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PHYTOREMEDIATION OF CONTAMINATED SOILS IN THE VICINITY OF PROBISHTIP, THE REPUBLIC OF NORTH MACEDONIA

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Abstract

Phytoremediation is one of the most suitable techniques for the remediation of heavy metals from polluted soils. The purpose of this paper is to determine the efficiency of several plants varieties for phytoremediation of soils contaminated with lead, iron and arsenic in the vicinity of the municipality of Probishtip.

The research was done in the period from May to September 2023. During this period, representative soil samples were taken from several locations in the vicinity of Probishtip. These samples were subjected to laboratory testing to determine the concentration of the elements that compose it. After receiving the results, certain types of plants are planted in the soil samples. The same soil samples are subjected to laboratory testing again after a certain period of time, sufficient for the plants to have an impact on the soil.

The results of the soil analysis and risk assessment indicate the presence of heavy metal contamination in the soils surrounding Probishtip, especially in samples near Cranfield and the old tailings dump. The most frequently found metals include iron, lead, zinc, arsenic, copper, and chromium, all of which are the focus of our research. It can be inferred that the heavy metal content before planting the crops is higher than after planting, suggesting a process of soil remediation is underway.

Key words: *Phytoremediation, contamination, heavy metals, plants, Probishtip.*

INTRODUCTION

The Republic of North Macedonia is faced with a multitude of environmental challenges, including soil contamination. With its deep-rooted past of industrial pursuits and agricultural practices, the country's soils retain the marks of contaminants such as heavy metals, organic compounds, and agricultural residues [1]. Faced with these environmental legacies, North Macedonia stands at a crossroad, striving for innovative and sustainable approaches to remediate its polluted lands and safeguard its natural resources.

The soils of North Macedonia, despite their agricultural richness, carry the wounds of historical industrial activities and contemporary farming practices. The utilization of soy for phytoremediation is a relatively rare practice in the Republic of North Macedonia, where environmental remediation efforts have primarily focused on traditional methods. Only a small number of investigations have explored the potential of soybean in mitigating soil pollution caused by heavy metals [2]. Heavy metals like lead, cadmium, and chromium leached from mining ventures and industrial locations, pose a hazard to both ecological well-being and human health [3]. Similarly, the excessive use of agrochemicals and the improper disposal of organic refuse have bequeathed a heritage of soil contamination, imperiling the nation's agricultural yield and food security.

Within this realm of solutions, phytoremediation has emerged as a promising and environmentally sustainable approach for alleviating heavy metal pollution in soils [4].

The implementation of phytoremediation technologies entails introducing specific plant species to contaminated areas for the purpose of extracting pollutants from the soil. Unlike conventional remediation practices that often entail excavation and disposal of tainted soil, phytoremediation offers an in-situ, cost-efficient, and visually appealing resolution to soil contamination [4]. Moreover, it can enhance soil fertility and ecosystem functions, thereby augmenting the overall environmental quality.

Several investigations have showcased the plants' capacity to absorb, break down, or confine pollutants such as heavy metals, organic substances, and even radioactive components from soil, water, and air [5-10]. These studies have delved into the mechanisms driving phytoremediation procedures, elucidating the distinct capabilities of plant species in assimilating and neutralizing contaminants.

However, the journey towards integrating phytoremediation in North Macedonia is not without hurdles. The discovery of appropriate botanical species, adapted to the nation's climatic circumstances and adept at flourishing in polluted terrains, presents a significant challenge. Furthermore, the effectiveness of phytoremediation techniques may vary depending on soil properties, levels of pollutants, and the extent of contamination.

In spite of these obstacles, North Macedonia stands ready to utilize its inherent assets and human ingenuity to unleash the possibilities of phytoremediation. The country's multifaceted biodiversity, incorporating a broad spectrum of native plant varieties, offers a wealth of resources for exploring and implementing phytoremediation. Additionally, partnerships with international organizations, research institutes, and community initiatives can provide valuable support and expertise in propelling phytoremediation initiatives forward.

In conclusion, phytoremediation emerges as a guiding force in North Macedonia's quest for environmental sustainability. By embracing this innovative approach, the country can pave the way toward a greener, cleaner future. Through phytoremediation, polluted soil can be transformed into fertile landscapes brimming with life and vitality.

Through collaboration, creativity, and dedication, North Macedonia can reverse the trajectory of soil pollution and lay the groundwork for a more promising tomorrow.

DETERMINATION AND LOCATION OF THE STUDY AREA

In the Republic of North Macedonia, there are many places where the soil is polluted. It has been found that approximately 1000 hectares of land in the area along the course of the Zletovska Reka river are contaminated with cadmium [11]. This includes not just large areas affected by mining, but also many other places where industry and other human activities have caused pollution [12].

For the purposes of this research, we chose the area around the city of Probishtip to investigate the potential of phytoremediation, using different plant species to clean up soil pollution caused by heavy metals (Figure 1).

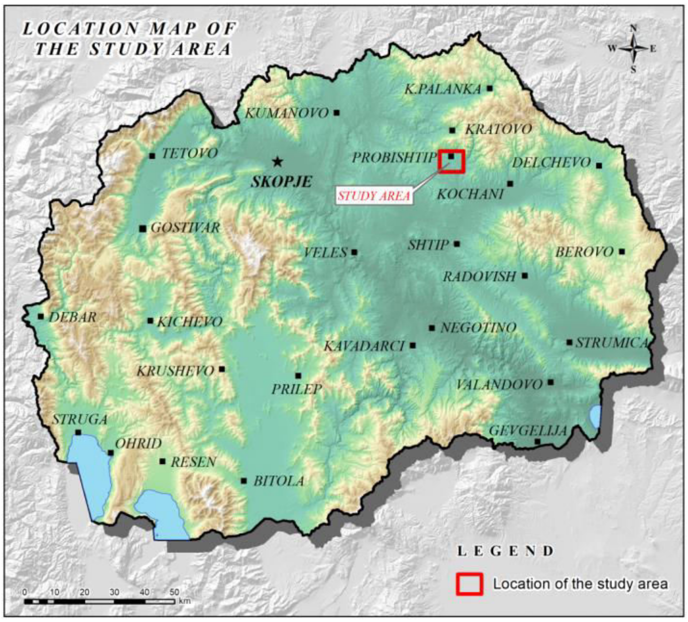

Figure 1. Location map of the study area

Probishtip, which is in the center of the Kratovo-Zletovo volcanic area, recognized for its storied mining past, plays a pivotal role in this research initiative. The extensive mining activities in the vicinity have left a significant impact on the local environment, profoundly affecting soil quality and ecological balance [12]. In close proximity to the city lies the lead and zinc mine "Zletovo" Alongside mining activities, Probishtip boasts industrial capabilities for manufacturing batteries and accumulators.

"ZLETOVO" MINE AND INDUSTRIAL HAZARDOUS WASTE FROM THE MINE'S HYDRO TAILINGS POND

The first mining activities in the Zletovo area were registered in the Roman period. The earliest reliable data on mining in this region date back to the 20th century AD. In the period 1935 - 1941 technological tests and preparations for the opening of the "Zletovo" mine were being carried out, as well as the construction of the necessary infrastructure for the exploitation and processing of the ore [12].

After the end of the Second World War, the rebuilding and establishment of the mine and the Probishtip municipality commenced, setting the stage for the accelerated progress at the "Zletovo" mine. The initial exploitation operations at the "Zletovo" mine, marking a significant milestone in Macedonia, begin with 1,000,000 tons of confirmed geological reserves of the lead-zinc ore. The content of the main metals on average is: 9% lead, 3% zinc, 45 g/t silver, and increased contents of cadmium, bismuth, gold, arsenic, indium, gallium, germanium, etc. The main ore minerals and carriers of lead and zinc are galena and sphalerite, regularly followed by chalcopyrite, tetrahedrite, tennantite, pyrite, bornite, enargite, etc.

The old hydro tailing of the lead and zinc mines "Zletovo" - Probishtip (Fig. 2), dates back to the beginning of the exploitation of the mine and for decades huge amounts of material have been deposited at this location [12]. In the period so far, about 6,000,000 m^3 of hydro tailings have been deposited at the old hydro tailings. The reservoir is a plateau in five fields, with a total height difference of about 30 m. By depositing the hydro tailings, the original relief of the terrain has been completely changed. There are flat surfaces, plateaus, in five different levels (fields 1 to 5) and a slope that follows the bed of the River Kiselička from the beginning of field 1 to the end of field 5. The hydro tailing contains heavy metals (lead, cadmium, zinc, mercury, iron, copper, arsenic, uranium), which pose a danger to all environmental media [13].

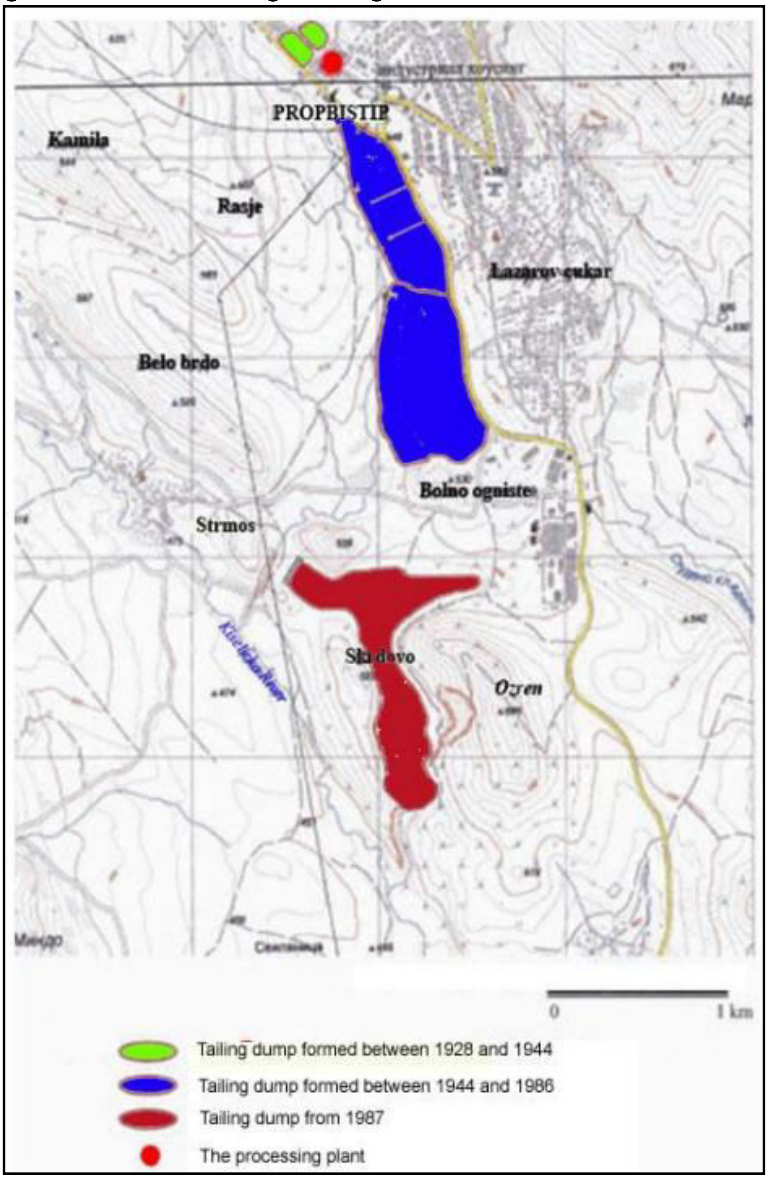

Figure 2. Location of tailings from the Zletovo mine

The hydro tailing also contains chemicals, such as: copper, sulfate, sodium cyanide, potassium amyl-xanthate, calcium oxide, calcium carbonate, which were used in the process of wet chemical processing of ore during wetting, after mills and spiral classifiers as reagents for pH adjustment.

In the agricultural land in the area of the villages of Strmosh, Neokazi, Petrishino and Buchishte, there is an increased percentage of lead, zinc, copper, cadmium and manganese [13].

The tailing pond lacks adequate environmental protection measures, such as covering its surface, treating wastewater released from its base, and controlling dust emissions during windy conditions.

SAMPLE LOCATION & METHODS

A total of four locations were selected in the vicinity of the town of Probishtip (Figure 3). From these four locations, a total of 8 samples were collected, with two samples taken from each location, each from different depth, at the surface of the field to a depth of 20 centimeters and from a depth between 20 and 40 centimeters. The first two samples were taken on an area of 80x80 cm, while the last two samples on an area of 40x40 cm (Table 1). The samples were collected mostly from moist soil, from which three samples were collected from uncultivable land and one sample was taken from cultivated land (vineyard).

Table 1. Main data for the samples taken

SAMPLE	LOCATION	COORDINATES		SAMPLING DEPTH [cm]		SURFACE AREA [cm]
		Y	X	0-20	20-40	
Sample 1	Stara Kasapnica	7 597 886	4 650 871	x		80x80
					x	
Sample 2	Old tailings dump	7 598 079	4 649 439	x		80x80
					x	
Sample 3	Vineyard - v.Strmosh	7 598 182	4 649 237	x		40x40
					x	
Sample 4	Cranfield	7 599 271	4 648 070	x		40x40
					x	

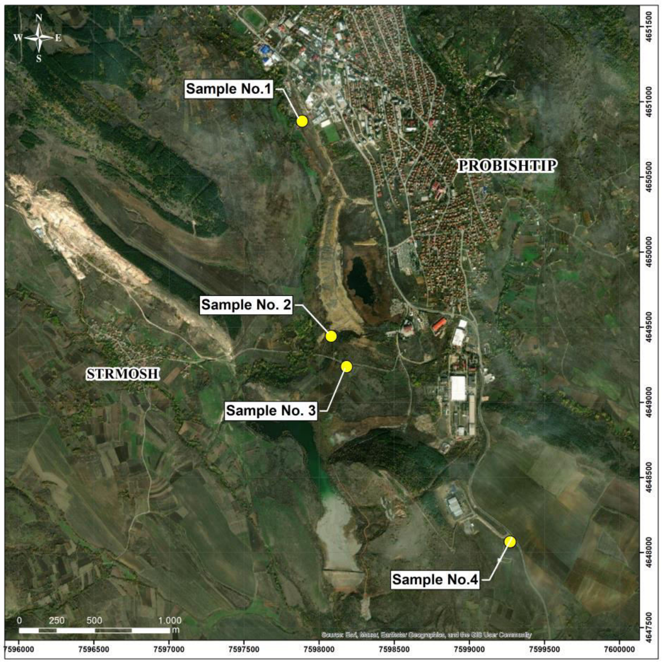

Figure 3. Sample location map

All samples were collected with appropriate equipment, in order to minimize contamination and ensure representativeness. Precisely marked, the samples are submitted for further laboratory processing. Since the primary sample is inhomogeneous in composition, it is subject to its processing. Processing of the sample consists of three basic operations: crushing and grinding, mixing (homogenization), and shortening. If the weight of the primary sample is two or more times greater than the minimum required, the procedure can be modified, that is, crushing may be omitted. The mixing operation should be repeated three to four times, followed by shortening of the sample. In this research, mechanical shortening, that is, Johnson's splitter, was used (Fig. 4, b).

a) b) c)

Figure 4. Research methodology: sampling, sample reduction and chemical analysis with a fluorescence spectrometer (from left to right).

Finally, X-ray fluorescence spectrometer using semi-quantitative analyses was used to determine the chemical composition of the soil. The Energy Dispersive X-ray Fluorescence Spectrometer (EDX) is an instrument for performing qualitative and quantitative analysis of elements ranging from Na to U and it is an ideal tool for non-destructive applications (Fig. 4c).

CHEMICAL ANALYSIS OF SOIL BEFORE PLANTING

Table 2 shows the results of the chemical analysis of the collected soil samples using a fluorescence spectrometer. The composition of elements in the soil is analyzed, with particular focus on heavy elements, which are of significant research interest.

Table 2. Results of chemical analysis of the soil samples before planting

Element	**S1 (mg/kg)**	**S1 (mg/kg)**	**S2 (mg/kg)**	**S2 (mg/kg)**	**S3 (mg/kg)**	**S3 (mg/kg)**	**S4 (mg/kg)**	**S4 (mg/kg)**
	Depth (0-20cm)	Depth (20-40cm)	Depth (0-20cm)	Depth (20-40cm)	Depth (0-20cm)	Depth (20-40cm)	Depth (0-20cm)	Depth (20-40cm)
Si	410980	430260	<1	<1	460610	437990	551510	579530
Fe	599460	341035	743640	755370	260620	259050	269410	277215
Mn	154170	106680	52500	50620	23055	21440	20025	11110
Ba	57340	36890	20270	12040	12920	15080	12705	12645
K	44220	38550	92870	82400	53995	51440	63560	71270
Pb	68870	31650	35670	35240	6580	3875	4425	1645
Zn	50070	22355	24190	22710	6490	3095	3335	1030
Ca	9520	7395	2480	4530	173860	199040	68850	60560
Sr	6560	2850	7250	10970	8460	11885	7590	7520

Element	S1 (mg/kg)	S1 (mg/kg)	S2 (mg/kg)	S2 (mg/kg)	S3 (mg/kg)	S3 (mg/kg)	S4 (mg/kg)	S4 (mg/kg)
	Depth (0-20cm)	Depth (20-40cm)	Depth (0-20cm)	Depth (20-40cm)	Depth (0-20cm)	Depth (20-40cm)	Depth (0-20cm)	Depth (20-40cm)
As	2800	1595	6630	5610	<1	<1	<1	<1
Cu	3220	1500	1660	1580	905	800	790	915
Zr	3760	<1	6680	10460	1900	1060	1440	1820
Sn	830	<1	<1	<1	1810	1110	1040	1320
Cr	285	120	<1	790	615	1005	705	755
Ti	2230	<1	9200	12880	10730	12890	12650	12760
Ag	<1	520	1840	<1	<1	1020	<1	<1
Ac	<1	<1	4180	<1	4090	<1	<1	<1
Rb	<1	<1	3300	3670	1000	1405	1640	1570
Y	<1	<1	<1	<1	165	430	325	290
S	<1	<1	<1	<1	10920	<1	<1	<1
Mo	<1	<1	<1	<1	<1	90	<1	<1

From the above table, we observe the presence of heavy metals, posing a serious problem and threat to agricultural production, which forms the fundamental basis for food production. These heavy metals reach the soil via the air/atmosphere, where they are absorbed by plant roots and accumulate in the plant's softer tissues. Eventually, they are deposited in soils and water bodies as poorly soluble compounds, such as carbonates, sulfates, or sulfides.

PLANTING

In this research, the purpose of planting the plants is to see how many elements the plants can extract from the soil and if the same soil in which they are planted is suitable for sprouting a crop. In this research, we are talking about wheat, grass, soybeans, and beans (Fig. 5).

Figure 5. Plantations with soybeans, wheat, and beans (from left to right)

CHEMICAL ANALYSIS AFTER PLANTING

After the sprouting of the plants, a chemical analysis of the soil was done again, which determined the representation of the elements in the soil after planting, that is, how much concentration the plants extracted from the soil.

The analysis of the soil was done with a fluorescence spectrometer, and the obtained results are shown in Table 3.

Table 3. Results of the chemical analysis of the soil samples after planting

Element	S1 (mg/kg)	S1 (mg/kg)	S2 (mg/kg)	S2 (mg/kg)	S3 (mg/kg)	S3 (mg/kg)	S4 (mg/kg)	S4 (mg/kg)
	Grass	**Wheat**	**Grass**	**Wheat**	**Soya**	**Beans**	**Soya**	**Beans**
Si	<1	427910	<1	<1	435020	339810	480700	559330
Fe	353330	327800	727120	749010	342850	357815	312910	238770
Mn	99540	93675	63340	62160	11180	15910	8440	7760
Ba	29260	31165	16700	8280	12190	13925	11180	12490
K	39055	36600	89130	80200	48110	47050	58395	65445
Pb	32015	29290	34210	34830	<1	2070	1760	1600
Zn	19235	20010	21090	20240	<1	1610	800	730
Ca	8495	6960	5210	8390	100105	178810	84870	49145
Sr	2495	2720	6950	9140	6700	8220	7440	6580
As	1570	1330	5320	5300	<1	<1	<1	<1
Cu	1580	1460	1270	1290	600	510	685	750
Zr	1430	<1	4560	<1	<1	420	940	1510
Sn	<1	<1	<1	<1	<1	<1	<1	<1
Cr	<1	<1	<1	<1	550	690	700	720
Ti	<1	<1	9100	9100	8220	11190	10895	12395
Ag	<1	<1	<1	<1	<1	<1	<1	<1
Ac	<1	<1	<1	<1	1060	<1	<1	<1
Rb	<1	<1	3160	3170	950	880	1325	1430
Y	<1	<1	<1	<1	170	200	300	200
S	<1	<1	<1	<1	1540	<1	<1	<1

From the results shown in Table 3, it can be noted that the soybeans planted in the locations "Vineyard" and near "Cranfield" have significant differences in the concentration and content of the elements represented in the soil.

The contents of Fe, Mn, Ba, K, Ca, Sr, As, Rb, Cr, Ti and Y are reduced, i.e., "extracted" from the soil and occur in lower concentration, while the contents of Pb and Zn are completely extracted. In the soil taken from the site of the old tailings dump where grass and wheat were planted, nothing sprouted at all, which indicates the negative influence and the presence of heavy metals that prevent the growth of any culture. In that trial, from the results of that experiment, only small differences were observed in the content of the elements present in the soil.

The sources of soil contamination with heavy metals in the area are related to mining and related activities, but there are also zones of natural mineralization, geological units, etc. The sources of contamination in the area consist of the old tailings dump of mining waste, tailings, etc. Although primary soil contamination occurs in and around the original sources, heavy metal contamination gradually spreads over a larger area through surface water, groundwater, and dust. The agricultural risk of crops is assessed using standard values of heavy metals in our country.

In the following part, there are diagrams (Fig. 6) of the contents of metals in the soil before and after planting the plants, which shows the difference in their concentrations. From the diagrams it can be concluded that plants are good absorbers of soil elements.

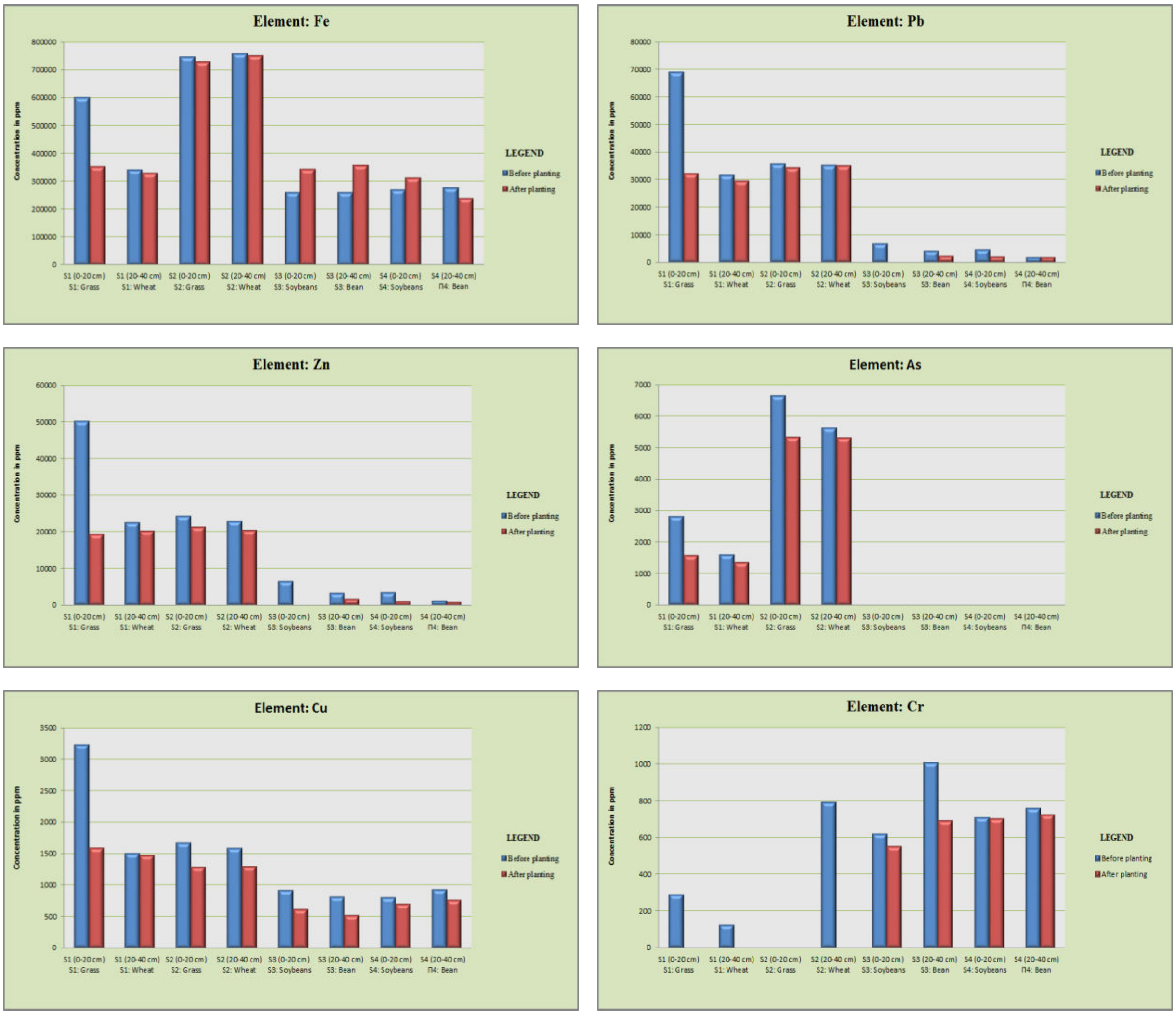

Figure 6. Content of certain elements before and after planting

SOYBEANS PLANT TEST RESULTS

Since soybean is recognized for its ability to bioaccumulate elements, particularly heavy metals, from the soil, we chose to include it in this experiment. As previously discussed, the use of soy for phytoremediation is a relatively uncommon practice in the Republic of North Macedonia, with only a limited number of similar investigations conducted before.

Table 4 shows the results of the concentrations of elements extracted by soybeans from the soil samples taken near "Cranfield".

Table 4. Results of the content of elements extracted by soybeans from the soil samples taken near "Cranfield"

ELEMENT	SYMBOL	CONTENT [mg/kg]	ELEMENT	SYMBOL	CONTENT [mg/kg]
Aluminum	Al	5.19	Lithium	Li	0.83
Antimony	Sb	<0.0001	Magnesium	Mg	744

ELEMENT	SYMBOL	CONTENT [mg/kg]	ELEMENT	SYMBOL	CONTENT [mg/kg]
Arsenic	As	<0.0005	Manganese	Mn	10.8
Copper	Cu	14.8	Molybdenum	Mo	0.73
Barium	Ba	0.084	Sodium	Na	134
Beryllium	Be	0.0046	Nickel	Ni	0.72
Bismuth	Bi	<0.001	Lead	Pb	0.19
Boron	V	7.62	Selenium	Se	0.033
Vanadium	V	0.18	Palladium	Pd	<0.0005
Gold	Au	<0.001	Platinum	Pt	<0.001
Indium	In	0.067	Silver	Ag	<0.001
Gallium	Ga	<0.0005	Strontium	Sr	5.63
Germanium	Ge	0.019	Thallium	Tl	0.0017
Iron	Fe	112	Tellurium	Te	<0.0005
Mercury	Hg	<0.001	Titan	Ti	0.038
Rubidium	Rb	0.59	Chromium	Cr	0.67
Cadmium	Cd	0.041	Phosphorus	P	2274
Potassium	K	4.775	Zinc	Zn	45.8
Calcium	Ca	1.766	Cesium	Cs	0.031
Tin	Sn	<0.0001	Rhodium	Rh	<0.001
Iridium	Ir	<0.001	Ruthenium	Ru	<0.001
Cobalt	Co	1.11			

While they may not be as commonly linked to phytoremediation as other plants, as can be seen from the table, soybeans show excellent abilities to absorb elements from the soil. This is primarily due to the deep root system of soybeans, which enables deep penetration into the soil, enabling them to reach contaminants that have permeated the soil.

CONCLUSION AND RECOMMENDATIONS

The results of the soil analysis as well as the risk assessment reveal the presence of heavy metal contamination in the soils around Probishtip, particularly in the soil samples near Cranfield and the old tailings dump. The most common metals are: iron, lead, zinc, arsenic, copper and chromium, which are actually the subject of research. It can be concluded that the content of heavy metals before sowing the plants is higher than the content of heavy metals in the soil after sowing, and this is a good indicator for the conclusion that there is a process of reanimation of the soil. Certain forms of legumes such as soybeans have been shown to be bio-extractors and can be used to extract heavy metals from soil.

Taking into account the results obtained from the chemical examination of the soil samples, as well as the results obtained from the results of the elements extracted from the soil by soybeans, the question of increased use of soybeans for this purpose arises.

Taking into account that in the vicinity of Probishtip arable land is spread over large areas, it is recommended that part of that area be planted with soybeans in the future, as one of the steps to remediate the soil, which in this area is quite contaminated, especially by heavy metals. Soybeans are known for their relatively fast growth rate, which means they can quickly establish themselves and begin the remediation process.

We can summarize that the phytoremediation process represents an extraordinary synergy between nature and technology in the pursuit of environmental restoration. By harnessing the inherent abilities of plants to absorb, accumulate and transform pollutants, this innovative approach offers a sustainable solution to a variety of pollution challenges, from heavy metals and pesticides to hydrocarbons and industrial waste. As research progresses and new plant species are identified for specific pollutants and environments, the scope and effectiveness of phytoremediation continues to expand.

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