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Analysis of S, Cl, K, Ca, Cr, Mn, Fe, Zn, Rb, Sr, Mo, Ba and Pb concentrations in the needles of *Abies alba* and potential impact of paper mill industry

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Abstract. In our work we studied samples of *Abies alba* Mill., - specifically the needles – as a bio indicator of pollution in Ružomberok (not far from the paper mill) and Liptovský Hrádok, Slovakia - the West Carpathians. Samples were collected from two locations, Choč (experimental area) and Čierny Váh (control area). At each locality we selected five field sites, from which we collected samples of needles once a month from May 2016 to February 2017. Various differences in the concentration of elements were detected in the samples based on sample age and the time of year samples were collected. We discovered that concentrations of potassium decreased as the needles aged, while calcium increased. Concentrations of toxic elements increased during autumn and winter and decreased during summer months. Ca, Fe, Rb, Sr, Cr and Pb, were more prevalent in the sample location at Choč (not far from the paper mill) in comparison to the control location.

Key words: Abies alba, heavy metals, bioindicators, pollutants

Introduction

Toxic substances are released into the environment through industrial processes, and increasing road transport results in air pollution by heavy metals. Motor-vehicle pollutants include toxic metals, such as lead and cadmium (Kmeť 1996). *Abies alba* Mill., from the *Pinacea* family belongs to the most important softwood species and is highly responsive to any change in the atmosphere. Using plant tissues for sample collection is a well-established and effective indicator of air pollution (Goodman and Roberts 1971).

Toxic substances enter plants mostly via a soil reaction. Accumulation of these substances is highly dependent on the content and quality of organic substances, plant nutrition, and cation exchange absorption capacity. Concentrations of elements in the soil depend on the soil pH value. Elements such as Cd, Cr, Pb, Zn and Ni have maximum mobility in the soil at pH levels below 5.5. In addition, the mineral partition of the soil plays an important role in metal immobilisation (Barančíková 1998). Plants have the potential to tolerate a large amount of metal concentration, known as hyper-accumulation. Hyperaccumulators are defined as higher plant species whose shoots contain > 100 mg Cd kg^{-1}, > 1,000 mg Ni, Pb and Cu kg^{-1}, or 10,000 mg Zn and Mn kg^{-1}, when grown in metal-rich soils (Baker *et al.* 1994).

Lead is accumulated in the plants as an aerosol from the atmosphere. It is caught on the surface of the plants due to atmospheric deposition. The highest content of lead is found on the leaves, followed by the stalks, and the smallest content is found in the plant tissues (Svičeková and Havránek 1993). Lead is found only in the upper humus layer to a depth of 50 mm. In the deeper tissues lead content decreases (Beneš and Pabiánová 1987).

Plants receive sulphur from mineral compounds in the form of the highest oxide; the SO_4 anion. (Kmeť 1996). A higher concentration of sulphur in plants occurs due to sulphate pollutants, such as NO_2 and H_2S. Two main reasons for sulphur toxicity are known: root damage due to higher sulphur concentrations in the humus complex; and damage to leaves caused by sulphur metabolites as a result of the excessive uptake of SO_2 and H_2S. Sulphur is accumulated mainly in older tissues, and high sulphur concentrations result in a decrease in plant growth (Innes 1995).

Coniferous trees are a significant bio indicator of environmental pollution as their needles have a long lifespan. Bio indicators are useful for defining threshold values for sulphur, chlorine, fluorine and heavy metals (Arndt *et al.* 1987). A wide range of biochemical methods are available for bio indication purposes, such as pigment concentrations, buffer capacity and PEP-carboxylase activity (review by Wild and Schmitt 1995). These methods are developed mainly when measuring and assessing chloroplast pigment, especially chlorophyll (Lichtenthaler 1993). Diminution of forests is closely related to chlorophyll-degrading needle chlorosis. Biochemical damage to chlorophyll has already occurred before visible symptoms emerge, such as yellowing of needles, or the subsequent loss of needles or decrease in the height profile (Oren *et al.* 1993).

The aim of this study is to show the degree of air pollution in selected locations, namely the polluted area of Choč, in the region of Ružomberok and the unpolluted area of Čierny Váh, in the Liptovský Mikuláš region. Further, we want this study to highlight the movement of toxic substances in needles as determined by factors such as the age of needles, locality, and seasonality.

Material and Methods

Samples were collected from two different remote locations. The first location was the hill of Choč in the region of Ružomberok located at an altitude of 1,611 m. The second control location was Čierny Váh in the region of Liptovský Mikuláš at an altitude of 1,160 m. We created 5 principal sample locations, from which we collected needle samples once a month throughout the year (May 2016 – February 2017). At each site we collected one twig, which was subsequently divided into one, two, and three-year-old needles. The collected samples were stored in polythene bags and transported to the laboratory.

In the laboratory, we placed the divided twigs on a Petri dish and then into the Memmert IF 160 Plus biological thermostat. Samples were dried at 75° C for 12 hours using natural circulation of air. Dried *Abies alba* needles were homogenized to a fine powder in a Retsch CroMill cryogenic ball mill. The CroMill was designed for grinding using nitrogen. Samples were ground without the use of an integrated cooling system and we used one of three available modes – the dry mode. Grinding jars produce radial oscillations in a horizontal position and the movement of grinding balls inside caused a high energy impact on the sample in the rounded ends of grinding jars. The grinding container holds 20 ml of material (Korčeková 2015). Samples were measured using X-ray fluorescence. Samples (1.5 g - 2 g) were placed into a special cuvette and put into the DELTA CLASSIC XRF spectrometer (US), which is able to measure elements.

Data was processed using the STATISTICA 12.1. statistical program. Element measurements were analysed using principal component analysis (PCA), with the determination of cross-correlation based on factor scores. For the analysis of differences between groups of parameters one-way or two-way analysis of variance – ANOVA – was used. Values of $p<0.05$ were considered as statistically significant (Table 1).

Variable	Factor 1	Factor 2	Factor 3	Factor 4	Factor 5	Factor 6	Factor 7	Factor 8
S	**-0.85**	0.06	-0.18	0.02	0.17	0.02	-0.002	0.03
Cl	-0.61	0.07	-0.18	0.22	**0.37**	**-0.60**	-0.08	0.02
K	-0.60	-0.33	-0.12	0.39	**-0.47**	-0.07	-0.14	-0.20
Ca	-0.62	**0.58**	-0.004	0.15	0.32	0.07	0.17	0.02
Cr	**-0.87**	-0.18	0.11	-0.26	-0.13	-0.13	-0.09	-0.06
Mn	-0.41	0.44	**-0.66**	-0.03	-0.22	0.19	0.15	0.03
Fe	**-0.72**	-0.17	0.02	-0.39	0.01	0.12	**-0.38**	0.14
Zn	-0.57	0.14	0.33	0.36	**0.34**	**0.40**	-0.23	-0.14
Rb	-0.32	**-0.60**	-0.41	**0.47**	0.01	0.16	0.13	0.03
Sr	-0.06	0.43	0.48	**0.52**	**-0.45**	-0.10	-0.04	0.22
Mo	-0.62	-0.26	0.49	-0.13	0.04	-0.02	**0.41**	-0.27
Ba	-0.66	0.43	-0.04	-0.30	**-0.37**	-0.02	0.01	-0.18
Pb	**-0.73**	-0.28	0.18	-0.11	-0.06	0.07	0.21	**0.47**
Variance (%)	38.93	12.27	10.02	9.20	7.85	4.93	3.95	3.51

Table 1. Principal component weights of the first eight factors of principal component analysis of data on element concentrations in the fir leaves.

Results

Tin and Antimony

At Choč hill tin was detected in 42 of 150 samples and antimony in 88 of 150 samples. At the control locality, Čierny Váh, tin was found in 49/150 and antimony in 93/150 samples. Occurrence of tin or antimony likely does not depend on the sample location (Sn-Chí^2 = 0.99, p = 0.31, Sb - Chí^2 = 0.06, p = 0.8).

Elements found in all samples

Sulphur

Detected sulphur measurements in various chemical forms proved that its occurrence in leaves was dependent neither on the variability of locations, nor on the needle age (Table 2). However, sulphur occurrence was definitely season-dependent with the lowest values occurring in the late summer and autumn periods (Fig. 1). Taking into account the synergistic effect of Cl, K, Ca, Cr, Mo, Ba, and Pb, sulphur presumably is present in higher concentrations in the form of various compounds mainly in winter, but also occurs significantly in spring (Fig. 2).

Chlorine

Chlorine occurs especially in winter in synergy with the other elements shown in Fig. 2. Overall analysis showed that the amount of chlorine was the same in the needles of different age classes, but its levels in Choč were significantly higher than in Čierny Váh (Fig. 3). The amount of chlorine in the needles increased during winter in each location, and by needle

Age - Years NS	Weighted average (μg)	Standard error	N
1	636.78	21.79	100
2	634.06	17.99	100
3	654.81	21.56	100
Months			
5	648.07	35.36	30
6	620.50	31.86	30
7	603.43	30.85	30
8	531.70	16.41	30
9	534.17	25.76	30
10	437.17	13.33	30
11	604.47	30.48	30
12	760.50	25.84	30
1	864.70	40.56	30
2	814.13	35.16	30
Localities NS			
Choč	632.89	16.60	150
Čierny Váh	650.88	16.86	150

Table 2. Sulphur and age classes of needles, months (F=19.9, p=0.00) and localities NS (F=0.18, p=0.9).

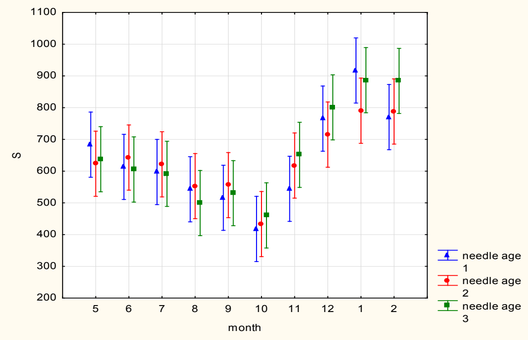

Fig. 1 Mean values of S in different age classes of needles (vertical bars denote 0.95 confidence limits) in dependence on seasons. Two way ANOVA effects: F (month)=19.9, p=0.00, F (age): =0.47, p=0.6). Amount of sulphur increases in all age classes in winter.

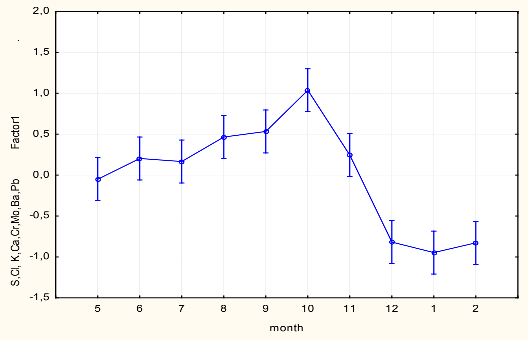

Fig. 2. Factor of general pollution in seasons. The leaves of fir suffer from polution mainly in winter, effects of pollution decrease in late summer and mainly in autumn. Numbers (Y-axis) represent means of factor scores, vertical bars denote 0.95 confidence limits, the lower value of a mean score, a higher pollution by the elements (month F=21.2, p=0.00). The pollution was higher in the Choč than control Čierny Váh area (F=0.18, p=0.00), and contamination of leaves did not different among their age classes (age F=0.8, p=0.4).

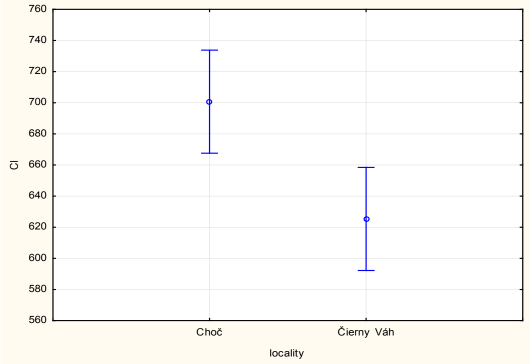

Fig. 3. At locality Choč witch, is situated closely to paper mill industry, the average values of Cl in needles were significantly higher than at the control locality – Čierny Váh.

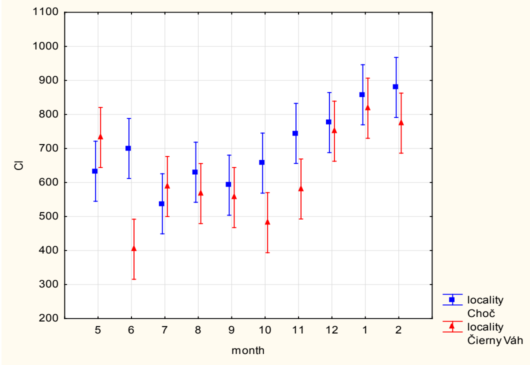

Fig. 4. The amount of chlorine significantly decreased in summer until autumn months.(F=12.29, p=0.00). In winter concentration of Cl increases and relatively high level of retained spring in May. Chlorine concentrations depend on the locations. In the picture is seen that at Choč were higher values of chlorine than in Čierny Váh.

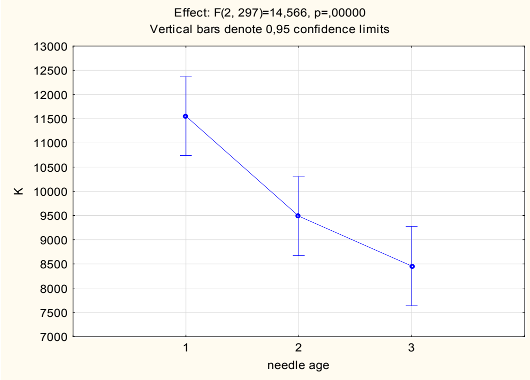

Fig. 5. The amount of K (in ppm) significantly decreased with age of needles.

age, independently (Fig. 4). Chlorine probably inhibits the metabolism of potassium and strontium (Fig. 5).

Potassium

As mentioned above, under particular conditions, potassium participates in the synergistic occurrence of harmful substances in the environment, mainly in winter (Fig. 2). Its concentration decreases in deteriorated conditions where chlorine is present (Fig. 5). Overall,

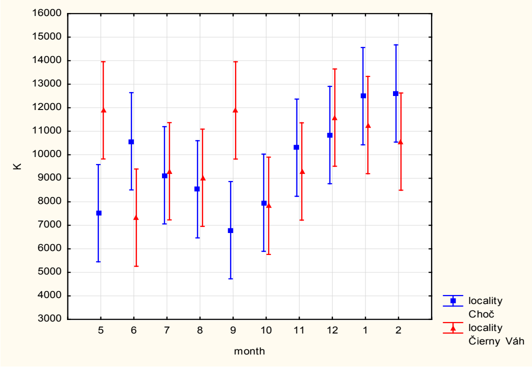

Fig. 6. There was not difference in the level of K between two locations (F=0.5, p=0.5) but K significantly increased in winter months (F=3.1, p=0.001) at both localities. The lowest levels of K were found at the end of summer and in the autumn. Because the number of needles of different age classes was equal in each month group, the age difference in the amount of K does not connect with K variability in seasons.

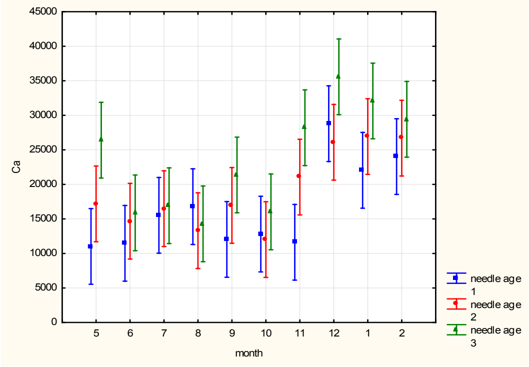

Fig. 7. The amount of Ca increases in winter is all age classes of needles (F=14.1, p=0.000). Older needles contain more Ca than younger needles (F=16.3, p=0.000). In both localities the amount of Ca increases in winter period but at Choč the level of Ca is always higher than in Čierny Váh. (F=14,14, p=0.001).

in its various chemical forms, potassium concentration decreases as the needles age (Fig. 6). Potassium concentration increases in winter and spring independent of the age of needles or sample location (Fig.7).

Calcium

In general, the calcium concentration trend in needles is the opposite to that of potassium. Older needles contain more calcium than the younger ones and the amount of calcium significantly increases with the age of the needles (Fig. 8). Calcium levels were higher in the location of Choč specifically during winter months. These concentrations of calcium are presumably related to the complex compounds involved in the seasonal cycle of harmful substances (Fig. 2). There is a synergistic effect with chlorine and sulphur as well as with other elements. The second principal component denotes the mutual interrelationship between calcium and rubidium contamination. When calcium increases, rubidium decreases and vice versa. The proportion of Rb to Ca increased at Choč (Fig. 9) in winter (Fig. 10).

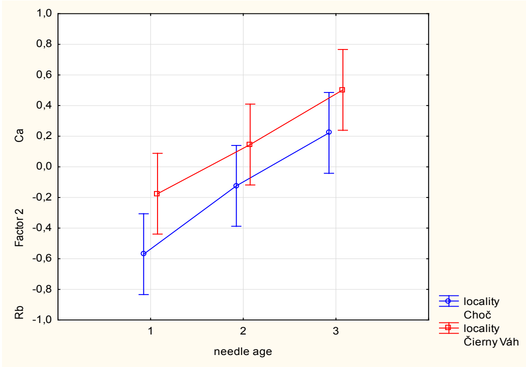

Fig. 8 Principal component 2 denotes the mutual interrelationship between Ca and Rb contamination. When Ca increases Rb decrease and vice versa. Locality Choč is proportionally more polluted by Rb than Čierny Váh (F=8.28, p=0.004), and this pollution increases with age of needles at both locations (F=15.0, p=0.000).

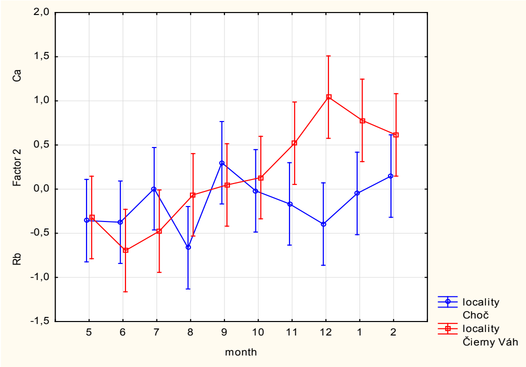

Fig. 9. Proportional increase of Rb and decrease of Ca is mainly in winter at location Choč (F=4.36, p=0.000).

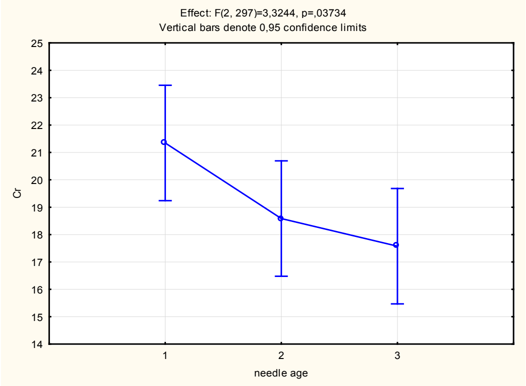

Fig. 10. The amount of Cr (in ppm) significantly decreased with age of needles.

Chromium

Under the synergistic effect of pollutants, chromium also occurs in increased concentrations during winter months (Fig. 2). Similar to potassium, the amount of chromium in the needles significantly decreases with the age of needles (Fig. 11) and the overall chromium concentration in the leaves increases in spring and in winter in a location-independent manner (Fig. 12).

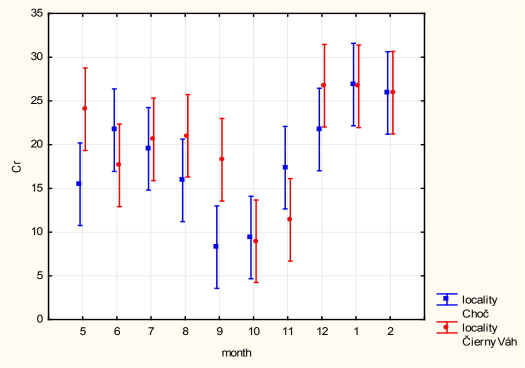

Fig. 11. The amount of Cr in the fir needles varies by the same way as amount of K. The highest levels of Cr are in young leaves (Fig. 1, Cr), and the lowest values of Cr may be found in the leaves from the autumn (F=11.2, p=000). The amount of Cr was not significantly different between two studied localities.

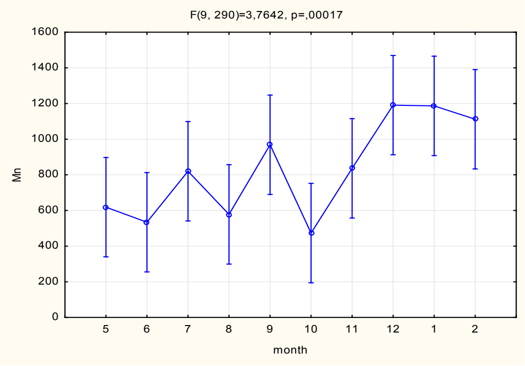

Fig. 12. Mn significantly increased in winter months, the amount of Mn in the leaves did not differ between two localities (F=0.69, p=0.4), nor among the age classes (Table 5).

Needle age in years	Weighted average - Mn	Standard error	N
1	828.69	81.92	100
2	868.19	82.36	100
3	798.93	78.46	100

Table 3. Age classes of needles and the average values of concentrations of manganese in them. The difference was not statistically insignificant.

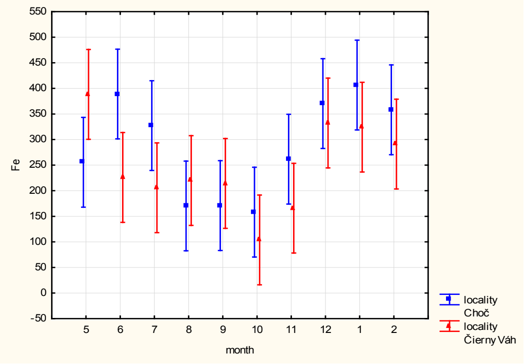

Fig. 13. Seasonal differences in the amount of Fe were comparable to the variation known from Ca or Cr the lowest values of Fe where found in the autumn leaves (F=6.4, p=0.000) the amount of Fe was higher at Choč hill than at the locality Čierny Váh – mainly in winter(F=2.03, p=0.036).

Manganese

The amount of manganese does not differ between different age classes of needles (Table 3), however, the manganese concentration does significantly increases in winter months (Fig. 13).

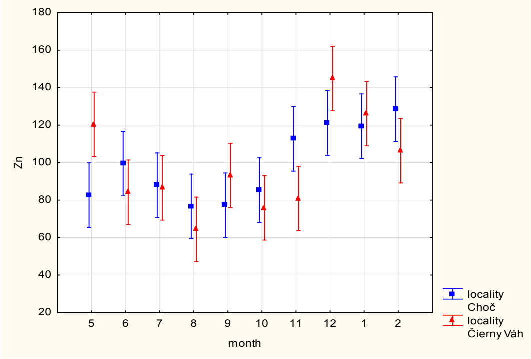

Fig. 14. The lowest values of Zn were found in autumn – September, October (F=10.49, p=0.000). The seasonal trend of Zn accumulation was equal in both studied locations but there was not difference in the amount of zinc between locations. The variability of Zn is comparable to the seasonal variation of K , Cr and Mn.

Needle age	Weighted average	Standard error	N
1	297.68	20.82	100
2	259.20	16.39	100
3	244.12	19.06	100

Table 4. Age classes of needles and the average values of iron in them in ppm.

Iron

The amount of iron was higher at the Choč hill location than at Čierny Váh – mainly in spring and in winter (Fig. 14). No needle age-dependent differences were found (Table 4).

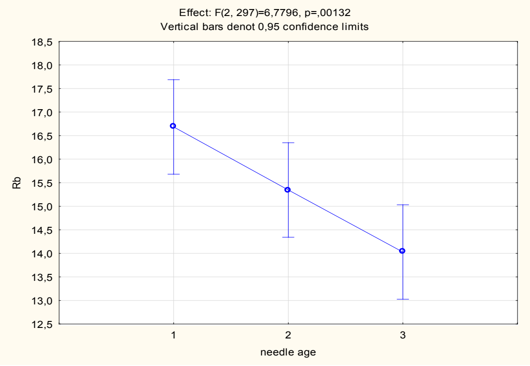

Fig. 15. The amount of Rb (in ppm) significantly decreased with age of needles.

Needle age	Weighted average	Standard error	N
1	94.01	3.13	100
2	97.16	3.85	100
3	104.91	4.61	100

Table 5. Age classes of needles and the average concentrations of Zn in them (ppm). Non-significant differences between the groups.

Needle age	Weighted average	Standard error	N
1	25.60	3.45	100
2	29.41	3.73	100
3	33.35	4.25	100

Table 6. Age classes of needles and the amount of strontium in them, there was not significant difference in average values among the groups.

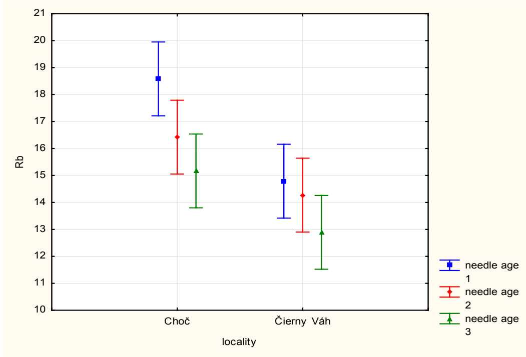

Fig. 16. Levels of Rb were higher at Choč than in Čierny Váh (F=23.3, p=0.000). In both locations the level of Rb was higher in younger than older leaves (F=7.3, p=0.001). Concentration of Rb in the leaves of fir is not seasonally dependent (F=0.82, p=0.6).

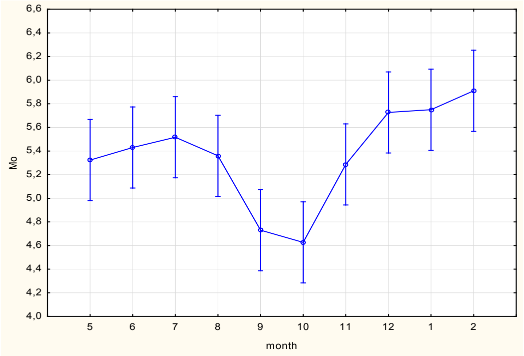

Fig. 17. Mo in needles has very similar seasonal variation to variation of K, Cr, Zn, Fe. The amount of Mo depends on seasons, the lowest concentrations were found in autumn (F=5.7, P=0.000), and does not depend on locations. (F=0,74, p=0,4). The concentration of Mo does not differ among different age classes of needles (Table 8).

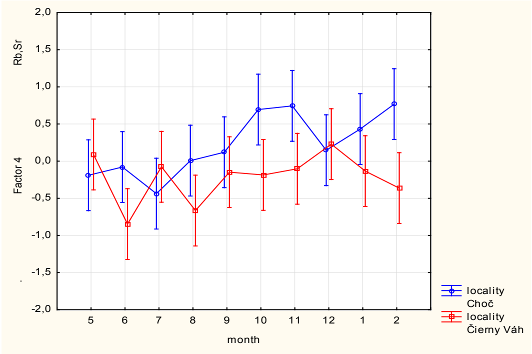

Fig. 18. Principal component 4 denotes the mutual contamination of needles by Rb and SR. It increases in autumn and winter months (F=2.2, p=0.02), mainly at Choč hill (F=15.15, p=0.000). The contamination does not differ among different age classes of leaves (F=0.1, p=0.9).

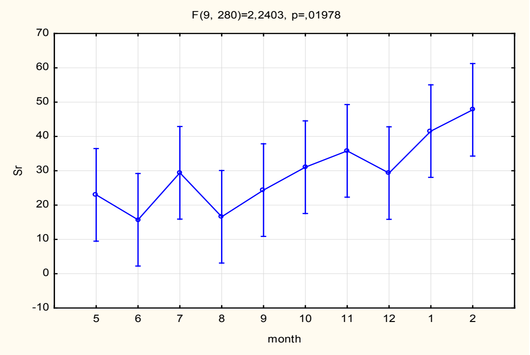

Fig. 19. There was not difference in the concentration of Sr in the leaves between two localities (F=0.05, p=0.08). Strontium increases in the winter (F=2.2, p=0.02), the variation is also known in Ca and Mn.

Zinc

Zinc does not appear as a pollutant (Fig. 2), and its amount does not differ by the age of needles (Table 5). The amount of zinc was higher in spring and in winter, but there was not any difference in the amount of zinc between locations (Fig. 15).

Rubidium

The hazard represented by rubidium results from the fact that it presumably displaces calcium from the needles. This was mainly seen to occur at the Choč location during the winter season. (Figs. 9–10). The amount of rubidium significantly decreased with the age of needles. Concentration of rubidium in the leaves of fir is not seasonally dependent. Levels of rubidium were higher at Choč (Fig. 16). An inverse mutual rubidium and strontium contamination has been observed in needles. The ratio of Rb/Sr increased in autumn and winter months, mainly at Choč sample location (Fig. 17).

Strontium

Under certain conditions, mainly in the deteriorated conditions of Choč, the synergistic effect of strontium and rubidium was observed, mainly in winter (Fig. 17). The strontium concentration increased in winter (Fig. 18), but did not vary among different age classes of needles (Table 6).

Needle age	**Weighted average**	**Standard error**	**N**
1	5.42	0.10	100
2	5.39	0.10	100
3	5.29	0.11	100

Table 7. Age classes of needles and the amount of molybdenum in them, there was not significant difference in average values of element concentrations among the groups.

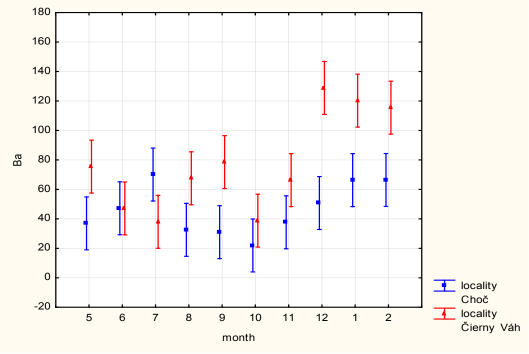

Fig. 20. Amounts of Ba were higher in Čierny Váh than in Choč (F=59.9, p=0.000), and higher in winter than in summer (F=11.2, p=0.000). The concentration of Ba does not reflect the age of needles (Table 9).

Needle age	**Weighted average**	**Standard error**	**N**
1	65.14	5.18	100
2	61.45	3.97	100
3	58.96	4.39	100

Table 8. Age classes of needles and average values of Ba in them. The differences among means were not statistically significant.

Molybdenum

This element interacts synergistically with other harmful elements mainly in spring and in winter (Fig. 2). The concentration of molybdenum did not differ among different age classes of needles (Table 7), and the lowest concentrations were found in autumn (Fig. 19).

Barium

Due to the synergistic effect, it behaves the same way as molybdenum (Fig. 2). The concentration of barium was higher at control locality than at Choč study area, specifically in winter (Fig. 20). The concentration of barium does not reflect the age of needles (Table 8).

Lead

Lead, together with sulphur, chlorine and other elements has an unfavourable environmental impact, mainly in winter. The concentration of lead did not differ among different age classes of needles (Table 9). It increases in spring and most drastically in winter (Fig. 21). The amount of lead in the needles was not different between the two locations. Under certain conditions, lead can partially counteract the content of potassium and molybdenum in needles (Table 1, Factor 8). The intensity of this effect depends on the level of deterioration of the sample site, as has been observed in Choč (Fig. 22). This type of lead concentration is likely not seasonally dependent.

Needle age	**Weighted average**	**Standard error**	**N**
1	12.01	0.30	100
2	11.55	0.27	100
3	11.34	0.27	100

Table 9. Age classes of needles and mean values of lead concentrations in them. There was not significant difference among the groups in the amount of Pb in the needles.

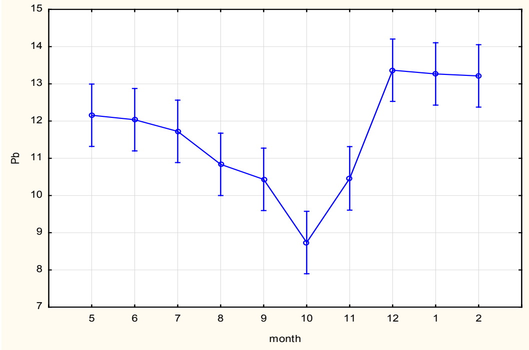

Fig. 21. Concentration of Pb highly depends on seasons beeing very low in autumn (F=12.51, p=0.000), Amount of lead in the needles was not different between two locations (F= 0.24, p= 0.7), and did not depend on age of needles (Table 9).

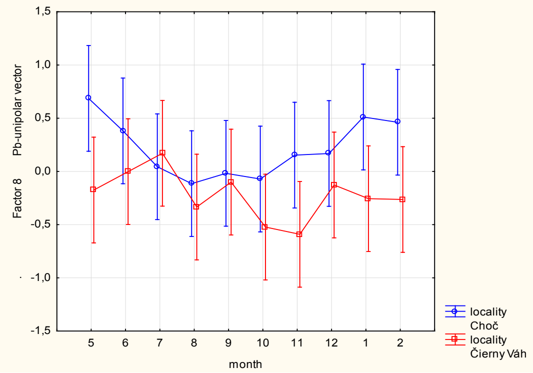

Fig. 22. Principal component 8 denotes the seasonal effects in lead contamination. The amount of this type of lead is significantly higher at Choč Hills than at the control location Čierny Váh (F=15.24, p= 0.000), mainly in the winter period. This type of lead concentration is probably not so intensively seasonally affected (F=1.15, p=0.327), and also does not differ among age classes of fir needles (F=0.664, p=0,516).

Discussion

In our work we have analysed the movement of toxic elements within the polluted location of Choč in the region of Ružomberok. The non-polluted location of Čierny Váh in the region of Liptovský Mikuláš was chosen as a control location.

We predicted that the contamination of *Abies alba* Mill. needles by toxic agents would not increase. As a conifer, the needles of *Abies alba* Mill. in particular, are sensitive to air pollution changes, and were used as a bio indicator.

In general we can say that winter and spring have a significant impact on the increased concentration of detected elements in the needles. Elements such as Ca, Fe, Rb, Sr, Cr and Pb, occur in higher quantity in the location of Choč compared to the control location, and we can infer that these concentrations are a result of the significant impact of the paper industry located nearby. Concentrations of some elements in the needles, namely calcium and potassium, vary based on age of the needles. This highlights their role in biogenic processes. Heavy metals in the tissue disrupt metabolic paths of biogenic elements, such as Ca and K. This could explain the high frequency of calcium oxalate crystals in the Choč location. The massive presence of these crystals can indicate uncontrolled calcium ingression to cells (Gostin 2010). Such results were also reported in the Ceahlău National Park in Romania where calcium concentrations increased with age. Similar research was conducted in Šumava National park (Novotný *et al.* 2010).

Potassium content was higher in one-year needles than in two- and three-year needles. The control of the plant's water regime is the most important function of potassium (Novotný *et al.* 2010). It is crucial in opening pores and participates in the creation of polymers, such as starch and protein (Kmeť 1996).

Pollutants can accumulate in plants through the root system or due to transpiration. Because of the deep stake root system of *Abies alba* Mill, we can assume that these toxic agents are accumulated by the needles through aerosols, as the elements mentioned above are usually accumulated only in the surface layer of soil. It is reported that while Cd, Cu, Co, Fe and Mo are accumulated mainly in plant roots, Pb, Sn, V, Ag and Cr are well-transported to aboveground plant parts. Zn, Mn and Ni are well-distributed to the entire plant and accumulated both in the roots and in the aboveground plant parts (Fargašová 2009).

With regard to metal distribution to the plants, consideration should be given not only to their accumulation in particular parts, but also to their distribution, which can depend on transpiration vapours (Fargašová 2009). This is caused by the negative water potential gradient between the transpiring surface and the adjacent air layer. Evaporation occurs inside the leaf, and water vapour released to the external atmosphere is physiologically controlled by the plant (Kmeť 1996).

Sulphur is present in the plants in the form of SO_4 oxide. Sulphur is metabolised early in the roots or transformed into leaves. Acting in synergy with Cl, K, Ca, Cr, Mo, Ba, and Pb, sulphur occurs in the leaves, in various compounds, both in winter and spring in a significant manner. This means that in spring, water flows intensively though conducting tissues and swells up the cytoplasm in awakening buds. The increased sulphur concentration causes damage to leaves. Sulphur is mainly accumulated in older tissue (Belicová 2010). The major producer of SO_x is the paper industry located close to our sample collection location of Choč.

Zinc is a trace element present in all live organisms. Zinc concentrations ranged from 10 to 100 ppm. It plays a major role in chlorophyll generation and participates in many metabolic processes. It also ensures the synthesis of enzymes. Zinc content correlates with magnesium. Excess zinc can result in decreased concentration of manganese and copper. Conversely, lack of phosphate may also cause reduced zinc in the plants. The lack of zinc in the plants leads to delayed cell growth (Raven 1986). In our measurements, we found that zinc concentrations were higher during autumn months (September and October). The variability of zinc correlates to seasonal changes, similar to K, Cr and Mn.

The hazard represented by rubidium results from the fact that it appears to displace calcium from the needles, specifically in the Choč location during the winter season. The rubidium concentration in the needles is not seasonally-dependent. In nature, rubidium is mainly found in earth's shell and has no biological role in metabolism. Rubidium has chemical and physical properties similar to potassium, and metabolises faster. A plant with a potassium deficiency will produce rubidium, allowing rubidium to enter the food chain (www.lenntech.com 2017).

The increased concentration of barium was observed to a greater extent during winter than in summer, in the control location of Čierny Váh. This pollution is likely a result of the distant industrial plants in the towns of Liptovský Hrádok and Liptovský Mikuláš. High concentrations result in the decrease of pore development, closure of pores and decrease in photosynthetic activity (Kmeť 1996).

Strontium has chemical and physical properties similar to calcium. It is found in small quantities in the majority of tissues. It has been found that it participates in their growth and development. Strontium is massively accumulated in the plants through the root system (Kmeť 1996). In our study we have shown that strontium and rubidium inter-contaminate needles.

Chromium was the next measured element. The amount of chromium increases in spring and in winter irrespective of location. The highest content of chromium was found in one-year needles. Chromium is a toxic and non-essential element for plants. The increased chromium concentration leads to chlorosis manifestations on the leaves and restricts normal root growth (Shanker *et al.* 2005). High chromium concentration in the atmosphere occurs due to the chromate of lead ($PbCrO_4$) released to the environment from industrial activities and the automobile industry (Al-Shayeb *et al.* 1995).

In both locations, we observed increased lead concentration. In the location of Choč, the measured concentrations were significantly higher than in the control location of Čierny Váh. This pollution could be a result of the Mondi SCP activities, as well as from road transport. Lead is accumulated in the plants mainly in the form of aerosols from the atmosphere. It is caught on the surface of the plants due to atmospheric deposition. The highest content of lead was found on the leaves, followed by stalks, and the lowest content was found in plant tissues (Svičeková and Havránek 1993). Lead is found only in the upper humus layer up to a depth of 50 mm. In the deeper tissues, lead content decreases (Beneš and Pabiánová 1987).

Elements such as iron, molybdenum and manganese should only be briefly mentioned, as no major differences were observed with regard to location, needle age, or season.

Concentration of iron was higher in the location of Choč than in the control location of Čierny Váh. The lowest iron values were measured in the autumn. Iron is an element that controls physiological functions of enzymes. It is mostly accumulated in chloroplasts, (up to 90%), and participates in the creation of the photosynthetic apparatus. A deficiency of iron leads to decreased content of chlorophyll. Needles turn yellow when affected by iron deficiency due to calcium chlorosis (Kmeť 1996).

Molybdenum values showed no difference among various age classes of needles. Differences in concentration were observed mainly in autumn months. Molybdenum plays an important role in the reduction of molecular nitrogen by nodule bacteria, and it is a part of nitrate reductase (Kmeť 1996).

Increased chlorine concentration measured on the south slopes of the Choč hill shows the negative impact of the emissions from the Mondi SCP factory. According to Xu *et al.* (2000), the higher concentration of chlorine in the leaves is partially caused as it travels from the roots to the leaves of the plants.

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