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EFFECT OF SOIL POLLUTION WITH PAHs ON THE AMOUNT OF MAIZE BIOMASS AND ACCUMULATION OF CADMIUM AND LEAD

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Abstract: Trace element bioavailability is of key importance for stimulation or inhibition of plant growth and development processes at later stages of their life and in result leads to changes of biomass biological value, *eg* limiting its use. As assessment of artificial soil pollution with benzo(a)pyrene, chrysene and fluorene effect on the quantities of biomass and accumulation in it of cadmium and lead was conducted as a pot experiment. The soil pollution with the studied aromatic hydrocarbons did not inhibit growth or development of maize shoots and roots. The greatest amount of biomass was obtained in the object where the soil revealed an elevated contents of the analyzed aromatic hydrocarbons. Value of tolerance index in the objects where the stress agent was introduced was above one, which indicates the absence of soil pollution with benzo(a)pyrene, chrysene and fluorene effect on plant biomass quantity. Value of tolerance index below one concerned only biomass from the control object. Significant increase in Cd content and its quantities taken up by maize shoots was registered on the treatments where dichloromethane and polycyclic aromatic hydrocarbons were added to the soil in comparison with the unpolluted objects. The content and amounts of absorbed lead were the lowest on the object where the soil was the most polluted. Values of maize shoot biomass Cd pollution index were apparently higher in the objects where the soil was contaminated with polycyclic aromatic hydrocarbons in comparison with the values obtained in the object where soil received only mineral nutrient solution. Values of translocation index do not point to cadmium accumulation in plant shoots. Both the values of pollution index and translocation index for lead in the objects with elevated PAHs content in soil and in the object where the soil was contaminated with these substances were below one, which did not confirm excessive lead accumulation in maize shoots.

Keywords: soil, pollution, polycyclic aromatic hydrocarbons, cadmium, lead

Introduction

Polycyclic aromatic hydrocarbons and heavy metals are the factors which most frequently cause chemical pollution of soil [1–4]. The main sources of chemical soil

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pollution are industry, agriculture and transport [5]. Depending on the pollutant emission, the rate of soil degradation may be different, conditioned also by its kind, which is connected with buffering properties and functioning of natural self-cleaning mechanisms in soil [6].

Soil pollution affects changes of not only its chemical properties but also alters quantitative and qualitative composition of soil microflora. Quantitative and qualitative changes of soil microflora directly influence a majority of biochemical processes occurring in the soil environment, which beside the processes of pollutant substances degradation, concerns also processes accompanying transformations of macroelements and trace elements [7]. In case of oil derivative pollutants a problem of unfavourable physical soil properties appears, which results in worsening of water-air relationships.

The outcome of these changes is worsening of plant living conditions, which affects the obtained crop yield and its quality. The quality of crops is of key importance regarding its destination. Anti-nutritional substances load in biomass results in its limited use. Toxic trace elements, such as cadmium or lead contained in the polluted soil may easily accumulate in plants. The content of these elements in plants requires monitoring because their potentially dangerous levels (without the symptoms of harmful effect on plants) may be reached in diets of both animals and people [8, 9].

The research was conducted to assess the effect of artificial soil pollution with polycyclic aromatic hydrocarbons on the amount of produced maize biomass and its accumulation of cadmium and lead.

Material and methods

The investigations were conducted in 2009–2010 as a pot experiment on soil material collected from Ap (0–20 cm) layer of an arable field.

The experimental soil was polluted with three hydrocarbons from PAH group: benzo(a)pyrene, chrysene and fluorene with diversified physicochemical properties. Because of the fact that the soil environment is never polluted with single compounds of this group, their mixtures were used for the experiment [10–12]. Benzo(a)pyrene (BaP), chrysene (Ch) and fluorene (Fl) were added to the soil as liquids dosed 0.1 $mg \cdot kg^{-1}$ d.m. of each substance and 10 $mg \cdot kg^{-1}$ d.m. of each substance. An appropriate amount of PAHs was dissolved in dichloromethane. The experiments comprised: the control (K) – soil with natural concentrations of studied PAHs and without mineral salt supplement; object (0) – soil with natural concentrations of studied PAHs and mineral salt supplement, object (I) – soil with dichloromethane and mineral salts supplement, object (II) – soil with a supplement of 0.3 $mg \cdot kg^{-1}$ of soil d.m. PAHs (0.1 mg BaP + 0.1 mg Ch + 0.1 mg Fl) + mineral salts – the amount of PAHs introduced to the soil on this object was level with elevated content, and object (III) – soil with an addition of 30 $mg \cdot kg^{-1}$ soil d.m. of PAHs (10 mg BaP + 10 mg Ch + 10 mg Fl) + mineral salts, PAH quantity introduced to the soil on this object was equivalent to very strong pollution [13].

The experiment was conducted on the soil material of sandy silt loam grain size composition containing 26 % of the < 0.02 mm fraction. The soil material revealed a slightly acid soil reaction (pH H_2O = 6.27), hydrolytic acidity (Hh) assessed after soil extraction with 1 mol · dm^{-3} CH_3COONa solution was 23.9 mmol (+) · kg^{-1} of soil d.m. Organic carbon concentration was 15.99 g C · kg^{-1} of soil d.m. and total nitrogen 1.54 g N · kg^{-1} of soil d.m.

The pot experiment was conducted in PCV containers to which 8.6 kg of air-dried soil material was weighted. In order to meet the plant nutritional requirements, the soil of all objects, except the control (0), received nutrients in the form of chemically pure salts containing nitrogen, phosphorus and potassium. The quantities of nutrients (N, P, K) introduced per 1 kg of soil were respectively 0.12 g N (NH_4NO_3); 0.06 g P ($Ca(H_2PO_4)_2 \cdot H_2O$); 0.19 g K (KCl). The research was conducted in four replications; the soil material moisture during the vegetation period was maintained on the level of 60 % soil water capacity. The test plant was maize “San” c.v. Five plants per pot were kept until harvest which was done at the stage of 7–9 leaves.

Following maize shoots harvest, roots were taken from the soil lump, washed and then the plant material was dried in an air flow dryer (at 70 °C) until a constant weight in order to determine the dry weight. Subsequently, dried biomass was crushed in a laboratory mill and mineralized in a chamber furnace (at 450 °C for 5 hours). The residue was dissolved in a diluted nitric acid 1:2 (v/v) [14]. The content of studied trace elements was assessed in solutions prepared in this way by means of ICP-AES method on JY 238 Ultrace apparatus.

The quantities of absorbed trace elements was computed on the basis of biomass amounts and a component concentration in the biomass. On the basis of a total maize biomass (shoots and roots) the tolerance index was computed as a ratio of the plant yield dry mass in the objects 0, I, II and III and in the object where mineral medium was introduced to the unpolluted soil (object 0). The indicator of pollution degree was calculated on the basis of the element concentrations in plant shoots and as a ratio of the element content in plants from objects K, I, II and III and in the object in which a mineral medium was introduced to the unpolluted soil (object 0). Translocation coefficient was calculated as a product of the element content in plant shoots and roots [15].

Analysis of plant material was conducted in four replications. The precision of the assessments was determined using reference material NCS DC733448 (China National Analysis Center for Iron & Steel). The data concerning the precision and exactness of the assessments were given in Table 1 [16].

Table 1

Amounts (mean ±SD) of metals released for material NCS DC733448, as well as data for analytical precision and accuracy

Metal	The value obtained in current study [mg · kg^{-1} d.m.]	Recommended value [mg · kg^{-1} d.m.]	Precision	Accuracy
Pb	6.8 ± 0.1	7.1 ± 1.1	1.47	–4.22
Cd	0.18 ± 0.01	0.14 ± 0.06	5.55	28.57

The obtained results were elaborated statistically according to a constant model in which a factor was the level of PAH pollution. Conducted statistical computations considered a one-way ANOVA and the significance of differences between arithmetic means was estimated by means of t-Tukey’s test at the significance level $\alpha < 0.05$ [17].

Results and discussion

The effect of soil pollution with PAHs on plant growth and development depends not only on the species but also, as stated by Maliszewska-Kordybach and Smreczak [18], mainly on the soil properties, primarily on its organic matter concentration. In the presented experiment, the amount of maize biomass in the objects where the soil was polluted with PAHs was bigger than on the control (unpolluted soil and without mineral salt supplement – object K) (Table 2). Therefore it may be stated that soil pollution with the analyzed PAHs did not inhibit maize growth or development. An analysis of maize biomass (shoots and roots) revealed the greatest amount in the object where the soil was characterized by a lower level of pollution – object II. Also Maliszewska-Kordybach and Smreczak [18] described a stimulating effect of PAHs on plant yield at the level of these substances below or slightly exceeding 1 mg · $kg^{-1}$of soil d.m. Kummerova et al [19] demonstrated that PAHs concentration not exceeding 10 mg · kg^{-1} d.m. in a solution may intensify plant biomass increment.

Table 2

The amount of biomass of maize [g d.m. · pot^{-1} ± SD] and the value of tolerance coefficient (mean ± SD)

Objects	Parts aboveground	Roots	Total biomass yield	Tolerance coefficient
K	$72.2^{a} \pm 4.0$	$12.6^{a} \pm 2.4$	$84.8^{a} \pm 6.3$	$0.58^{a} \pm 0.04$
0	$127.9^{b} \pm 4.1$	$17.6^{ab} \pm 1.1$	$145.5^{b} \pm 4.3$	1.00*
I	$135.3^{b} \pm 5.5$	$19.1^{ab} \pm 3.7$	$154.4^{b} \pm 9.1$	$1.06^{b} \pm 0.07$
II	$140.0^{b} \pm 7.0$	$20.4^{b} \pm 3.5$	$160.4^{b} \pm 10.0$	$1.10^{b} \pm 0.07$
III	$133.3^{b} \pm 1.9$	$17.8^{ab} \pm 0.9$	$151.2^{b} \pm 2.5$	$1.04^{b} \pm 0.03$

* Object 0 = 1.00. Means followed by the same letters in columns did not differ significantly at $\alpha < 0.05$ according to the t-Tukey test.

Beside the assessment of plant biomass quantity in conditions of increased stressor content in soil, also various other indices are being tested, which could more reliably show the effect of stress agent. The value of tolerance index in the objects where the stress agent was introduced (objects II and III) in the presented experiment was above one, which points to the absence of the effect of soil pollution with PAHs on the amount of plant biomass (Table 2). Lower than one value of the discussed index was noted only for the biomass from the control (the soil with natural PAH content and without the mineral medium – object K), which may be associated with a deficiency of available nitrogen, phosphorus and potassium forms. Aromatic hydrocarbons with bigger weights and molecular structures similar to gibberilines may affect plants as growth stimulators. The mechanism of plant growth stimulation at small concentrations of toxic substances is explained by a specific evolution of physiological control in organisms in result of which an excessive response to small deviations from standard are observed [20]. This phenomenon is also interpreted by increasing microbiological activity in result of which a biodegradation of polycyclic aromatic hydrocarbons occurs, which leads to an improvement in conditions for plant development [20, 21]. PAHs effect on plants may result from the soil properties. The result of elevated PAHs toxicity is observed in plants cultivated in soils poor in organic matter and in acid soils. On the other hand, lesser plant sensitivity is observed in soils abundant in organic matter or fertilized with it, which may result from a stronger PAHs sorption by this soil element [22–24], and therefore a lesser availability to plants [21–25].

Lead content in maize shoots did not exceed 0.50 mg · kg^{-1} d.m. Biomass from the objects where the soil was polluted with the analyzed hydrocarbons contained smaller amounts of this element (Table 3). Lead contents in the roots proved between over three and eight times higher, whereas the highest were registered in maize root system on the control (K). Like in the shoots, the least quantities of lead were assessed in maize roots from the object in which the soil was the most contaminated with PAHs (object III).

Table 3

Content of trace elements in maize biomass [mg · kg^{-1} d.m. ± SD]

Objects	Parts aboveground	Roots	Parts aboveground	Roots
	Cd		Pb	
K	$0.20^{a} \pm 0.01$	$2.57^{b} \pm 0.43$	$0.40^{b} \pm 0.06$	$3.11^{c} \pm 0.66$
0	$0.27^{a} \pm 0.05$	$1.43^{a} \pm 0.06$	$0.40^{b} \pm 0.10$	$2.50b^{c} \pm 0.44$
I	$0.43^{bc} \pm 0.02$	$1.02^{a} \pm 0.10$	$0.43^{b} \pm 0.10$	$1.59^{ab} \pm 0.34$
II	$0.37^{b} \pm 0.02$	$1.09^{a} \pm 0.07$	$0.26^{ab} \pm 0.04$	$2.07^{abc} \pm 0.42$
III	$0.47^{c} \pm 0.06$	$0.92^{a} \pm 0.13$	$0.19^{a} \pm 0.05$	$1.27^{a} \pm 0.27$

Means followed by the same letters in columns did not differ significantly at $\alpha < 0.05$ according to the t-Tukey test.

The amounts of lead taken up by maize shoots were the resultant of the biomass quantity and this element content (Table 4). Maize shoots absorbed the smallest quantities of lead in the object where the soil was contaminated by a bigger amount of the studied hydrocarbons (object III). A similar tendency was observed for maize root system.

Table 4

Uptake of trace elements with maize biomass [mg · pot^{-1} ± SD]

Objects	Parts aboveground	Roots	Parts aboveground	Roots
	Cd		Pb	
K	$0.014^{a} \pm 0.01$	$0.032^{b} \pm 0.01$	$0.029^{ab} \pm 0.01$	$0.041^{b} \pm 0.02$
0	$0.034^{b} \pm 0.01$	$0.025^{ab} \pm 0.01$	$0.051^{bc} \pm 0.01$	$0.044^{b} \pm 0.01$
I	$0.059^{c} \pm 0.01$	$0.019^{a} \pm 0.01$	$0.059^{c} \pm 0.01$	$0.032^{b} \pm 0.01$
II	$0.052^{c} \pm 0.01$	$0.022^{a} \pm 0.01$	$0.036^{abc} \pm 0.01$	$0.043^{b} \pm 0.02$
III	$0.063^{c} \pm 0.01$	$0.016^{a} \pm 0.01$	$0.026^{a} \pm 0.01$	$0.022^{a} \pm 0.01$

Means followed by the same letters in columns did not differ significantly at $\alpha < 0.05$ according to the t-Tukey test.

The index of maize shoot biomass pollution with lead was higher than one only in the object where dichloromethane (object I) was introduced to the soil, indicating lead accumulation in the biomass due to the applied agent (Table 5). The value of the discussed parameter for maize shoots from the other objects was much below one.

Table 5

The value of contamination coefficient parts aboveground of maize with trace elements (mean ± SD)

Objects	Cd	Pb
K	$0.43^{a} \pm 0.07$	$0.58^{a} \pm 0.08$
I	$1.79^{bc} \pm 0.48$	$1.27^{a} \pm 0.62$
II	$1.58^{bc} \pm 0.32$	$0.73^{a} \pm 0.16$
III	$1.91^{c} \pm 0.40$	$0.55^{a} \pm 0.22$

* Object 0 = 1.00. Means followed by the same letters in columns did not differ significantly at $\alpha < 0.05$ according to the t-Tukey test.

Lead translocation coefficient was much below one and did not reveal any significant diversification among the objects (Table 6), which evidences a lack of soil contamination with PAHs effect on this element migrations to maize shoots.

Table 6

The value of translocation coefficient trace elements (mean ± SD)

Objects	Cd	Pb
K	$0.08^{a} \pm 0.02$	$0.13^{a} \pm 0.03$
0	$0.19^{ab} \pm 0.04$	$0.16^{a} \pm 0.03$
I	$0.43^{cd} \pm 0.05$	$0.28^{b} \pm 0.06$
II	$0.34^{bc} \pm 0.03$	$0.13^{a} \pm 0.02$
III	$0.54^{d} \pm 0.14$	$0.16^{a} \pm 0.05$

Means followed by the same letters in columns did not differ significantly at $\alpha < 0.05$ according to the t-Tukey test.

Cadmium content in maize dry mass depended on the plant part and soil pollution with aromatic hydrocarbons (Table 3). More of this element was assessed in the root system. Maize shoots in the objects where dichloromethane was supplied to the soil and in the object where the soil was contaminated with benzo(a)pyrene, chrysene and fluorene contained significantly biggest cadmium quantities. Assuming that the value of $0.15 \text{ mg} \cdot \text{kg}^{-1}$ dry mass has been regarded as permissible in the biomass destined for consumption, the amount should be seen as considerably exceeded [26]. From the point of view of the biomass intended for fodder, cadmium concentration in the analyzed biomass did not raise objections. Maize root system contained much more of cadmium. The highest amounts were registered in root biomass from the control (K).

Amounts of cadmium taken up by maize shoots were significantly bigger in the objects in which aromatic hydrocarbons were supplied to the soil (II and III) and in the object in which dichloromethane was added to the soil (I). The amounts of cadmium taken up by maize root system were smaller, whereas maize root system absorbed the greatest quantities of cadmium in the control (K) (Table 4).

The indices of the degree of maize shoots pollution with cadmium were above one in all objects, except the control, whereas the highest value of this parameter was registered in the object where the soil was polluted with a bigger quantity of aromatic hydrocarbons (object III) (Table 5).

The values of translocation coefficient for this element were characterized by a relatively low diversification among the objects and confirmed a lower cadmium accumulation in maize shoots as compared with roots (Table 6).

Discussion of the research results of heavy metal content in maize in conditions of soil pollution with polycyclic aromatic hydrocarbons is made difficult by a limited number of literature positions addressing the presented issue. The problem of heavy metal bioavailability has been widely discussed in the aspect of these elements effect on biochemical processes occurring in a plant in the context of remediation of the chemically polluted lands, but also at the environmental application of waste materials [27–29]. Plants growing in the polluted environment may accumulate considerable amounts of toxic trace elements, which poses a serious hazard for animals and people [30, 31].

Heavy metal detoxification mechanisms developed by plants, after taking them up from soil solution allow these organisms to function in the polluted environment without any visible symptoms of phytotoxicity [7]. Many investigations demonstrated considerable differences in trace element uptake depending on the soil grain size composition, pH, organic matter content, or sorption capacity, however plant species is not without importance, either [26, 32]. According to Joung and Thorton [33] and Rosselli et al [34] increasing the soil pH value and organic matter content results in diminishing trace elements availability. However, Khan et al [35] and Young et al [36] call attention to the fact that trace element concentrations in plants may be also significantly influenced by the soil pollution with among others polycyclic aromatic hydrocarbons. The presented investigations make possible a statement that trace elements were accumulated mainly in maize root system. Investigations conducted by MacNicol and Beckett [37] confirm that roots constitute the first barrier restricting trace element translocation to the shoots, irrespectively of the stressor, although as stated by Batty and Anslow [38], there are plants which accumulate more of trace elements in the shoots. The barriers on the way of trace element transport from the roots to the shoots, generally act effectively in all plants towards lead. On the other hand, considering cadmium, they obviously depend on the cultivated plant species. According to Chu and Wong [39] such barriers exist also on the way of metal transport within the shoots. Batty and Anslow [38] revealed that soil contamination with pyrene did not reduce zinc uptake, nevertheless zinc and pyrene application to the soil markedly decreased Brassica juncea growth.

In the conducted experiment, cadmium concentrations in maize shoots were higher in the objects where dichloromethane and polycyclic aromatic hydrocarbons were introduced to the soil. As stated by Hart et al [40] higher content of cadmium in plant shoots may be connected with the role of phloem in this element transport to the shoots.

Conclusion

Soil pollution with the analyzed aromatic hydrocarbons did not inhibit either growth or development of maize roots or shoots. The greatest amount of biomass was obtained in the object where the soil was characterized by elevated content of the analyzed aromatic hydrocarbons . The value of tolerance index in the objects where stressor was introduced was above one, which indicates a lack of soil pollution with PAHs effect on plant biomass quantity. The value of tolerance index below one referred only to biomass from the control. Significant increase in Cd content and its quantities taken up by maize shoots was registered on the treatments where dichloromethane and polycyclic aromatic hydrocarbons were added to the soil in comparison with the unpolluted objects. The content and amounts of absorbed lead were the lowest on the object where the soil was the most polluted. Values of maize shoot biomass Cd pollution index were apparently higher in the objects where the soil was contaminated with polycyclic aromatic hydrocarbons in comparison with the values obtained in the object where soil received only mineral nutrient solution. Values of translocation index do not point to cadmium accumulation in plant shoots. Both the values of pollution index and translocation index for lead in the objects with elevated PAHs content in soil and in the object where the soil was contaminated with these substances were below one, which did not confirm excessive lead accumulation in maize shoots.

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WPŁYW ZANIECZYSZCZENIA GLEBY WWA NA ILOŚĆ BIOMASY KUKURYDZY ORAZ AKUMULACJĘ KADMU I OŁOWIU

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Abstrakt: Biodostępność pierwiastków śladowych ma kluczowe znaczenie w kwestii stymulacji lub hamowania procesów wzrostu i rozwoju roślin w późniejszych etapach ich życia, a w konsekwencji prowadzi do zmian wartości biologicznej biomasy np. ograniczając jej wykorzystanie. Ocenę wpływu sztucznego zanieczyszczenia gleby benzo(a)pirenem, chryzenem oraz fluorenem na ilość biomasy kukurydzy oraz akumulację w niej wybranych pierwiastków śladowych przeprowadzono w doświadczeniu wazonowym.

Zanieczyszczenie gleby badanymi węglowodorami aromatycznymi nie hamowało wzrostu i rozwoju części nadziemnych i korzeni kukurydzy. Największą ilość biomasy uzyskano w obiekcie, w którym gleba charakteryzowała się podwyższoną zawartością badanych węglowodorów aromatycznych. Wartość wskaźnika tolerancji w obiektach, w których wprowadzono czynnik stresowy, kształtowała się powyżej jedności, co wskazuje na brak wpływu zanieczyszczenia gleby benzo(a)pirenem, chryzenem oraz fluorenem na ilość biomasy roślin. Wartość wskaźnika tolerancji poniżej jedności dotyczyła jedynie biomasy z obiektu kontrolnego. Istotnie większe zawartości Cd oraz ilości pobrane tego pierwiastka przez części nadziemne kukurydzy stwierdzono w obiektach, w których do gleby wprowadzono dichlorometan i wielopierścieniowe węglowodory aromatyczne w porównaniu do obiektów niezanieczyszczonych. Zawartość i ilości pobranego ołowiu były najmniejsze w obiekcie, w którym gleba była najbardziej zanieczyszczona. Wartości wskaźnika zanieczyszczenia biomasy części nadziemnych kukurydzy Cd były wyraźnie większe w obiektach, w których glebę zanieczyszczono węglowodorami aromatycznymi w porównaniu do wartości, jakie uzyskano w obiekcie, w którym do gleby wprowadzono tylko pożywkę mineralną. Wartości wskaźnika translokacji nie wskazują na nagromadzenie kadmu w częściach nadziemnych roślin. Zarówno wartości wskaźnika zanieczyszczenia, jak również wskaźnika translokacji dla ołowiu, w obiektach o podwyższonej zawartości WWA w glebie oraz w obiekcie, w którym glebę skażono tymi substancjami kształtowały się poniżej jedności nie potwierdzając nadmiernego nagromadzenia ołowiu w częściach nadziemnych kukurydzy.

Słowa kluczowe: gleba, zanieczyszczenie, wielopierścieniowe węglowodory aromatyczne, kadm, ołów