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Comparison of Fe, Zn, Mn, Cu, Cd and Pb concentration in spruce needles collected in the area of Gdansk and Gdynia in Northern Poland

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Abstract
The aim of the investigation was to assess the utility of spruce needles as a potential material to be used for monitoring the contamination level of the environment of Tricity agglomeration (represented by Gdansk and Gdynia). This aim was realized by determining the levels of selected essential elements indispensable for the life of living organisms, such as Fe, Zn, Mn and Cu, as well as toxic, namely Cd and Pb, in spruce needles collected in the locations in Gdynia and for comparison, in Gdańsk. Due to this, the collected samples of needles were dried, and next digested by microwave technique in order to prepare them for quantitative analysis by atomic absorption spectrometry. Moreover, the same metallic elements were determined in the soil samples collected under the spruces, from which needles were taken for the investigation. The concentrations of the studied elements were found in the range of mg/kg of dry mass in the following order: Fe, Mn, Zn, Pb, Cu and Cd (needles), and Fe, Mn, Cu, Zn, Pb and Cd (soils). By application of statistical methods (correlation, variance and principal component analyses), the differences in the elemental composition of spruce needles were identified, as well as sources of this differentiation.

1. INTRODUCTION

Nowadays, the growing problem for human population on Earth is monitoring of environment, since the extensive development of industry and agriculture has its significant impact on soil, plant kingdom, animals and human health. Therefore, the need to investigate the concentrations of metallic elements in various natural materials is very important. One of the good bio-indicators are needles originating from spruce (*Picea abies* (L.) H. Karst). In literature, there are numerous reports about studying the level of metals in needles of spruce, pine and other plants used as suitable bio-indicators of environmental contamination.

As example of such studies, the investigation of lead level and persistent organic pollutants in pine needles originating from a plant *Pinus mugo* Turra, collected in High Tatra mountains in Slovakia can serve [Chropenova *et al*. 2016]. It was found that lead content increased with needle age, as well as the correlation between Pb level in the studied needles and pollen abortivity was revealed too. In another study, six trace elements, namely Cd, Ni, Cr, Cu, Pb and Zn were determined in the selected seven alpine species and in two mosses growing in the Eastern Tibetan Plateau, in order to monitor their contamination [Bing *et al*. 2016]. The studies have revealed that according to the biological and enrichment factors, and based on the factor analyses results, Cd, Pb and Zn in the studied samples were impacted by the anthropogenic emission, while Cr, Cu and Ni were related to the level of these metals in soils.

The samples of moss (*Pleurozium schreiberi*), grass (*Avenella flexuosa*) and spruce (*Picea abies*) originating from Czech Republic were analysed for 39 elements by ICP-MS and ICP-AES, to compare these plants as bio-indicators of environmental pollution [Suchara *et al.* 2011]. It was revealed that at the scale of the whole country, moss was the best indicator of contamination sources, but also spruce needles on a local scale appeared to be well suited for the identification of urban zones as contamination sources. In another study, the toxic elements present in forest soils of the Jizera Mountain Region were evaluated [Kvacova *et al.* 2015]. It was demonstrated that enrichment factors proved the anthropogenic origin of Cd, Cu, Pb and

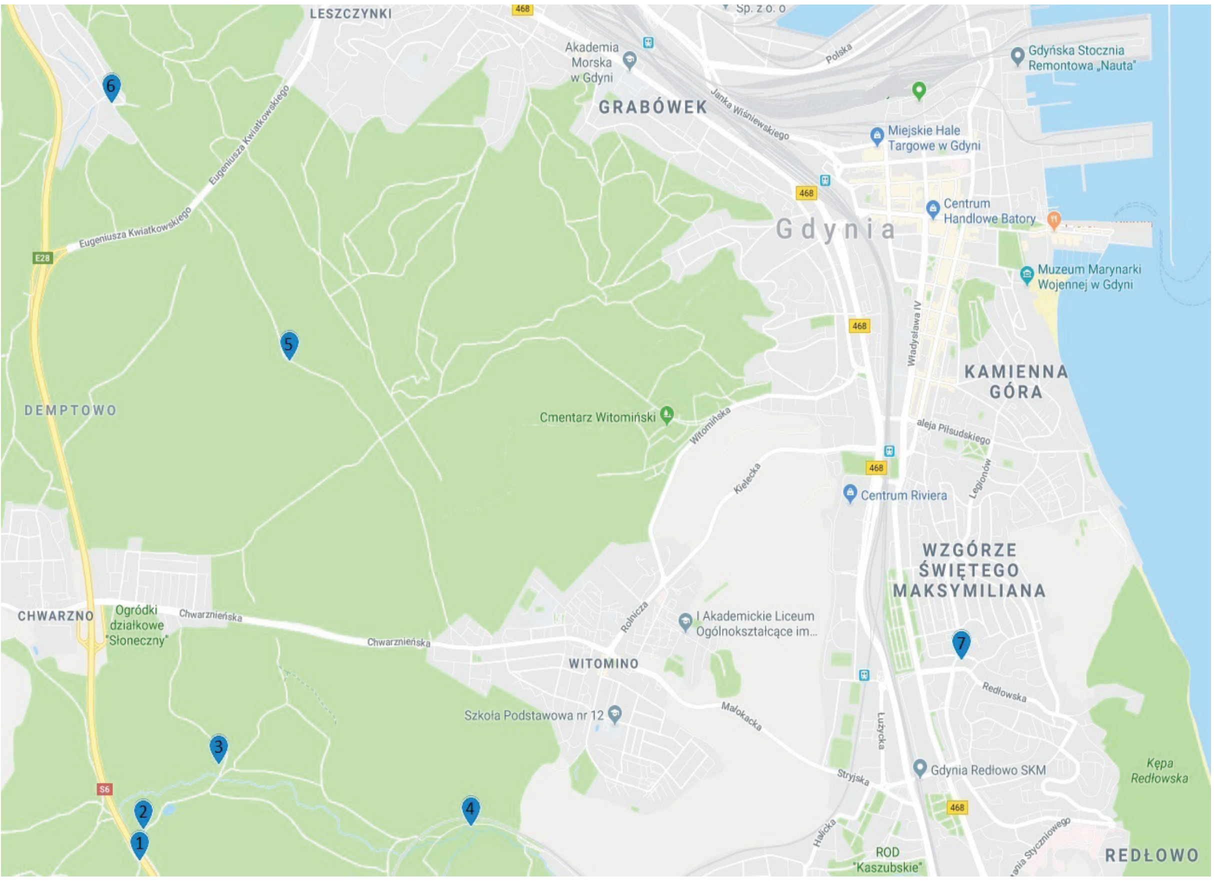

Figure 1. Map of harvest in Gdynia

Zn, while Mn was of geologic origin in the investigated samples.

There are other reports, in which the level of metallic elements, both essential and toxic were monitored to gather the knowledge about plants serving as bio-indicators of environmental contamination, and also about soil analyses [Łukasik *et al*. 2017, Jonczak and Parzych 2014].

Taking all the above points into consideration, the objective of the study was to assess the possibility to use the levels of selected essential elements indispensable for life of living organisms, such as Fe, Zn, Mn and Cu, as well as toxic, namely Cd and Pb, in spruce needles collected in locations in Gdynia and Gdansk, for monitoring the environmental pollution. Moreover, it was also interesting to relate the concentration of the studied metals in spruce needles with their level found in soil samples collected under the investigated trees. To fulfil these goals of study, metallic elements were determined both in needles and soil samples by flame atomic absorption spectrometric technique, and to interpret the obtained results, such statistical methods, as correlation analysis, one-way analysis of variance (ANOVA) and principal component analysis (PCA), were applied.

2. MATERIAL AND METHODS OF STUDY

Within the study spruce needles originated from 7 locations in Gdynia and 6 locations in Gdansk were used, as shown in Figures 1 and 2. The characterization of the collection places is presented in Table 1. To identify the samples, symbols GA and GK were used throughout for samples from Gdynia and Gdansk, respectively. Spruce needles were collected during summer months in triplicate from trees under study with age about 100 years at 1 m height above the ground, in amounts about 10–20 g of fresh weight. Then, the needles were air-dried and kept until analysis in plastic bags. For comparison purposes, the soil samples were also collected under the spruces, from which needles were taken for the investigation, and analysed for the same metals. In all collection sites, the samples of soils were taken up in triplicate using plastic spatula from 0–10 cm layers just under the investigated trees.

To prepare the plant materials for the determination of metals, the needles were ground in a Knifetec sample mill 1095 (Höganäs, Sweden), next digested by microwave-assisted digestion procedure (UniClever BM-1z, Plazmatronika Wrocław, Poland). The following

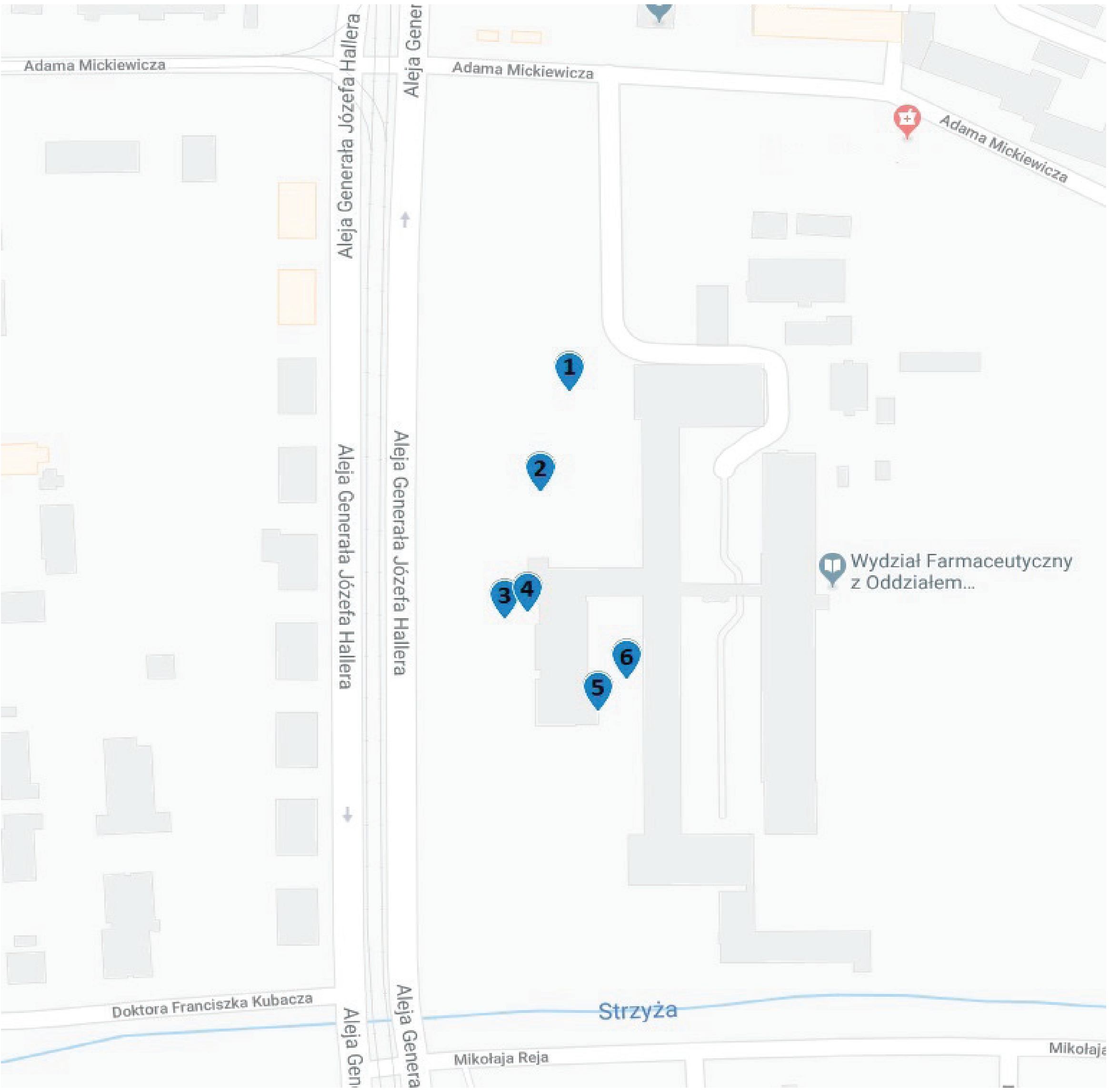

Figure 2. Map of harvest in Gdansk

parameters of this process were applied: pressure from 31 to 45 atm, temperature between 250 and 350°C, the power of microwaves set to 85% of the maximum value, time of digestion 7 min and time of cooling 5 min. After digestion, the samples were transferred into volumetric flasks and diluted up to 50 ml with redistilled water (Heraeus Quarzglas, Switzerland). To prepare the soil samples for analysis of plant-available metallic element species, extraction with the 1 mol/l solution of hydrochloric acid (pure for analysis grade reagent, the Polish Company of Chemistry, Gliwice) was applied, using 5 g of soil and 50 ml of redistilled water, mixed with electromagnetic stirrer for 1 h [Karczewska and Kabała 2008].

For the determination of concentrations of Fe, Mn, Zn, Cu, Pb and Cd in the digests obtained from the spruce needles and in the extracts from the soil samples, A250 plus atomic absorption spectrometer (Varian, Australia) in the flame mode of operation was used. Standard procedures were applied, and analytical wavelengths [nm] were as follows: Fe – 248.0; Zn – 214.0; Mn – 280.0; Cu – 325.0; Pb – 217.0; Cd – 228.8), while the used gas mixture was acetylene-air.

3. RESULTS AND DISCUSSION

3.1. Metallic elements in spruce needles and soils samples

Based upon the results of quantitative analysis of metallic elements in spruce needles, as well as in the soil samples

Table 1. Characterization of studied samples of spruce needles and soil under trees

Sample No	Sample id	Plant species	Location (geographical coordinates)	Height above sea level [m]	Collection date
1	GA 1	*Picea abies* (L.) H. Karst	N 54° 29′ 15.969″ E 18° 27′ 59.098″	135	2017
2	GA 2		N 54° 29′ 21.59″ E 18° 28′ 1.046″	143	2017
3	GA 3		N 54° 29′ 30.002″ E 18° 28′ 25.54″	141	2017
4	GA 4		N 54° 29′ 22.203″ E 18° 29′ 49.919″	173	2017
5	GA 5		N 54° 30′ 44.061″ E 18° 28′ 50.811″	147	2017
6	GA 6		N 54° 31′ 30.169″ E 18° 27′ 50.759″	90	2017
7	GA 7		N 54° 29′ 52.019″ E 18° 32′ 32.5″	50	2017
8	GK 1	*Picea abies 'concolor'*	N 54° 22′ 56.773″ E 18° 37′ 21.781″	8	2018
9	GK 2		N 54° 22′ 55.758″ E 18° 37′ 21.338″	8	2018
10	GK 3	*Picea abies 'omornica'*	N 54° 22′ 54.491″ E 18° 37′ 20.755″	8	2018
11	GK 4	*Picea abies 'nidiformis'*	N 54° 22′ 54.559″ E 18° 37′ 21.129″	8	2018
12	GK 5	*Picea abies 'concolor'*	N 54° 22′ 53.569″ E 18° 37′ 22.259″	8	2018
13	GK 6	*Picea abies 'omornica'*	N 54° 22′ 53.886″ E 18° 37′ 22.706″	8	2018

GA = Gdynia; GK = Gdansk

taken from the same locations as trees (shown in Tables 2 and 3), the order (considering the arithmetic mean from all collection sites) in which the studied elements occurred is as follows: Fe, Mn, Zn, Pb, Cu and Cd for the needles, and Fe, Mn, Cu, Zn, Pb and Cd in soils, respectively. Regarding the results in Table 2, one can see that Fe level in needles remains in the range from almost 60 mg/kg in spruce needles collected from a tree growing in location GA 5 to 644.5 mg/kg in the Medicinal Plant Garden in Gdansk (location GK2). Location GA 5 in Gdynia is placed in the middle of Tricity Landscape Park (Trójmiejski Park Krajobrazowy), so relatively far away from traffic. One-way ANOVA has revealed that the differences between Fe level in samples collected in Gdynia and Gdansk are not statistically significant. As for the Fe level in soil, it was determined in a rather narrow range of concentrations, from 21.4 to 27.4 mg/kg.

Manganese level in the studied samples of spruce needles was found from 4.7 to almost 496 mg/kg, whereas in soil samples, it was determined on similar level, from 5.0 to 8.8 mg/kg. In the case of needles, the range of concentration is large, similar to the one obtained for Fe. The lowest Mn level was determined in the sample GK3, whereas the highest in samples GA3 and GA5. Zinc concentration in spruce needles was assayed from 47.4 (sample GK3) to 111.2 mg/kg (sample GA6), while the level of this element in soil samples was from 1.1 (sample GA3) to 5.6 mg/kg (sample GK1). Characteristic is the fact that Zn level in almost all soil samples from Gdansk was significantly higher than the concentration of this metal in soils from Gdynia. The exception is the sample GA6, where Zn was found on comparable level as in samples collected in Gdansk.

Copper level in the studied samples of spruce needles was found from 1.7 (GK3) to 14.7 mg/kg (sample GA5). As for the soil samples, similarly like in Zn concentrations, Cu level was significantly higher in the samples originating from Gdansk. The range of Cu concentration was determined from 0.6 mg/kg (GA2) to 8.9 mg/kg (GK1). Literature data about the level of metallic elements in spruce or another plant material confirms the range of concentration obtained in our study, not counting Pb and Cd level [Kvacova *et al.* 2015, Ceburnis and Steiness, 2000].

Table 2. Concentration of analysed elements in spruce needles [mg/kg d.m.]. The arithmetic mean ± SD is shown (n = 3)

Element	GA 1	GA 2	GA 3	GA 4	GA 5	GA 6	GA 7	GK 1	GK 2	GK 3	GK 4	GK 5	GK 6
Fe	169.18 ± 1.02	122.71 ± 9.67	65.60 ± 2.24	65.67 ± 5.97	59.93 ± 7.68	162.86 ± 6.13	312.84 ± 3.03	197.85 ± 1.54	644.47 ± 5.83	117.65 ± 6.56	105.56 ± 0.34	272.74 ± 1.70	320.03 ± 0.66
Zn	64.55 ± 2.29	96.56 ± 3.64	81.41 ± 2.14	83.82 ± 3.62	109.15 ± 7.28	111.24 ± 2.43	64.73 ± 1.39	49.77 ± 0.55	67.37 ± 1.65	47.44 ± 0.88	64.77 ± 3.25	68.64 ± 2.41	53.04 ± 2.59
Mn	26.76 ± 0.31	260.02 ± 2.42	422.82 ± 9.62	54.82 ± 2.18	495.97 ± 7.31	98.62 ± 1.71	62.65 ± 3.41	31.45 ± 1.81	62.42 ± 5.55	4.68 ± 0.97	10.63 ± 1.65	64.50 ± 2.77	85.88 ± 9.70
Cu	7.56 ± 0.50	13.21 ± 1.07	14.46 ± 0.64	12.68 ± 1.85	14.71 ± 2.09	9.36 ± 0.25	7.62 ± 0.85	3.87 ± 0.32	12.37 ± 1.08	1.66 ± 0.25	2.28 ± 0.28	2.62 ± 0.15	2.38 ± 0.34
Pb	6.15 ± 0.72	7.37 ± 0.48	8.29 ± 0.37	8.98 ± 0.29	10.93 ± 1.33	11.14 ± 0.37	12.41 ± 0.71	18.57 ± 1.84	23.94 ± 2.61	14.51 ± 1.47	16.42 ± 0.93	18.48 ± 3.21	14.04 ± 1.63
Cd	0.37 ± 0.06	1.50 ± 0.14	1.62 ± 0.21	1.67 ± 0.20	1.60 ± 0.08	1.47 ± 0.10	1.52 ± 0.10	0.69 ± 0.09	0.73 ± 0.05	0.70 ± 0.10	0.73 ± 0.06	0.70 ± 0.01	0.76 ± 0.06

GA = Gdynia; GK = Gdansk

Table 3. Concentration of analysed elements in soil samples [mg/kg d.m.]. The arithmetic mean ± SD is shown (n = 3)

Element	GA 1	GA 2	GA 3	GA 4	GA 5	GA 6	GA 7	GK 1	GK 2	GK 3	GK 4	GK 5	GK 6
Fe	21.44 ± 0.97	24.50 ± 0.66	25.13 ± 0.63	23.24 ± 1.14	26.03 ± 0.52	26.20 ± 1.35	25.43 ± 0.70	27.40 ± 0.45	27.01 ± 0.18	26.90 ± 1.08	26.73 ± 1.05	25.60 ± 1.28	25.03 ± 1.11
Zn	1.34 ± 0.05	1.28 ± 0.05	1.08 ± 0.06	1.72 ± 0.04	1.76 ± 0.07	5.39 ± 0.09	3.44 ± 0.32	5.64 ± 0.06	5.15 ± 0.12	5.29 ± 0.14	5.18 ± 0.12	4.98 ± 0.20	5.10 ± 0.25
Mn	5.27 ± 0.18	7.11 ± 0.06	8.84 ± 0.17	5.07 ± 0.14	7.21 ± 0.13	7.87 ± 0.25	6.65 ± 0.33	7.24 ± 0.16	5.00 ± 0.23	7.41 ± 0.49	7.10 ± 0.45	7.38 ± 0.37	6.47 ± 0.36
Cu	0.65 ± 0.01	0.59 ± 0.05	0.66 ± 0.03	0.66 ± 0.04	1.07 ± 0.02	0.96 ± 0.06	0.91 ± 0.01	8.87 ± 0.20	7.85 ± 0.41	8.36 ± 0.34	7.90 ± 0.18	5.55 ± 0.13	6.85 ± 0.04
Pb	0.50 ± 0.07	0.51 ± 0.02	0.47 ± 0.07	0.45 ± 0.04	0.37 ± 0.01	0.12 ± 0.01	0.85 ± 0.04	3.62 ± 0.05	2.14 ± 0.07	2.64 ± 0.01	2.56 ± 0.02	3.19 ± 0.03	2.98 ± 0.07
Cd	0.18 ± 0.01	0.19 ± 0.01	0.21 ± 0.01	0.21 ± 0.02	0.22 ± 0.01	0.25 ± 0.01	0.23 ± 0.02	0.35 ± 0.02	0.26 ± 0.02	0.27 ± 0.01	0.25 ± 0.01	0.29 ± 0.01	0.29 ± 0.01

GA = Gdynia; GK = Gdansk

Toxic elements, Pb and Cd were also determined in the range of concentrations of mg/kg, but Pb was present in higher amounts than Cd in needles. Lead was determined from 6.2 (GA1) to almost 24 mg/kg (GK2),while Cd from 0.4 (GA1) to 1.7 mg/kg (GA4). In the case of the analysed soil samples, Pb was found from 0.12 (GA6) to 3.6 mg/kg (GK1), and Cd from 0.18 (GA1) to 0.35 mg/kg (GK1). Based on the comparison with the other studies concerning toxic elements in spruce needles, the level of Pb was determined in higher amounts in our study, since Staszewski et al. [2012] has found this metal in spruce needles at average concentration equal to 1.73 mg/kg, and this value in the samples from Gdansk and Gdynia was about 10 times higher (Table 2). Similarly, Cd level was higher in our material in comparison with the results obtained by Staszewski et al. However, the fact should be taken into consideration that Staszewski et al. have analysed the Pb level in spruce needles growing in the Polish national parks, areas relatively free from contamination sources, while our samples originated from trees growing in the centre of Gdansk, and in the forest close to Gdynia centre and the highway characterized by intensive traffic (the state way No 6 leading from Tricity to Szczecin and to Hel peninsula).

When comparing our results of Pb and Cd concentrations in spruce needles with those obtained for spruce needles growing in the Eastern Carpathian mountains [Szponar et al. 2009], it can be seen that Pb level was about 2–4 times higher in the needles of *Picea excelsa* originating from the mountains. On the other hand, the Cd level was about 2–3 times lower in the spruce samples from the mountains in comparison with our results.

Ceburnis and Steiness [2000] has also determined the metals in spruce needles originating from Lithuania, and

Table 4. Results of correlation analysis between concentrations of the studied metals in spruce needles and soil samples (element symbol with S in upper index). Coefficients of correlation higher than r = 0.7 statistically significant (α < 0.05) are shown in bold font

	Fe	Zn	Mn	Cu	Pb	Cd	Fe^S	Zn^S	Mn^S	Cu^S	Pb^S
Fe	1										
Zn	-0.34	1									
Mn	-0.36	0.64	1								
Cu	-0.10	**0.74**	**0.71**	1							
Pb	**0.71**	-0.46	-0.41	-0.42	1						
Cd	-0.39	**0.73**	0.63	**0.72**	-0.50	1					
Fe^S	0.26	-0.14	-0.02	-0.27	**0.73**	-0.07	1				
Zn^S	0.46	-0.46	-0.62	**-0.72**	**0.80**	-0.53	**0.71**	1			
Mn^S	-0.46	0.21	0.46	-0.07	-0.13	0.29	0.43	0.08	1		
Cu^S	0.41	**-0.72**	-0.52	**-0.70**	**0.83**	**-0.71**	0.66	**0.81**	-0.04	1	
Pb^S	0.36	**-0.77**	-0.51	**-0.79**	**0.77**	**-0.71**	0.53	**0.76**	0.02	**0.93**	1
Cd^S	0.33	-0.53	-0.40	-0.65	**0.76**	-0.46	**0.70**	**0.84**	0.18	**0.81**	**0.87**

when analysing the Pb results, again ours are higher. However, it can be noticed that the Cd concentrations determined in the samples from Gdansk and Gdynia are on similar level, as found in the samples of spruce needles from Slovakia [Mankovska 1998].

In general, the metallic elements in soil samples were found in lower amounts than the same elements in spruce needles. It must be emphasized that the method using 1 mol/l solution of hydrochloric acid for the metal's extraction of soils, caused that only plant-available metallic element species were extracted, not their total concentrations [Karczewska and Kabała 2008].

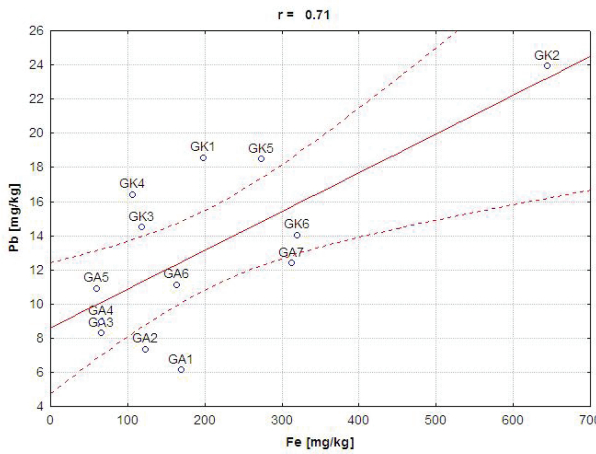

Figure 3. Correlation between Fe and Pb levels in spruce needles

3.2. Correlation analysis

The results of correlation analysis are presented in Table 4. Among the elements determined in spruce needles, depending on which level was related to each other, were the following pairs: Cu-Zn, Cu-Mn, Fe-Pb, Cd-Zn and Cd-Cu. There are even more statistically significant relations between metals occurring also in soil samples (r > 0.8), such as Cu-Zn, Pb-Cu, Cd-Zn, Cd-Cu and Cd-Pb. These relations point towards high correlation of heavy metals in the studied soils in Tricity, which is an effect of their contamination, probably caused by anthropogenic factor. The relations among the selected micro-elements, such as the correlation: Zn – Mn, were obtained previously also for medicinal plants [Konieczynski and Wesolowski, 2015]. These relations can be explained by the role of micro-elements as co-factors in numerous biochemical processes in plants [Farago, 1994]. However, it is not easy to explain why several correlations were statistically significant, as noticed for the relation between Pb contents in spruce needles and Fe in soils. Perhaps the explanation can be a

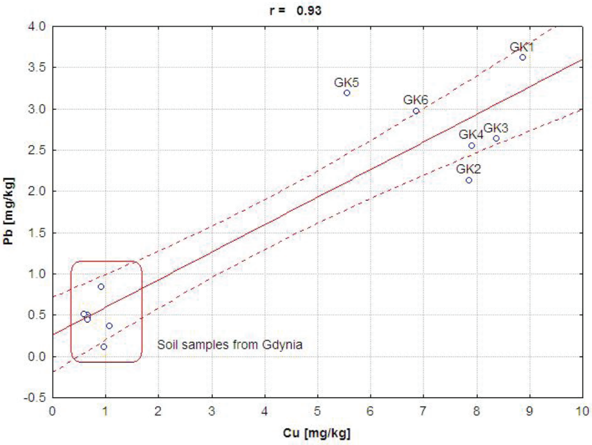

Figure 4. Correlation between Cu and Pb levels in soil samples

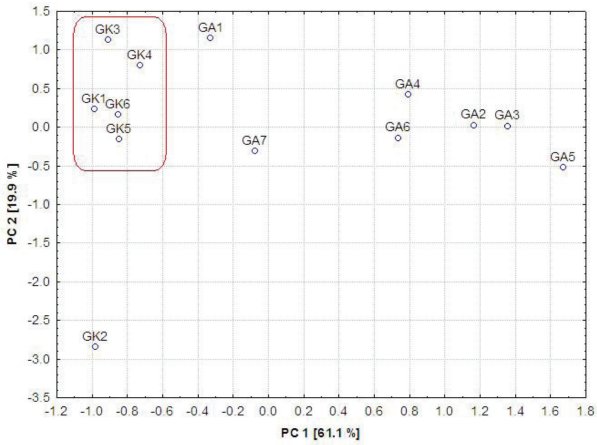

Figure 5. Principal component analysis scores plot

relatively limited number of samples under study. On the other hand, the statistically significant correlation relation between Pb in needles and the same element in soil samples (r = 0.77) has its logical justification.

Only a few significant correlations were also found between the level of the studied elements in needles and soils samples, such as negative relations in the following pairs of elements: Zn in needles – Cu in soil, or Zn in needles – Pb in soil. Positive relations were found for the pairs: Pb in needles – Fe in soil, Pb in needles – Cu in soils. Moreover, the highest correlation coefficients were calculated for the relations: Cd in soil – Pb in soils (r = 0.87), and Pb in soil – Cu in soil (r = 0.93). Some characteristic examples of relations among the studied elements obtained for spruce needles and soils, are shown in Figs 3 and 4, respectively.

3.3. Analysis of variance

Using one-way ANOVA calculations, it was found that statistically significant ($\alpha < 0.05$) differences exist between collection places in Gdynia and Gdansk for the following metallic elements determined in spruce needles: Zn, Cu, Pb and Cd. All the samples of spruce needles and soils originating from the Medicinal Plant Garden of the Faculty of Pharmacy in Gdansk were characterized by a higher level of Pb. This situation can be justified by a fact that the garden is located along one of main roads (Gen. J. Hallera Avenue) in Gdansk; so, the high concentration of lead in spruce needles and soils was caused due to the high traffic in this area for many years (Fig. 2). On the other hand, Cd level was similar in the samples originating from both cities, and didn't differ significantly. Within Gdynia, the collection places were more differentiated, because some of them, such as samples GA1 to GA5 are located in the middle of the forest of Tricity Landscape Park (Fig. 1), and due to this, the level of the studied elements, such as Pb and Zn, was significantly different in comparison with the samples from Gdansk.

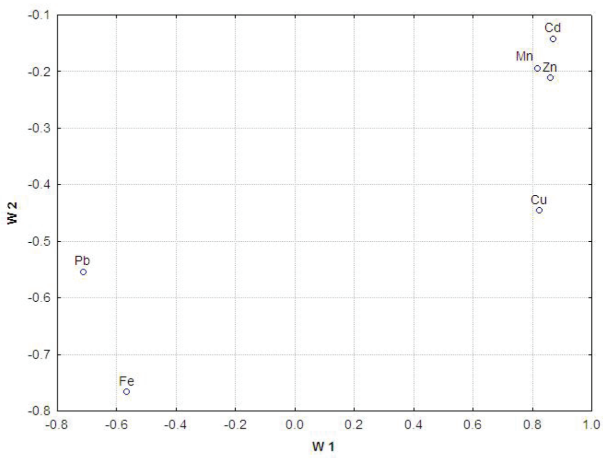

Figure 6. Principal component analysis loadings plot

3.4. Principal component analysis (PCA)

The results of the determination of Fe, Zn, Mn, Cu, Pb and Cd in the studied 13 spruce needles originating from the trees growing in Gdynia and Gdansk were used for principal component analysis calculations. The two databases were obtained: principal component scores and principal component loadings. Two first principal components accounted for 81% of variability among the investigated plant material, and the *eigen*-values for them were equal to 3.67 and 1.19, respectively. As it can be seen in Figure 5, the samples of spruce needles growing in Gdansk are located in the upper left corner of the two-dimensional plot of PC1 and PC2. Only one of them, sample GK2 is located in the left down area of the plot. This sample (GK2) contained higher amounts of Fe and Pb in comparison with others, which is the reason for its location visible in Figure 5.

In general, all the analysed samples of spruce needles originating from Gdynia can be found in the upper area of the plot, and are well separated from those collected in Gdansk. The elements, which are mainly responsible for such a separation of spruce needles samples, are concentrations of Zn, Mn, Cu and Cd, which are correlated to PC1, and Fe, related to PC2, as it can be seen in Figure 6. The effect of geographical origin on the content of metallic elements was proved in other investigations, for example, on tea samples [Polechońska et al., 2015], as well as the concerning medicinal plants [Konieczynski et al., 2018]. Thus, the fact that the place of origin of plant samples determined mostly the level of the studied elements in the spruce needles, was confirmed in this study too.

4. CONCLUSIONS

Quantitative analysis of the selected metallic elements, such as Fe, Zn, Mn, Cu, Pb and Cd in spruce needles, and in the soils originating from Gdynia and Gdansk, can be a valuable tool for the evaluation of environmental pollution of Tricity region, represented by the samples from Gdansk and Gdynia. The most important elements for studying the differences among the investigated samples appeared to be the level of Zn, Mn, Cu and Cd, as proved by PCA. It was also found that Pb in spruce needles was significantly higher in the samples from Gdansk. Moreover, several significant correlations were found between the level of the studied elements in spruce needles and soils samples, such as the positive relation between the Pb concentration in the needles and the level of this element in soil samples. Moreover, numerous correlations were found between heavy metals in soils, such as Cu-Zn, Pb-Cu, Cd-Zn, Cd-Cu and Cd-Pb, which indicate pollution caused probably by anthropogenic factor.

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