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ASSESSMENT OF HEAVY METAL CONTAMINATION IN TOPSOIL AND SOIL INVERTEBRATES FROM THE NIEPOLOMICE FOREST

OCENA ZAWARTOŚCI METALI CIĘŻKICH W GLEBIE I BEZKRĘGOWCACH GLEBOWYCH W PUSZCZY NIEPOŁOMICKIEJ

Abstract: The purpose of the research was to determine the respective contents of heavy metals (Pb, Cd, Ni, Zn and Cu) in soil and extracted soil fauna in humid mixed coniferous forest (MHCF), fresh mixed coniferous forest (MFCF) and in fresh mixed forest (MFF) of Niepolomice Forest. The contents of heavy metals varied, depending on the forest type and trophic group. The highest values of cadmium, lead, and copper content were recorded in MFCF, while of nickel in MHCF. Analysis of variance and post-hoc Tuckey's showed significant differences between the concentrations of Zn and other metals in invertebrates in different types of forest habitats. Bioconcentration factor shows that both soil saprophages and predacious invertebrates accumulated the largest amounts of Cd and smaller amounts of Zn, while the accumulation of remaining metals depended on the type of forest habitat. In fresh mixed coniferous forest predators accumulated heavy metals in the following order: Cd > Zn > Pb > Ni > Cu, in the case of saprohages it was: Cd > Zn > Ni > Cu > Pb.

Keywords: Niepolomice Forest, heavy metals, trophic groups, saprophags, predators, bioconcentration factor values

Introduction

Soil is a major element of all land ecosystems. Processes taking place there are crucial for their sustainability and efficiency [1]. One of its characteristic features is the abundance and diversity of organisms found in it. In one square meter of soil one can find up to 1000 species of pedofauna, including approximately 200 species of arthropods. Invertebrates play significant roles in the delivery of ecosystem services by soils at plot. They participate actively in the interactions that develop in soil among physical, chemical and biological processes. Soil invertebrates are enormously diverse

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[2]. According to recent estimations, soil animals may represent as much as 23% of the total diversity of living organisms that has been described to date [3].

Anthropogenic activity brings about essential changes in the circulation of elements in the environment, which leads to the pollution by these elements of individual environmental components. Particularly hazardous is the process of accumulation of cationic trace elements of heavy metal groups. Heavy metals in excessive amounts pose serious risk to plants, animals and human beings [4–8]. The presence of trace metals in the soil, plants and in soil fauna is an environmental indicator [2, 9–11]. Accumulation of heavy metals in soil is conditioned by the form of land use as well as physical and chemical properties of the soil and by weather conditions [12, 13].

That is why there is a need to systematically monitor their concentrations in environmental components [14]. Since invertebrates are very susceptible to changes of soil conditions, they may be considered valuable indicators of soil disturbances [15–23].

The purpose of the research was to determine the respective contents of heavy metals (Pb, Cd, and Ni, Zn and Cu) in soil and extracted soil fauna in three types of forest habitats – fresh mixed coniferous forest, humid mixed coniferous forest and in fresh mixed forest of Niepolomice Forest.

Materials and methods

Niepolomice Forest is a huge forest complex situated approx. 25 km to the east of Krakow. Forests cover nearly 90% of its area. Niepolomice Forest is dominated by pine coniferous forests: mixed fresh coniferous forest (MFCF), mixed humid coniferous forest (MHCF), marshy coniferous forest (MCF); mixed fresh forest (MFF), mixed humid forest (MHF), mixed marshy forest (MMF), alder forest (AF), and ash-alder forest (AAF).

The research was carried out in three types of forest habitats: in mixed humid coniferous forest (MHCF) which occupies the largest area in Niepolomice Forest, in mixed fresh coniferous forest (MFCF) (9.8%), and in mixed fresh forest (MFF) (10.5%) [24]. The localities of taking of the samples were marked in Fig. 1.

Soil samples were taken during autumn of 2011–2012 vegetation season, in tree stands of age category III (called stickstand), where trees have between 40 to 60 years of age.

Soil samples were collected from the surveyed positions with soil frames sized 25 × 25 cm and with 1 m^2 surface area and absent of soil fauna using the dynamic method with a modified Tullgren apparatus. Soil fauna were separated into trophic groups – saprophags, predators, and phytosaprophags – in which heavy metals were then marked. The heavy metal content in soil and soil fauna was determined by FAAS after previous mineralization of the soil and animal test materials. For this purpose, animals and soil samples were dried at 105^{o}C to obtain a dry weight. After obtaining the dry weight, each trophic group was weighed. Also, 2 g of dried soil from each location was weighed.

The soil and soil organisms underwent the mineralization process in a Velp Scientifica DK-20 mineralizer in concentrated nitric acid at 120^{o}C until the tissue was completely dissolved. Then, the resulting solutions were poured into measuring flasks filled with distilled water up to 10 cm^3. In the solutions thus prepared, the heavy metal content was determined content – cadmium, lead, nickel, copper and zinc in a spectro-

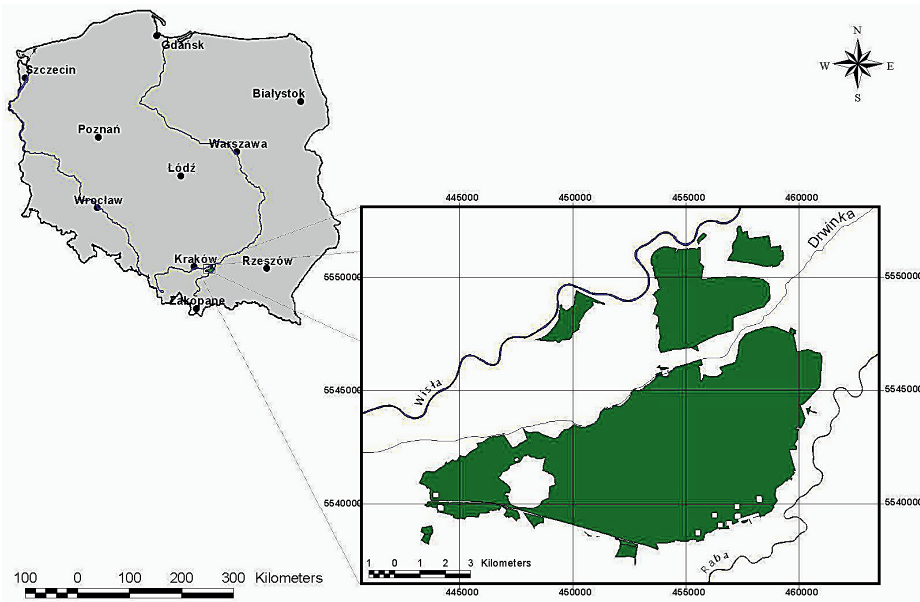

Fig. 1. Location of the study area in Niepolomice Forest

photometer. The heavy metal content in soil and soil fauna was determined by Buck Scientific 200A Flame Atomic Absorption Spectrophotometer FAAS. The AAS limits of detection (LoD) and quantification (LoQ) for Pb were set at 0.027 ppm and 0.083 ppm (mg/dm^3), respectively, for Cd 0.011 ppm (LoD) and 0.033 ppm (LoQ), for Ni 0.017 ppm (LoD) and 0.05 ppm (LoQ), for Cu 0.012 (LoD) and LoQ = 0.035 ppm and for Zn: LoD = 0.023 ppm and LoQ = 0.069 ppm. Reference material was BCR-185, Bovin Liver.

Moisture content and pH of the investigated soils were determined as well. PH of soil was measured using a WTW 330/SET-1 pH-meter (Wissenschaftlich – Technische Werkstatten 82362 Weilheim).

The paper also analyzes the significance of differences between the concentration of heavy metals in soil fauna in three types of forest habitat MFCF, MHCF and MFF. After showing the statistical significance ($p < 0.05$) a Tuckey's test was chosen [25]. The content of heavy metal in studied soil of forest habitats were compared using t-Student's test, and in the case of heterogeneous variance was used t-Student's test with the separate estimation of the variance [26]. The differences were statistically significant at $p < 0.05$. All the analyses were performed using STATISTICA 10 computer program.

Results and discussion

The analysis of the pH value showed that the studied soils had acidic pH, from 4.5 to 5.5. The types of soil found in the analysed sites included: podzolic soils, podzolic and muck soils, muck and gley soils, brown acid soils (Table 1).

Table 1

Comparison of studied habitat types of the forest (±SD)

Parameters	MFF	MHCF	MFCF
Soil pH	5.5 ± 0.2	4.5 ± 0.3	4.8 ± 0.3
Soil temperature [°C]	3.1 ± 0.1	3.6 ± 0.2	4.9 ± 0.3
Area temperature [°C]	7.7 ± 0.4	7.4 ± 0.6	5.4 ± 0.4
Soil moisture [%]	10.2 ± 2.3	7.5 ± 1.5	7.9 ± 1.9
Sort of soil	brown acids, podzolic	podzolic and muck, muck and gley	podzolic

The highest values of cadmium, lead, and copper content were recorded in MFCF, while of nickel in MHCF. The largest differences in content in the analysed types of forest habitats were observed with respect to lead. The overall lead content in the soil of MFF and MFCF was nine times higher than in soil of MHCF. The concentration of this metal in the soil MHCF and MFF was statistically significant ($p < 0.001$) (Table 2).

Table 2

Concentration of heavy metals in soil [$mg \cdot kg^{-1}$ d.m.]
(Mean, SD, significant differences between the concentration of metals in soil)

Metals	MFCF			MHCF			MFF			Limit values[a]
	Mean	SD	p	Mean	SD	p	Mean	SD	p	
Cd	0.8	0.7	ND	0.7	0.1	*	0.5	0.2	*	4
Pb	78.1	25.3	ND	8.4	0.5	***	74.4	12.4	***	100
Ni	6.4	0.2	ND	12.4	1.8	***	7.1	3.2	***	100
Cu	15.9	1.6	ND	10.4	1.6	**	8.8	3.7	**	150
Zn	89.4	20.8	ND	71.6	11.4	NS	92.6	19.8	NS	300

[a] Limit values for the heavy metal content set out in the Minister of Environment Regulation on the soil quality standards and earth quality standards for group B– (Polish Journal of Laws 2002 No. 165, item 1359 of 4 October 2002); * 0,05 > p > 0.01; ** 0.01 > p > 0.001; *** p < 0.001; NS – no significant differences; ND – no data.

It should be pointed out that the accumulation of lead in forest litter is a long-lasting process, hence the found lead concentrations may result from the total pollutant amounts from deposition lasting many years. The results indicate that the concentrations of heavy metals do not exceed the standards set out in the Regulation of the Minister of the Environment on soil quality standards and earth quality standards [27]. Rozen et al [19] showed in their study in mixed oak-pine in Niepolomice forest much higher values of Cd, Pb, Cu and Zn.

In much of the soil was cumulated zinc (71–92 $mg \cdot kg^{-1}$ d.m.), but its content in the studied forest was not statistically significant ($p > 0.05$). In a smaller were cumulated lead, copper, cadmium, nickel, which the concentrations in the soil were statistically significant (Table 2). Higher average content of nickel (67.5 $mg \cdot kg^{-1}$ d.m.) in soils in coniferous forest in Olsztyn (northeastern Poland) found Modrzewska and Wyszkowski [13].

The analysed metal content in the studied soils did not correlate with the abundance and the biodiversity of the studied fauna. The highest density was recorded in fresh mixed forest MFF, and the lowest in fresh mixed coniferous forest MFCF. The largest systematic group were Acarina, which accounted for 37% of the MHCF to 83% in the total number of MFCF. Collembola were also large numbers until reaching in MFF 10880 $N \cdot m^{-2}$ (Table 3). Also, other authors showed a similar arrangement of taxonomic groups in their studies [10, 20].

Table 3

Density of pedofauna [$N \cdot m^{-2}$]

Systematic groups	Trophic groups[a]	MHCF	MFCF	MFF
Enchytraeidae	S	1 688		296
Lumbricidae	S	8		56
Isopoda	S			8
Collembola	S, P	7 688	120	10 880
Diptera l.[b]	S	232	8	96
Aranea	P	8		8
Symphyla	P			80
Chilopoda	P	40	16	72
Coleoptera im.[c]	P	16	32	56
Carabidae l.	P	216	200	32
Staphylinidae l.	P			56
Rhagionidae l.	P			48
Dolichopodidae l.	P		104	32
Thysanoptera	Ph		64	
Hymenoptera	P		80	272
Curculionidae l.	Ph			8
Bibionidae l.	Ph	24		
Cecidomyiidae im.	Ph	48		48
Acarina	S, P	5 992	3 008	13 160
Total		15 960	3 432	25 208

[a] Trophic groups: S – saprophag, P – predator, Ph – phytophag; [b] l. – larvae; [c] im. – imago.

Also tested was the trophic structure of pedofauna assemblages. In mixed humid coniferous forest and in mixed fresh forest were dominated by predatory invertebrates. The density of predatory forms ranged from 280 $N \cdot m^{-2}$ in MHCF to 656 $N \cdot m^{-2}$ in mixed forest fresh (Table 3). However, in mixed humid coniferous forest dominated by saprophags invertebrates, whose share was almost 85%.

The contents of heavy metals varied, depending on the forest type and trophic group. As regards lead, the largest values – between 40 $mg \cdot kg^{-1}$ d.m. and 220 $mg \cdot kg^{-1}$ d.m. – were observed in predators, while in phytophags and saprophages they ranged from 29 to 64 ad in earthworn found Rozen et al [19].

Cadmium content was higher in predators than in phytophags and saprophages and ranged from 8.0 to 22.6 mg · kg^{-1} d.m. The higher cadmium concentration was recorded in earthworm from oak-hornbeam in the northern part of the Niepolomice Forest [19]. Jelaska et al [20] showed that predators Carabides accumulated less cadmium (0.2–10.6) and lead (0.13–2.31) in forest ecosystem in Medvednicz Nature Park.

Nickel was present in the amount of 6.6 to 31.9 mg · kg^{-1} d.m., with higher values observed in predators than in saprophages and phytophags (Table 4). The highest copper contents were found in predacious invertebrates (91 mg · kg^{-1} d.m. in MHCF). Also Chrzan et al [28] showed a higher content of Pb, Ni and Cu in predatory invertebrates in forest soils.

Table 4

Content of heavy metals in trophic groups [mg · kg^{-1} d.m.] (±SD)

Metal	Saprophytophags			Predators		
	MFCF	MHCF	MFF	MFCF	MHCF	MFF
Cd	7.98 ± 1.19	13.73 ± 3.56	17.89 ± 2.34	22.61 ± 3.53	12.22 ± 1.58	13.64 ± 0.43
Pb	29.39 ± 4.6	64.38 ± 5.24	38.77 ± 15.92	220.04 ± 77.87	79.42 ± 22.11	40.03 ± 2.71
Ni	6.60 ± 0.23	10.13 ± 1.29	13.21 ± 1.59	14.78 ± 1.84	27.27 ± 4.19	31.92 ± 1.79
Cu	15.84 ± 2.53	17.99 ± 0.92	29.67 ± 4.69	18.75 ± 3.14	91.05 ± 10.97	35.79 ± 4.33
Zn	280.89 ± 36.79	860.18 ± 210.15	1081.15 ± 111.02	779.56 ± 83.41	937.96 ± 343.22	538.46 ± 156.35

Zinc is crucial for proper functioning of all cells of an organism and is present in many enzymes responsible, *inter alia*, for metabolism of proteins, carbohydrates, and fats. Zinc content in organisms is high. The highest value of 1081 mg · kg^{-1} d.m. was found in soil saprophages and phytophags. The high content of Zn in saprophagous Enchytraeides demonstrated by Rozen et al [19]. Heavy metal toxicity depends on the roles they play in the metabolic processes of the organisms and their susceptibility to bioaccumulation.

Significant correlations between the studied metals in each types of forest habitats separately demonstrated. Analysis of variance and post-hoc Tuckey's showed significant differences between the concentrations of Zn and Pb, Cd and Cu in invertebrates in different types of forest habitats (Table 5).

Table 5

Significant differences between the concentration of Pb, Cd, Zn, Cu [mg · kg^{-1} d.m.] in soil fauna of MFF and MHCF

MFCF	p	MHCF	p	MFF	p
Pb vs Cd	NS	Pb vs Cd	NS	Pb vs Cd	NS
Pb vs Zn	***	Pb vs Zn	**	Pb vs Zn	***
Pb vs Cu	NS	Pb vs Cu	NS	Pb vs Cu	NS
Cd vs Zn	**	Cd vs Zn	***	Cd vs Zn	**
Cd vs Cu	NS	Cd vs Cu	NS	Cd vs Cu	NS
Zn vs Cu	***	Zn vs Cu	***	Zn vs Cu	**

* $0.05 > p > 0.01$; ** $0.01 > p > 0.001$; *** $p < 0.001$; NS – no significant differences.

The potential risk may be expressed through the accumulation index representing the ratio between the average concentration of the element in the body to its content in the soil. Bioconcentration factor calculated for predatory and saprophytophag invertebrates, and presented in Table 6.

Table 6

Bioconcentration factor values (BCF) of the potentially toxic trace elements in saprophytophags and predators

Metal	Saprophytophags			Predators		
	MFCF	MHCF	MFF	MFCF	MHCF	MFF
Cd	10.0	19.7	35.8	28.2	17.4	27.2
Pb	0.4	7.7	0.5	2.8	9.4	0.5
Ni	1.0	0.8	1.8	2.3	2.2	4.1
Cu	1	1.7	3.4	1.2	8.8	4.5
Zn	3.1	12	11.8	8.7	13.1	5.9

Bioconcentration factor shows that both soil saprophags and phytophags and predacious invertebrates accumulated the largest amounts of Cd and smaller amounts of Zn, while the accumulation of remaining metals depended on the type of forest habitat and the trophic group.

In fresh mixed coniferous forest predators accumulated heavy metals in the following order: Cd > Zn > Pb > Ni > Cu, in the case of saprophytophags it was: Cd > Zn > > Ni = Cu > Pb. As regards fresh mixed forest, the lowest BCF value was recorded for Pb. In humid mixed coniferous forest, BCF with respect to Cd, Pb and Zn was similar both in saprophytophags and predators invertebrates. Assimilability of heavy metals by soil fauna in this type of forests was as follows: Cd > Zn > Pb > Cu > Ni (Table 6).

Conclusions

1. Content of the heavy metal in the forest soils did not exceed the limit values for forest land according to the Regulation of the Minister of Environment on standards for soil quality and earth quality standards of 4.10.2002.
2. Soil invertebrates accumulated the most of Cd and a little less of Zn.
3. Significant differences were noted between the content of Zn in soil and fauna.
4. More of heavy metals were accumulated in predatory invertebrates in humid mixed coniferous forest MHCF and in fresh mixed coniferous forest MFCF than in saprophytophags.

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OCENA ZAWARTOŚCI METALI CIĘŻKICH W GLEBIE I BEZKRĘGOWCACH GLEBOWYCH W PUSZCZY NIEPOŁOMICKIEJ

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Abstrakt: Celem badań było określenie zawartość metali ciężkich (Pb, Cd, Ni, Zn i Cu) w glebie i wyekstrahowanej faunie glebowej w trzech typach siedliskowych lasu w Puszczy Niepołomickiej – w lesie mieszanym wilgotnym, borze mieszanym świeżym i borze mieszanym wilgotnym. Zawartość metali ciężkich uzależniona była od typu lasu i grupy troficznej. Najwieksze wartości kadmu, ołowiu i miedzi odnotowano w glebie w borze mieszanym świeżym BMśw, a niklu w borze mieszanym wilgotnym BMw. Wskaźnik biokoncentracji wskazuje, że zarówno saprofagi i drapieżne bezkręgowce glebowe w największej ilości kumulowały Cd, w mniejszej Zn, natomiast pozostałe metale kumulowane były w zależności od typu siedliskowego lasu. W borze mieszanym świeżym drapieżne kumulowały metale w kolejności Cd > Zn > Pb > Ni > Cu, a saprofagi Cd > Zn > Ni > Cu > Pb.

Słowa kluczowe: Puszcza Niepołomicka, metale ciężkie, grupy troficzne, saprofagi, drapieżne, wskaźnik biokoncentracji