Report of STSM under COST Action FP1301: EuroCoppice

"PHYTOREMEDIATION POTENTIAL OF DIFFERENT POPLAR CLONE COPPICE PLANTATION"

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INTRODUCTION

The accumulation of metals in different environmental compartments poses a risk to both the environment and biota health, including humans, since these elements bioaccumulate in living organisms and suffer biomagnification processes. Plants can be used for phytoremediation via different physiological processes that allow metal tolerance and absorption capacity. Many types of soil clean-up techniques have been developed over the years, categorized by their physical, chemical and biological approaches. Phytoremediation takes advantage of the unique and selective uptake capabilities of plant root systems, and applies these natural processes alongside the translocation, bioaccumulation, and contaminant degradation abilities of the entire plant. One of main phytoremediation techniques is phytoextraction that uses the plants ability to extract and accumulate metals into their harvestable tissues. The efficiency of phytoextraction depends on many factors, such as metal bioavailability, soil properties, metal speciation and plant species and, mainly, on shoot metal concentration and biomass.

The main tree species used in short rotation coppicing (SRC) in Latvia now are the fast-growing hybrid aspens and willows. Poplars have great potential to become widely used as SRC culture in Latvia, as the growing conditions here are suitable, however their potential has not been fully investigated yet. Phytoremediation is being used in Latvia and its use tends to broaden with every year, when plants with phytoremediation properties are introduced in plantations and city landscapes with the goal to reduce pollutants in the environment.

PURPOSE

The aim of the STSM was to use laser ablation technique for evaluation of phytoremediation potential of coppice plantations of different Poplar clones. The STSM was carried out at the Masaryk University in Brno, Czech Republic from 16th to 22nd of October. During the STSM, the aim was met and results obtained to evaluate the phytoremediation potential of hybrid poplar SRC plantation and that of various clones of hybrid poplar.

MATERIALS AND METHODS

Preparation of samples. Altogether 10 poplar tree samples were analyzed during the STSM. The samples were collected from an experimental coppice plantation in the central part of Latvia (*Fig.1*). The experimental plot was established on agricultural land in the spring of 2011.

Fig.1. **Location of the experimental poplar plantation (N 56° 41.4950, E 025° 08.2624)**

Ten trees of various hybrid poplar clones (*Table 1*) were selected and sampled at the height of 0.20m. Each sample represented 6 tree rings with thickness of about 1 cm. The tree discs were dried at temperature of 70 °C and divided into smaller sections with diameter no greater than 4 cm due to sample chamber size restrictions of the laser ablation system.

Table 1

Characteristics of the samples

Sample ID	Clone	Tree height, m	Naturally wet stem biomass, kg	Naturally wet branches biomass, kg	$BHD_{1.3}$, mm
P13	AF2	6,12	0,84	4,82	46
P14	AF6	6,90	1,10	6,60	50
P15	AF8	5,70	0,64	4,00	40
P16	AF7	5,54	0,84	3,76	38
P17	AF8	5,20	0,98	4,08	63
P18	AF7	5,29	1,87	4,42	45
P19	AF2	4,64	1,2	4,14	46

P20	Auce 1	4,60	1,46	2,62	48
P21	LV14	8,22	2,06	7,36	52
P22	LV3	7,70	1,52	11,36	64

The clones AF2, AF6, AF7 and AF8 have been produced by an Italian company "Alasia New Clones" and the clones LV3, LV14 and Auce1 have been created in Latvia. Weed management for the plantation was performed once per season, and no additional fertilization of the soil has been performed.

After drying, the surface of the samples was treated with fine sandpaper to reduce the roughness of surface and increase the ability to obtain even portions of ablated matter throughout the analysis process.

Instrumentation. All samples were analyzed by laser ablation with mass spectrometry of inductively coupled plasma (LA-ICP-MS). The instrumentation consists of laser ablation system UP213 (NewWave, USA) that generates aerosol of sample from the sample surface and ICP-MS Agilent 7500ce (Agilent Technologies, Japan), used for isotope detection. Ablation parameters were optimized with respect to achieve best limit of detection and lateral resolution. Optimized parameters were applied for analysis of all measured samples and are summarized in Table 2. Altogether 15 isotopes were selected for determination in all samples: ^{13}C, ^{26}Mg, ^{27}Al, ^{31}P, ^{39}K, ^{44}Ca, ^{53}Cr, ^{55}Mn, ^{56}Fe, ^{60}Ni, ^{63}Cu, ^{66}Zn, ^{111}Cd, ^{202}Hg, ^{208}Pb.

Table 2

Laser ablation parameters

Parameter	**Value**
Laser beam wavelength	213 nm
Laser spot size	100 µm
Scan speed	70 µm/s
Laser beam fluence	2.5 J/cm^2
Repetition rate	10 Hz
Carrier gas flow	1.0 L/min He

Because all samples were split into 2 pieces, line scan pattern was used for their ablation. The sample was moved during laser ablation with constant scan speed with straight line trajectory. First ablation pattern was divided in two parts and went from the center of the tree ring to edge of the sample. Second pattern went perpendicularly to the first one (*Fig.2.*).

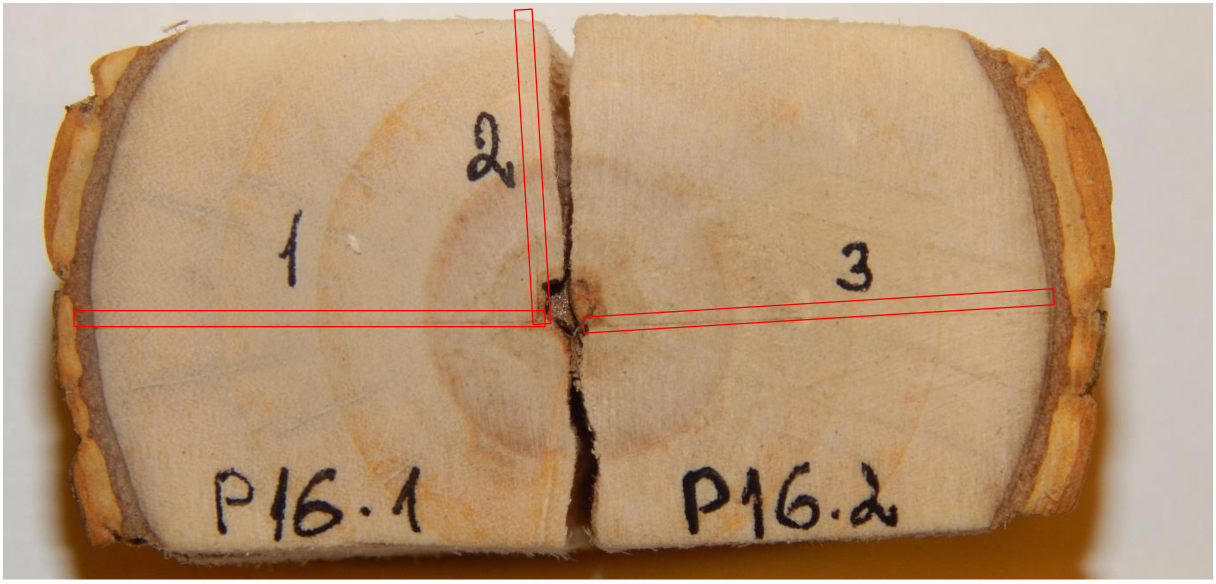

Fig.2. **Sample P16 after LA-ICP-MS run (numbers 1, 2 and 3 indicate ablation patterns).**

In case the surface of the samples was not straight and had the form of a hill (with central part of the line elevated in respect to start and end points), the line was divided in two parts to ensure proper focusing of the equipment and equal ablation conditions.

RESULTS AND DISCUSSION

In this report, only preliminary results will be discussed as the amount of results obtained (450 individual datasets) is too big to be properly analyzed during two weeks. In the following part of the report we will be focusing on the differences of trace element (Cr, Ni, Cu, Cd and Pb) content in poplar clones AF2, AF7 and AF8. A summary of average intensities for the selected elements in all ablation lines between the selected samples are summarized in Table 3. The results are being expressed as relative intensities because of the lack of proper reference material to carry out quantitative determination.

Table 3

The average intensity (cps) of selected trace elements in hybrid poplar clones AF2, AF7 and AF8

Sample	Clone	Line number	Cr		Ni		Cu		Cd		Pb	
			Int	±SD	Int	±SD	Int	±SD	Int	±SD	Int	±SD
P13	AF2	1	**9**	*11*	**1**	*2*	**10**	*5*	**4**	*3*	**7**	*11*
		2	**13**	*17*	**1**	*6*	**12**	*15*	**4**	*3*	**8**	*19*
		3	**11**	*23*	**2**	*4*	**14**	*13*	**6**	*9*	**12**	*22*

P19		1	**6**	*11*	**3**	*14*	**25**	*74*	**5**	*7*	**8**	*6*
		2	**17**	*38*	**6**	*7*	**33**	*32*	**4**	*3*	**21**	*22*
		3	**10**	*32*	**3**	*19*	**19**	*18*	**6**	*6*	**12**	*11*
P16	AF7	1	**19**	*35*	**6**	*4*	**38**	*22*	**6**	*4*	**37**	*21*
		2	**7**	*12*	**3**	*6*	**10**	*11*	**2**	*3*	**10**	*14*
		3	**8**	*28*	**4**	*5*	**25**	*13*	**4**	*3*	**19**	*15*
P18		1	**5**	*10*	**0**	*3*	**9**	*6*	**2**	*2*	**5**	*7*
		2	**7**	*7*	**0**	*1*	**10**	*6*	**1**	*2*	**6**	*6*
		3	**4**	*5*	**0**	*1*	**9**	*7*	**1**	*4*	**4**	*10*
P17	AF8	1	**16**	*45*	**1**	*6*	**15**	*7*	**3**	*4*	**13**	*12*
		2	**17**	*13*	**6**	*6*	**47**	*56*	**5**	*9*	**42**	*50*
		3	**10**	*14*	**1**	*5*	**15**	*9*	**3**	*10*	**13**	*17*
P15		1	**3**	*7*	**1**	*2*	**14**	*8*	**2**	*3*	**6**	*4*
		2	**8**	*17*	**4**	*14*	**23**	*10*	**3**	*4*	**16**	*9*
		3	**3**	*11*	**0**	*1*	**85**	*704*	**2**	*3*	**43**	*239*

The results show that for all selected trace elements the standard deviation value is between 50% and in some cases, more than 300% of the mean intensity value. This effect is due to the complex matrix of the analyzed sample that contains various high-molecular compounds. As the trace element content is very low, even slight variations in the amount of aerosol obtained in ablation process can have great impact on the results. For all samples and trace elements there are no significant differences between lines 1, 2 and 3 which means that the trace elements have been distributed evenly in both horizontal and vertical directions of the tree discs.

Figure 3 shows the relative intensities of chromium in hybrid poplar clones AF2, AF7 and AF8. The results show that highest chromium content is in the samples P16 and P18 and it suggests that the trees of clone AF7 have taken up higher amount of chromium from the soil thereby acting as a phytoremediator. For the samples P13 and P19 higher chromium content is observed closer to the core of the tree. For the clones AF2 and AF7 no considerable differences between samples are observable whereas for the clone AF8 the chromium content in samples P15 and P18 differs significantly. Therefore, it shows that the uptake of chromium depends on its content in the soil.

Fig.3. **Chromium content in hybrid poplar clones AF2 (samples P13, P19), AF7 (samples P16, P18) and AF8 (samples P15, P17).**

Figure 4 pictures the differences of nickel content between the selected samples and hybrid poplar clones. The sample P16 (clone AF7) has the highest nickel content while all other samples have lower nickel content with some visible peaks across the scan pattern. To evaluate the location of nickel in the sample with respect to tree rings, it is necessary to carry out precise measurements across the scan pattern to assess if the peaks shown below coincide with three rings or some imperfections in the wood. Altogether it can be concluded that hybrid poplars can take up the nickel from soil and act as phytoremediators.

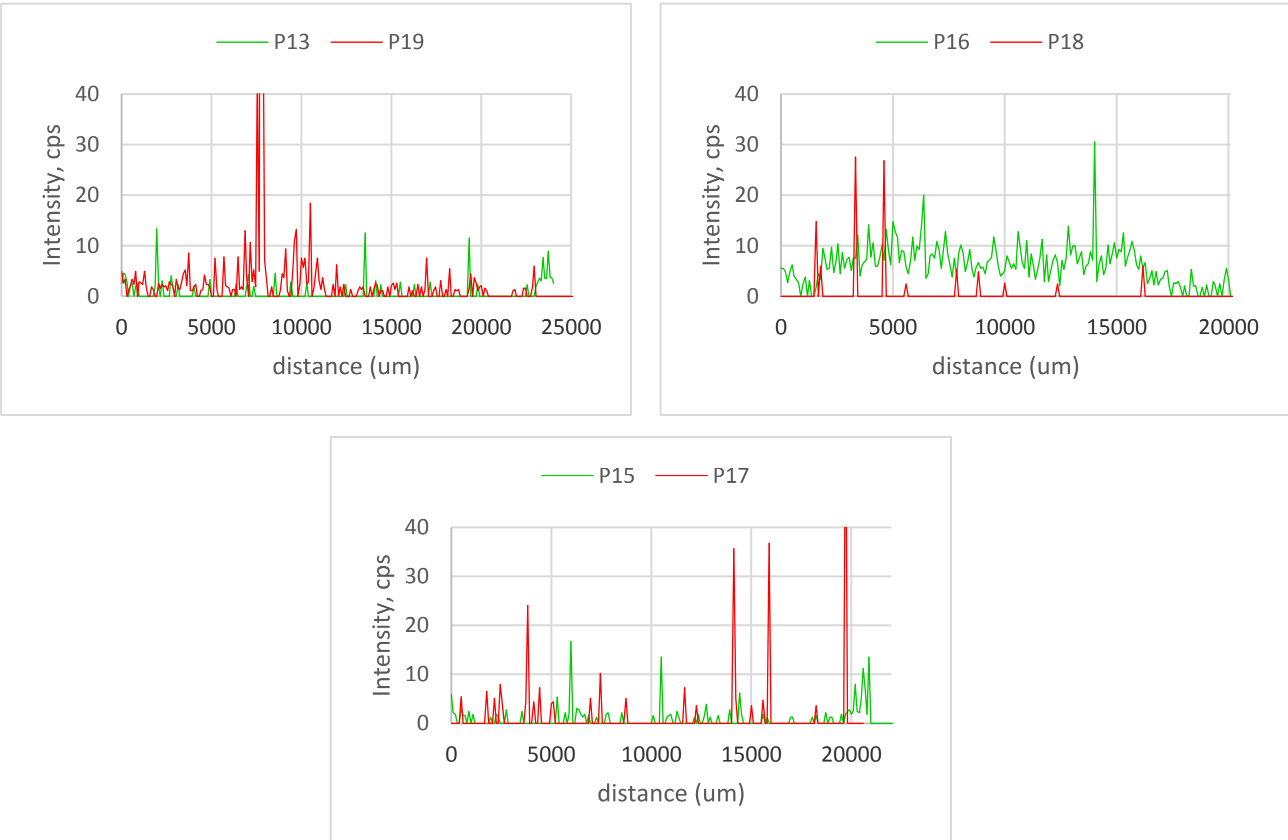

Fig.4. **Nickel content in hybrid poplar clones AF2 (samples P13, P19), AF7 (samples P16, P18) and AF8 (samples P15, P17).**

Figure 5 shows the copper content in hybrid polar clones AF2, AF7 and AF8. It is clearly visible that the sample P16 has higher copper content than any other sample. There is higher copper content also in the sample P19, but in comparison with sample P16 there are more peaks and the baseline of scan pattern is at 17-19 cps whereas for sample P16 it is at 30-40 cps. The mean values for all the poplar clones chosen coincide within the values of standard deviations and therefore there is no significant difference in the ability to uptake copper ions from soil. For the samples P13, P15, P16, P18 and P19 there is a visible peak at the end of scan pattern which indicates that the scan ended in the tree bark with higher trace element content.

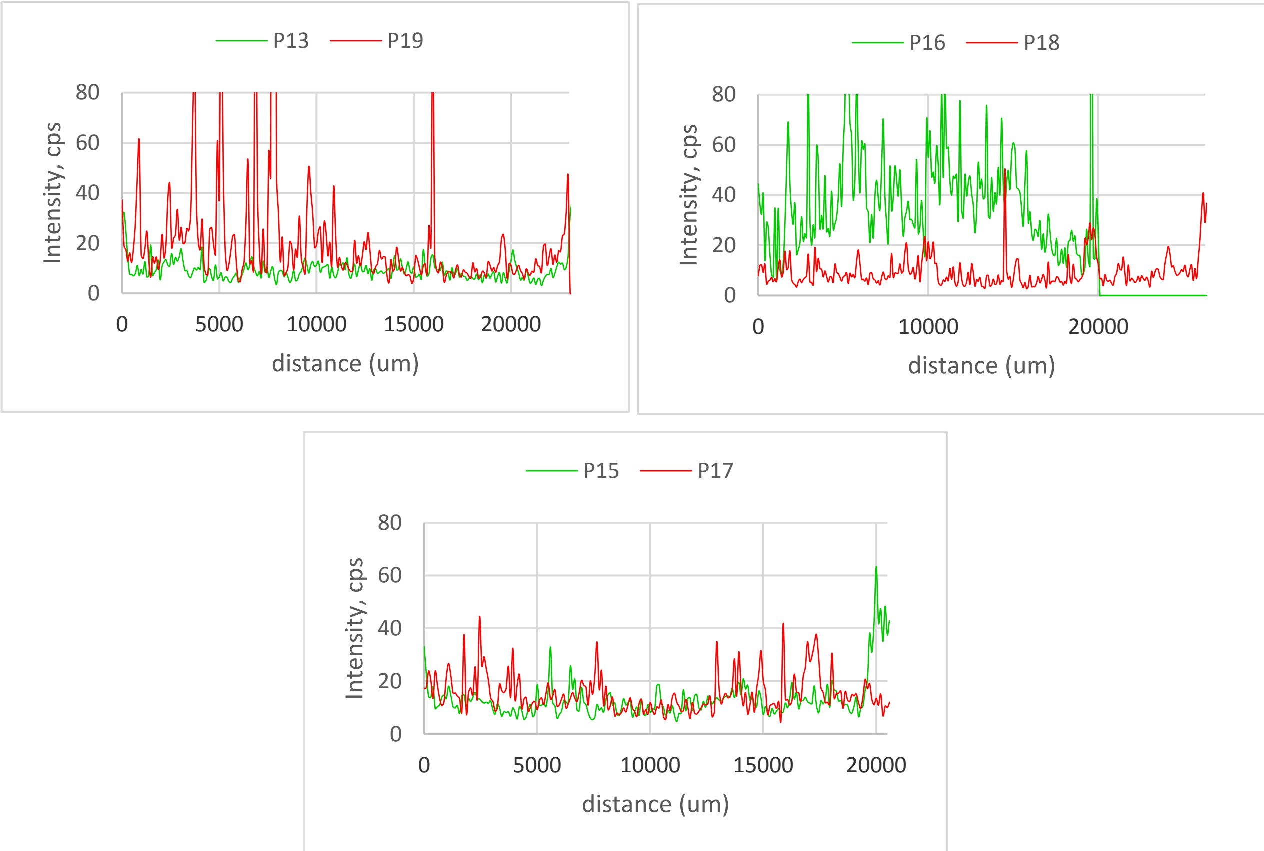

Fig.5. **Copper content in hybrid poplar clones AF2 (samples P13, P19), AF7 (samples P16, P18) and AF8 (samples P15, P17).**

Figure 6 shows the distribution of cadmium along the scan pattern – Line 1 of each of the chosen samples. It is clearly visible that the overall cadmium content varies only a little across the tree rings which indicates that cadmium has been taken up evenly throughout the growing seasons. Only sample that shows higher cadmium content is the sample P16 (clone AF7). The increment in cadmium content at the end of scan pattern for samples P13 and P19 indicates that the ablation pattern finished on the bark.

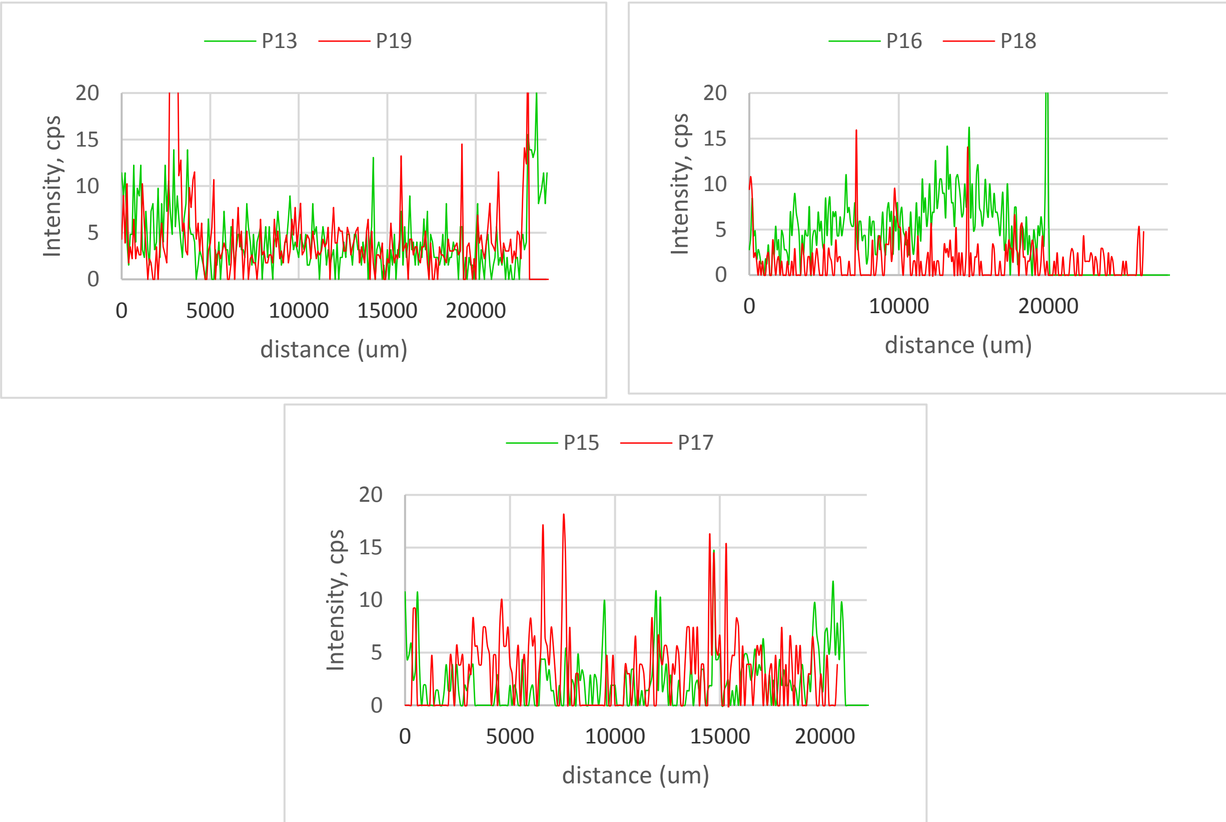

Fig.6. **Cadmium content in hybrid poplar clones AF2 (samples P13, P19), AF7 (samples P16, P18) and AF8 (samples P15, P17).**

Figure 7 shows the variations in lead content in the hybrid poplar clones AF2, AF7 and AF8. The lead content does vary between trees of the same clone, thus indicating that lead uptake depends on its content in the soil. Sample with highest lead content is P16. Next steps for proper data evaluation would be to measure the scan pattern length and compare placement of most intensive peaks with location of tree rings and other significant parts of the sample.

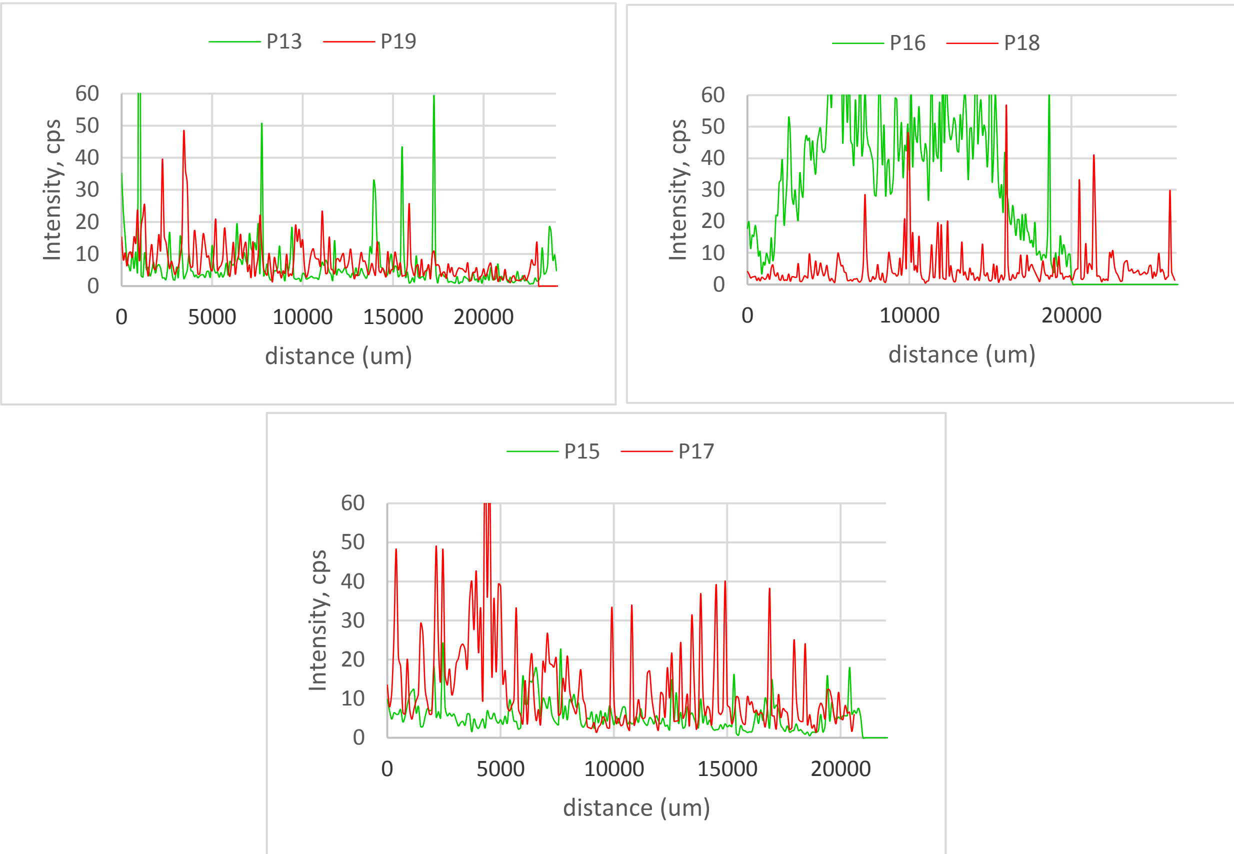

Fig.7. **Lead content in hybrid poplar clones AF2 (samples P13, P19), AF7 (samples P16, P18) and AF8 (samples P15, P17).**

Comparison of trace element content in the selected samples for hybrid poplar clones indicates the tendency for the sample P16 to have taken up considerably larger amounts of the trace elements. Meanwhile there are no significant differences of trace element uptake between clones. The presence of the selected toxic heavy metals in the selected samples indicates the potential of hybrid poplars to be use for phytoremediation. To describe the potential in detail, broader analysis of the obtained data will be performed.

CONCLUSIONS

- LA-ICP-MS is adequate method for evaluation of various macro and micro element uptake in wood from soil. The precision and trueness of results depends on the preparation of samples and the sample matrix. Elaboration of suitable reference material is necessary for future data acquisition and evaluation.
- There have not been observed any significant differences in the intensities of selected heavy metals between various clones of hybrid poplar which indicates that all the clones analyzed in the experiment show equal capacity for heavy metal uptake.
- Hybrid poplars are suitable to use for phytoremediation as they show the capacity of taking up various metallic elements. To describe the phytoremediation potential in detail, further data analysis will be performed and the results published.

Signed for acceptance

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