T_6 sites) and urban sites (sites V_3, V_4, and V_6) were also non-toxic. Nevertheless, a significant difference in shoot length and shoot dry weight was found between control and the other two site types. In particular, a significant decrease was observed in the latter parameter. The largest decrease was caused by soil samples from sites T_5, V_1, and V_2, where approximately 35 % less shoot mass was observed compared to control. Samples collected from site V_5 were also moderately toxic to this parameter, and soil from site T_1 was found to be mildly toxic.

According to the *F. candida* test, none of the soil samples from control sites was toxic to the collembolan species tested. In contrast, samples from urban and near-natural sites (except for the T_4 site) all reduced the number of adult as well as juvenile individuals. The sample from the T_1 site was found to be the most toxic of all samples in terms of both adults and juveniles. It caused a decrease of more than 70 % in the number of adults and more than 80 % in the number of juveniles.

Samples from T_2, T_3, and T_6 sites were also highly toxic to the reproduction of the animal, but only moderate toxicity was observed regarding the number of adults. Each of the samples from urban sites was moderately or highly toxic to *F. candida*. The sample from site V_2 was highly toxic to both parameters examined, but samples from sites V_1, V_5, and V_6 were also found to be highly toxic to reproduction. The latter is particularly interesting, as e.g. in plant tests, this sample did not cause any adverse effects at all. The same was true for the sample from the V_3 site, which was moderately toxic in this test. In the earthworm test, soil samples from control sites were also non-toxic, and samples from near-nature sites were not decreased the number of adult individuals. However, the number of juveniles was lower at both urban and near-natural sites (excluding the T_3 site) than at control sites. The number of adults was reduced the most by samples from sites V_4, V_2, and V_5, the former by nearly 50 % and the latter two by nearly 40 %. This means that all three samples were found to be moderately toxic to this parameter, while samples collected from sites V_1 and V_3 were slightly toxic. Among the near-natural sites, soil samples collected from the T_4 site were highly toxic to reproduction, with a reduction in juvenile numbers of nearly 70 %. Samples from sites T_1, T_2, and T_6 were found to be moderately toxic, while samples from site T5 were found to be mildly toxic to this parameter. Samples from urban sites all showed significant toxicity to this parameter, with even the least toxic soil from the V_3 site reducing the number of juveniles by nearly 50 %. The other urban soils were all highly toxic to reproduction, the most toxic soil samples (from sites V_2 and V_5) reduced the number of juveniles by more than 70 %.

3.4. Investigation of factors influencing the results of bioassays

According to the correlation analysis, some soluble element concentrations in the soils influence the studied microbiological parameters. The concentration of Pb was negatively correlated with the results of DHA (r= -0.40) and microscopic cell number (r= -0.44), while DHA was also negatively correlated with the

concentration of soluble Cr in the soil (r= -0.45). These results are not surprising, as the total concentrations of these two PTEs exceeded the contamination limits at most sites, and neither Cr nor Pb is an essential element for living organisms. Although the Zn content of the soil samples was relatively low, a negative correlation was found between the microscopic cell number and the soluble Zn concentrations (r= −0.41). Among the general physical and chemical parameters, the artefacts content was negatively correlated with all microbiological parameters examined, but the correlation coefficients (similar to PTEs) were relatively low (r< -0.52). Humus content was positively correlated with all microbiological parameters (0.44< r <0.56), and microbiological parameters were also positively correlated with the amount of at least one nutrient except DHA (0.41< r <0.87).

According to the correlation analysis, the soluble concentrations of Cr, Cu and Pb in the soils were negatively correlated with the ED_{50} values obtained in bacterial bioassays (-0.51< r <-0.63), which means that higher concentrations of soluble PTE increased the toxicity for the studied test bacteria. This explains the higher toxicity observed at urban sites, as the soluble concentrations of all three PTEs were significantly higher in the soils of these sites. Among the general physical and chemical characteristics, artefacts and humus content should be highlighted (similar to microbiological tests). The artefacts content was negatively (-0.56< r <-0.68), while the humus content was positively correlated with ED_{50} values (0.57< r <0.58).

In the case of plant tests, it was found that the concentration of soluble Pb in the soil samples was negatively correlated (-0.49< r <-0.67) with all examined parameters (except for the root length of white mustard), while the concentration of soluble Cr negatively affected the germination of white mustard (r= -0.49). No significant correlation was found between the soluble concentrations of the other PTEs and the plant parameters. The results of plant bioassays are significantly influenced by the general properties of the soil, thus the correlation analysis is

even more justified in this case. However, in this case, only soil N concentration influenced the results, as it was positively correlated with all the parameters examined in the seedling test and also with the shoot weight of the ryegrass ($0.42 < r < 0.61$). These results are not surprising, as it is well known that the role of N in the (initial) vegetative phase of plant growth is very important. Thus, based on the results, the high N content of soils can reduce the harmful effects of PTEs on plants.

According to the correlation analysis, there was a negative correlation between the number of adult individuals of *F. candida* and the soluble concentrations of PTEs (excluding Zn) ($-0.42 < r < -0.63$). However, only the soluble concentrations of Co and Ni correlated significantly with the number of juveniles ($r = -0.60$ and $r = -0.64$, respectively). In the earthworm test, significant correlations were found in the case of Cr, Cu and Pb, their soluble concentrations were negatively correlated with all the measured parameters ($-0.42 < r < -0.71$). Based on these, it is clear that higher PTE concentrations in soil are responsible for the higher toxicity of urban soils to soil animals. At the same time, among the general physical and chemical parameters of the soil, artefacts content, pH and $CaCO_3$ content were also negatively correlated with some of the examined parameters in soil animal tests ($-0.41 < r < -0.56$). In contrast, the N concentration of the soil samples was positively correlated with the number of adults and juveniles of *F. candida* and the number of juveniles of *E. fetida* ($0.43 < r < 0.48$). The humus content was positively correlated with all measured parameters of earthworm ($0.43 < r < 0.54$).

4. Conclusions and recommendations

Based on the results, urban activities significantly modify the physical, chemical, biological and ecotoxicological characteristics of the soils at the studied sampling sites in Budapest.

The high content of artefacts (above 10%) in urban sites is a clear indication of human disturbance, as these substances can only enter the soil through human activities. The amount of artefacts can play a key role in soil modification as it changes the composition and structure of the soil, so it can also affect many natural processes in the soil. Based on my research, higher artefacts content contributes to more alkaline pH, higher $CaCO_3$ content, and lower humus content in urban areas.

Based on the results, it was clearly shown that the total and soluble concentrations of PTEs in soils within the city were also typically higher than those in soils outside the city. The total concentrations of Cr and Pb were particularly high, as in many cases they exceeded the contamination limit by several times. In addition, the measured concentrations are considered to be outstanding in Europe. As PTEs can enter the lower soil layers in some cases, thus posing a significant environmental risk, I recommend the assessment of elemental concentrations in the lower soil layers as well, especially at sites where significant soil contamination has been observed.

Changes in the physical and chemical characteristics of urban soils, but in particular their contamination with PTEs may be associated with a deterioration of natural soil functions and damage to soil organisms, which has been clearly demonstrated in the research.

The microbiological condition of the soil was more unfavorable in urban sites than outside the city. This can be related to human disturbance, as the artefact content of the soils is negatively correlated with the measured microbiological values. In addition, soluble concentrations of some PTEs, especially those

presented in relatively large amounts (Cr and Pb), were also negatively correlated with microbiological parameters. Furthermore, the reduced amount of humus and nutrients can also have an effect on this, which was also confirmed by the results. However, since microorganisms play a very important role in the circulation of nutrients and the decomposition of organic matter, the correlation may be reversed: lower microbiological activity may contribute to the reduction of humus and soluble nutrients.

It was found that all soil samples from urban and near-natural sites were toxic to at least two terrestrial test organisms at different trophic levels under laboratory conditions. As the applied bioassay methods model the natural conditions relatively well, it can be assumed that these adverse effects also occur in the real environment. In contrast, no toxicity was observed for non-urban samples (except for the *P. fluorescens* test).

Significant differences were found in sensitivity between the test organisms used. The two different types of plant tests, but especially the ryegrass bioassay, were less sensitive, which may be since plants generally tolerate lower concentrations of PTE contamination. This is also indicated by the fact that the soluble PTE concentrations of the soils had a much lesser effect on the test results according to the correlation analysis than e.g. for terrestrial animals. Based on seedling tests, the studied soils may have a negative impact on sensitive plant species, which may promote the spread of more tolerant plant species in urban areas. Soil animal tests, but especially animal reproduction, were highly sensitive to the samples tested, and adverse effects were clearly associated with soluble PTE concentrations in soils. If the reproduction of a species is inhibited, it is clearly accompanied by a decrease in the number of individuals. Of course, this has other disadvantages in urban soils: since the studied species play a very important role in shaping the structure of the soil, the physical condition of the soil may deteriorate. In addition to the reproduction of soil animals, the bacterium *P. fluorescens* has also been shown to be very sensitive in the study, even though

this species is well adapted to extreme environmental conditions, e.g. for higher PTE concentrations.

Although in many cases we found a correlation between the soluble concentration of a given element and the adverse effects, it cannot be ruled out that other contaminants (which could not be tested in the present research) may also have contributed to the toxicity of the soil samples. This confirms the need to use bioassays in soil testing, as they can indicate environmental risks even if we do not have information on the pollutant responsible. That is why I recommend the use of bioassays and ecotoxicological tests in the examination and environmental risk assessment of urban soils. In my experience, there are very few examples of this even in the international literature.

A further aim of the research was to identify risk areas from the sampling sites. Based on the contamination with PTEs, site V_5 can be highlighted, as we obtained the highest PLI value here, however, sites V_1 and V_2 were also classified as slightly contaminated. In these areas, a more detailed assessment of contamination is recommended in the future (e.g. examination of lower soil layers, identification of pollution sources), especially because the samples from here were typically the most toxic to the test species in bioassays. It would also be worthwhile to perform a broader analytical study on the most common urban pollutants (eg other PTEs, PCBs, etc.). I suggest the same in sites where the soil was not classified as contaminated based on PLI, yet significant toxicity was observed for some species (sites T_5, V_4 and V_6). These places can be considered environmentally risky.

Overall, based on my results, in the future, more attention should be paid to the condition of soils in suburban (transitional and suburban zones) areas, especially their complex assessment, which considers biological and ecotoxicological parameters in addition to physical and chemical data. My doctoral dissertation can also provide practical help for this.

5. New scientific results

During my research, the complex (physical, chemical, biological, ecotoxicological) effects of urban activities were studied on the soils of grasslands located in the transition and suburban zones of Budapest. The new scientific results obtained and the answers to the formulated hypotheses are the following:

1. **Urban activities modify the general physical and chemical characteristics of soils in the study area. The hypothesis is accepted.**

   Based on my research, the artefacts content in soils of urban sites is many times higher than in the soils outside the city, which contributes to the more alkaline pH of the soil, its higher calcium-carbonate content, and its lower humus content. In addition, the total nitrogen and ammonium-lactate soluble phosphorus and potassium contents of the soil are also lower in the soils of urban sites.

2. **Urban activities contribute to the increase of concentrations of potentially toxic elements in the soils (Co, Cr, Cu, Ni, Pb, Zn), thus to soil contamination in the study area. The hypothesis is partially accepted.**

   Based on my research, urban activities contribute to the increase of the concentrations of some potentially toxic elements (Cr, Cu, Ni and Pb) in the soil in the studied area, however, e.g. in the concentration of Co, there were no differences between the soils of urban and non-urban sites. Based on the integrated pollution index (PLI), some of the soils of the examined sites in Budapest (sites V_1, V_2, and V_5) were slightly contaminated, while the soils of the other sites were not classified as contaminated.

3. **The potentially toxic element contamination of the studied soils is similar to the values measured in other European cities. The hypothesis is rejected.**

The contamination of the studied soils (in Budapest) with potentially toxic elements differs from the values measured in other European cities with a similar area and/or population. I found that the concentrations of Cr and Pb were typically higher, while the concentrations of Cu and Zn were lower than in the 11 European cities used for comparison (Athens, Belgrade, Berlin, Vienna, Glasgow, Copenhagen, Krakow, Lisbon, Naples, Stockholm, Turin).

4. **Urban activities have an impact on the microbiological status of soils in the study area. The hypothesis is accepted.**

Dehydrogenase enzyme activity was 50% lower in urban soils than in control sites, and the number of fungi, as well as the number of microscopic cells, were also lower. In addition, the latter parameter was also lower in urban near-natural areas than in control sites. Based on these, the microbiological condition of the soils in the studied area deteriorates as a result of urban activities.

5. **Soils modified by urban activities are harmful (toxic) to various terrestrial organisms. The hypothesis is accepted.**

I found that all of the tested soils were toxic to at least two terrestrial test species under laboratory conditions among the following species: *Azomonas agilis, Pseudomonas fluorescens, Sinapis alba, Lactuca sativa, Lolium perenne, Folsomia candida, Eisenia fetida*. The extent of the adverse effects was related to the concentrations of soluble potentially toxic elements in the soils, especially those present in the samples in excess of the contamination limit (Cr and

Pb). The general physical and chemical parameters of the soils influence the results of the bioassays: The high artefacts content increased, while the high humus and N content reduced the toxicity.

6. The author's publication in the topic of the dissertation

Scientific article in international journal with IF:

Mónok Dávid, Kardos Levente, Pabar Sándor Attila, Kotroczó Zsolt, Tóth Eszter, Végvári György (2020): Comparison of soil properties in urban and non-urban grasslands in Budapest area. Soil Use and Management. In press. https://doi.org/10.1111/sum.12632

Scientific article in foreign language:

Mónok Dávid, Kardos Levente, Pabar Sándor Attila, Kotroczó Zsolt (2020): Physico-Chemical and Ecotoxicological Characterizations of Suburban Soils. International Journal of Environmental Pollution and Remediation 8, pp. 23-29. https://doi.org/10.11159/ijepr.2020.003

Scientific article in hungarian language:

Mónok Dávid, Füleky György (2017): A talaj kadmium szennyezettségének vizsgálata angolperje (*Lolium perenne* L.) bioteszttel. Agrokémia és Talajtan, 66 (2), pp. 333-347. https://doi.org/10.1556/0088.2017.66.2.3

Mónok Dávid, Kardos Levente, Végvári György (2019): Bársonyvirágfajok (*Tagetes* spp.) nehézfém fitoremediációs potenciáljának értékelése laboratóriumi tesztmódszerekkel. Agrokémia és Talajtan 68 (1), pp. 139-154. https://doi.org/10.1556/0088.2019.00036

Mónok Dávid, Strbik Dorina (2020): Az ólom hatása az angolperje (*Lolium perenne*) növekedési paramétereire komposzttal kezelt homoktalajon. Talajvédelem Különszám, pp. 245-253. http://real.mtak.hu/115042/1/Talajvedelem_Kulonszam_2020_Fekete_etal.pdf

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Mónok Dávid, Kardos Levente (2019): Examining soils of urban areas using plant biotests. In: Szigyártó, I-L; Szikszai, A (szerk.) XV. Kárpát-medencei Környezettudományi Konferencia (ISBN 1842-9815). Kolozsvár, Románia. Ábel Kiadó, p. 210.

Mónok Dávid, Kotroczó Zsolt, Pabar Sándor Attila, Tóth Eszter, Kardos Levente, Végvári György (2019): Városi talajok vizsgálata fizikai, kémiai és biológiai módszerekkel. In: Németh, Katalin (szerk.) Absztraktkötet. Tavaszi Szél Konferencia (ISBN 978-615-5586-42-2). Nemzetközi Multidiszciplináris Konferencia. Budapest, Magyarország. Doktoranduszok Országos Szövetsége, p. 314.

Mónok Dávid, Kardos Levente, Pabar Sándor Attila, Kotroczó Zsolt (2020): Applying Bioassays for Investigation of Soils from Suburban GreenSites. In: CSEE, Congress (szerk.), Proceedings of the 5th World Congress on Civil, Structural, and Environmental Engineering, CSEE'20. International ASET Inc. Paper: ICEPTP 108.

Mónok Dávid, Kardos Levente, Végvári György (2020): Investigation of physical and chemical properties of soils from different urban grasslands. In: Csiszár B, Hankó Cs, Kajos LF, Kovács OB, Mező E, Szabó R, Szabó-Guth K (szerk.), 9th Interdisciplinary Doctoral Conference Book of Abstracts. Pécsi Tudományegyetem Doktorandusz Önkormányzat, Pécs, p. 384.