

# Usage of Perlite in Polluted Sandy Soils for Potato Crop

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*After three years since pollution by oil, petroleum byproducts practically disappeared (it were found only 1.57 g/kg total petroleum hydrocarbons represented by heavy compounds such as asphaltenes) but, the soil was more salty of about 3.8 times higher (299.1 ppm) than the unpolluted one (78.7 ppm). A greenhouse experiment was carried out to study the response of potato (*Solanum tuberosum*) to different improved salt affected soils with Perlite (4 mm). The highest production (300g/plant), similar to the one obtained on unpolluted soil (320g/plant), was obtained on soil variant mixed with 75% Perlite while the production on plant grown on polluted soil was lowest (55g/plant). In the case of plants grown on the substrate that contained 75% Perlite, protein and sodium content both for tubers and leaves are very close to those as that found in plants grown on unpolluted soil. However, the sodium content in the tubers of potatoes grown on polluted soil was almost 2 time higher than in the tubers of potatoes grown on unpolluted soil. Our results show that the salty soil ameliorated with 75% Perlite offer growing conditions similar to those of an unpolluted soil.*

**Keywords:** salty soil, potato, production, protein, sodium, Perlite

In the process of oil extraction salt water injections are used under pressure in order to increase the production of crude oil drawing out. Thus, in the areas of extraction and of oil transport takes place pollution of the soil, underground and aquifers not only with oil products but also with saline solutions. In this context soil pollution is complex because concurrently with oil pollution occurs and of wastewaters pollution, salty waters capable of provoking a strong soil salinization, fact that cause a decline in or even destroy soil micro-fauna which seriously endanger the production capacity of the soil.

Phytoremediation, tested around the world through a series of techniques and plants, is considered a promising field for future remediation of the contaminated areas with various contaminants [1, 2]. At the roots level, plants and associated microorganisms adsorb the petroleum products. Some of these products pass into the plant where they accumulate, degrade or transform into volatile product that evaporates thus acting as filters. The adsorbed products in exudates from plant roots are metabolized and degraded by the microbial community [3, 4].

Most of phytoremediation research so far to remedy the petroleum product contaminated soils were geared specifically toward use of different varieties of grass, vegetables and grains [3, 5].

Thus, many studies show that the analysis of hydrocarbons degradation ability by the different herbs: *Cyperus rotundus*, *Axonopus compressus*, alfalfa, red clover, fescue, amaranth in temperate, tropical or cold climates [5 - 10].

One of the vegetables tested is potato a staple food for many people. By cultivating with potatoes of oil polluted soil has been shown that is obtained a significant decrease in the concentration of pollutants. When this concentration is in the range 1.5 to 3.2 ppm crude oil is obtained a decrease in range 75.4% to 77.5% through the potato

cultivation. At higher concentrations potato ability to metabolize hydrocarbons decreases [11]. But other vegetables (*Calopogonium mucunoides*, *Centrosema brasilianum*-butterfly peas, *Stylosanthes capitata*) showed a clear intolerance from petroleum products [9].

Were also tested and a number of other plants of the most consumed by people such as wheat, maize, soy, beans, sunflower, mustard. For instance, for some experiments with mixed grasses, mixed clover, sunflower and different grains cultivated on soils contaminated with used oil at a concentration of 1.5% after 150 days analyzes showed that the most effective phytoremediation process was the one in which clover was used followed by sunflower and mustard [12-14]. Cultivation of soybean (*Glycine max*) on soil polluted with crude oil but enriched with cow manure increases soil remediation capacity by this plant [15-17] tested in the laboratory peas and lentils tolerance to high concentrations of oil in the soil.

Interesting are the studies on maize tolerance towards petroleum products. Though they are made in the vast majority in the laboratory has been shown that corn has a high tolerance towards petroleum products, so the pollution of 21% leads to a production of about 60% compared to that obtained in the unpolluted soil conditions [18]. Several studies have taken into account the possibility of using Trees for remediation, especially willow, locust, poplar and mulberry. In the case of Acacia has been shown that on the polluted soil Acacia respond towards petroleum pollutants in soil by changing the content of flavonoids [19]. In some attempts to remedy polluted soils by using willow, has been shown that it does not tolerate larger amounts of 3 ppm petroleum product [20].

A promising technology is the use of plants whose rhizosphere contributes to the degradation of pollutants. Comparison of the capacity of bean (*Vicia faba*), of maize (*Zea mays*) and of wheat (*Triticum aestivum* L) to degrade

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desert soil polluted with oil products has led to the conclusion that the best results are obtained by cultivation in this type of soil of bean. In its rhizosphere oil degradation rate was 62.4%, while for the other plants tested was only 19.9% respectively 17.6% [21- 23].

At the same time, oil pollution is not confined only to the spread of the petroleum products on the ground. Oil extraction is done with large quantities of saltwater (deposit water). Although, the petroleum products are consumed by specific microorganisms occurring in the soil or added by humans, often the salts remain. Different studies established that some rhizobacteria help some plants to grow and provide its tolerance ability against salts [24].

In the salt-affected soils, the leaching of salts is very low, therefore, salt accumulates in soil surface layers. As a consequence, these soils have affected their physical properties by becoming clayey and losing the ability to be an agricultural land properly [25]. Numerous chemical, physical and biological methods were established to redress such soils [26 - 33].

On this premise, this study is aimed at assessing the effectiveness of *Solanum tuberosum* (potato) phytoremediation measure of salt-affected soils.

### Experimental part

The experiments were performed in the winter of 2014, in greenhouse of University of Agronomic Sciences and Veterinary Medicine - Bucharest with Colette variety potato (*Solanum tuberosum*).

The substrates used to experiment were unpolluted and polluted soils from Icoana commune, Olt County, Romania. Soil type from analyzed areas is black earth. It has been chosen this commune because it is often polluted with oil. The last pollution with oil held in autumn 2011 and soil sampling was conducted on 17.11.2014. The soils studied are of fertile black soil type and have an average EC of 1.8.

Potato crops were performed in a common greenhouse with glass walls and roof. Potato cultivars were conducted using unpolluted and polluted soils in five variants: V0-unpolluted soil, V1-polluted soil, V2-polluted soil plus 25% Perlite, V3-polluted soil plus 50% Perlite and V4-polluted soil plus 75% Perlite. Perlite used had a grain size of 4 mm, dry bulk density of 83 kg/m<sup>3</sup>, wet bulk density of 114 kg/m<sup>3</sup>, moisture capacity of 37, total porosity of 75%, air porosity of 36% and pH of 4.1.

Reason of using perlite in order to prepare soil variants is on one hand in that Perlite is widely available in Romania and, on the other, its price is much lower than other possibilities to remedy contaminated soil characteristics. In addition, perlite is inert, so in any way it not influence the composition and properties of the soil.

Potatoes were grown in pots, filled with the different variants of polluted soil and mixed perlite. The pots have been 16 cm depth, the upper diameter of 18 cm and bottom diameter of 12.5 cm. In each pot was placed one potato tuber, variety Colette, of 30 g each. Each variant was conducted in ten replications. Unpolluted soil cultures were performed on 30 liters capacity 1 m long systems on mattresses (variant V0).

Nutrient solution had an EC of 0.7 and a pH of 5.8. Greenhouse temperature was set to 23-24 °C during the day and to 17-18 °C during the night. The culture fertilization was always the same for all types of substrate.

The modified Hartree-Lowry method using bovine serum albumine as a standard was used to assess the total soluble protein [34, 35].

For measurements of Na<sup>+</sup> concentrations, the samples (5 g soil and plant tissues) were dried in 60°C for 48 h. Then

1 gr of sample was powdered and burned in 560°C to obtain ash then ashes digested in 10 ml of 1N HCL. The concentration of Na<sup>+</sup> in the digested samples was determined using a flame photometer (Model 410, Sherwood).

All spectrophotometric measurements were made by using a Metertek SP830 Plus UV-VIS scanning spectrophotometer, calibrated at 705 nm, using the 0.2 nm width measuring band beam and 1 ml cuvette having a path length of 1 cm.

Analysis of variance for crop production, protein content and Natrium content determinations was performed by Duncan test.

Each biochemical assay was performed at least 3 times and the three closest results are presented as average values.

### Results and discussions

Petroleum compounds analysis showed that they were almost totally consumed by the plants grown in the last three years and microorganisms from soil, from rhizosphere. It were found only 1.57 g/kg total petroleum hydrocarbons represented by heavy compounds such as asphaltenes. As a result of oil pollution followed by salinization, soil is mostly compact. This process modifies the properties of the soil, such as porosity and permeability. The movement of gas and water through the ground is prevented by interruption of the pores, causing the existence of a small amount of water and oxygen. Root growth is hampered. Water deficit or osmotic effects are among the major factors which limit crop production [36].

Phytoremediation experiments were conducted using unpolluted and polluted soils. The soil samples were collected from fields of Icoana commune, where an area of 3.8 ha was polluted by oil in autumn (November) of 2011. There have been taken 10 samples each from both categories of soil, polluted and unpolluted. Soil sampling was conducted on 17.11.2014. Because, analyzes have shown a lack of oil in polluted soil, it was analyzed only the sodium content. Each analyze was made at least three time and the averages of obtained results are presented in the table 1.

From the data presented it can observed that, although three years have passed since pollution to sampling (2011 to 2014), the sodium content of the polluted soil is about 3.8 times higher than the one in unpolluted soil.

After determining the sodium content, the samples corresponding to the two soil types (polluted and unpolluted) were blended so as to achieve a polluted homogeneous soil respectively an unpolluted homogeneous one. These homogeneous soils were used for experiments, being performed the 5 soil variants (V0 - V4).

After a period of 30 days from sowing the 5 variants were evaluated in terms of appearance. Figure 1 presents the plants developed on the different variants of soil.

The figure 1 clearly shows that on the salted soil, resulted from oil pollution, plants are less developed. In the case of potatoes, only one plant in three is most developed, the other are tiny with a reduced number of leaves. Plants grown on variant 4 of soil (polluted soil mixed with 75% perlite), though less high than those grown on version 3 (polluted soil mixed with 50% perlite) are much richer in leaves and have a healthier appearance than all other alternatives. This variant is very similar with the plants grown on unpolluted soil (V4 compared to V0). So, in terms of potatoes development has a significant influence the concentration of added perlite.

**Table 1**  
THE SODIUM CONTENT IN SOIL SAMPLES

Sample	Sodium content, mg/kg(ppm)	
	Polluted soil	Unpolluted soil
1.	345.0	68.9
2.	292.1	78.5
3.	273.2	71.2
4.	274.4	76.8
5.	288.7	76.1
6.	321.7	82.1
7.	265.8	79.8
8.	301.2	82.9
9.	351.2	85.3
10.	277.5	85.2
Average	299.1	78.7
H.S*	298.5	79.1

H.S\* - homogenized soil of collected samples

This can be explained by the fact that if the soil is helped to allow the aeration and circulation of water in the root zone, the results are satisfactory. These results are confirmed at harvest time. Thus, the plants grown on the polluted soil produced an average of two medium-sized tubers. Also notice that at least some of these tubers have a green coloration, which means that they contain solanine, an alkaloid toxic to humans. Plants grown on the soil with 25% perlite produced on average 4 - 5 tubers of different sizes.

Plants grown on the soil enriched with 50% and 75% perlite produced almost the same number of tubers namely 7 - 9, with the difference that those produced on the ground with 75% perlite are higher.

In addition the tubers developed on V4 variant are similar to the ones obtained on unpolluted soil (fig. 2).

On average, it were obtained 250g potatoes per plant grown on 50% perlite and 300 g potatoes per plant grown on the variant with 75% perlite. For potatoes grown on unpolluted soil were obtained 8-11 tubers of different sizes and an average of 320g per plant. Thus, potato plants respond to conditions offered by the soils where on grown. Relationship between the amounts obtained on different variants is presented in figure 3 and table 2.

In terms of tubers production, the unpolluted soil has the highest production followed closely by variant V4 (with 75% perlite), while the lowest production was obtained on polluted soil. Coordination between the crops and the properties of each substrate, mainly the aeration capacity, is obvious. Although the availability of nutrients was the same for all soil variants, the growth, maturation and production of plants developed on V1 (polluted soil) were lower against the results obtained for the all other variant.

Analyzing statistical potato production levels obtained on unpolluted, polluted and that improved with perlite we could appreciate the following:

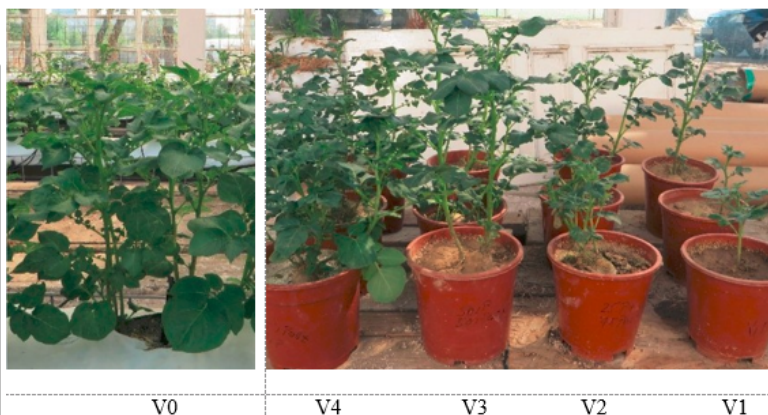


Fig. 1. Potato crops after 30 days from sowing on the 5 different soil variants

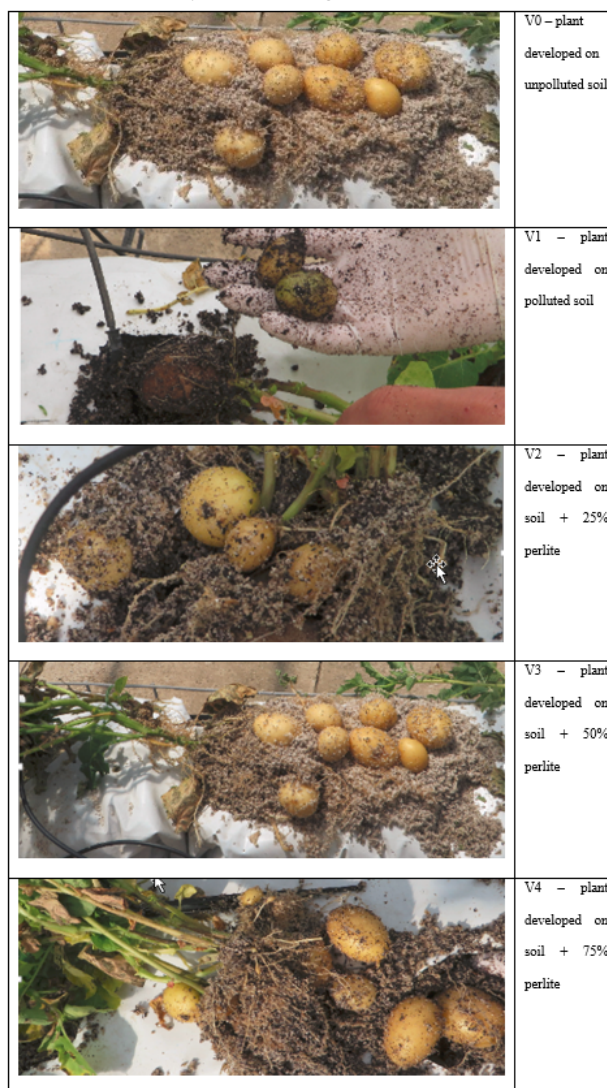


Fig. 2. Potatoes harvested from the 5 soil variants

-Harvests obtained on polluted soil, on plant were of 53.67 g with a difference under the control of 266.33 g. Compared to control the plant production was only 16.77%. The data are statistically confirmed by very significant negative difference.

-By improving polluted soil with perlite, in the case of variant V4 (Polluted soil + 75% perlite) the production increased to give 307.00 g / plant, which represents a difference compared to control of 95.94%. It is noted that of statistic point of view differences were significant negative.

The explanation reside in the clayey loam characteristics of the polluted soil which determine a slowly flow of the nutrient solutions and especially reduced

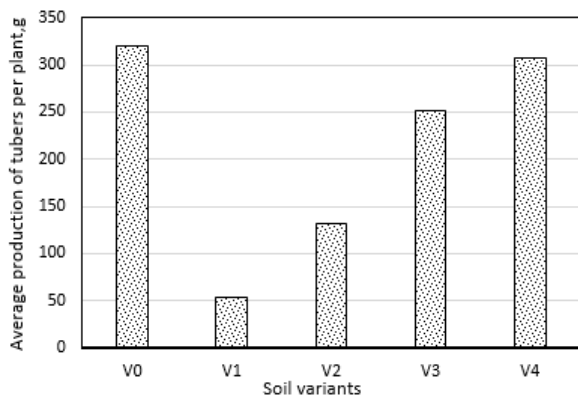


Fig. 3. The average amount of tubers per plant, in g, obtained on different soil variants

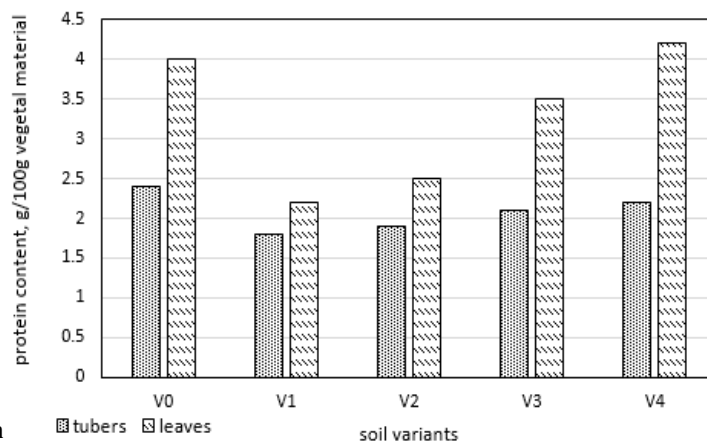


Fig. 4. Influence of culture substrates on potato leaves and tubers protein concentrations

Variant	Production, g potatoes per plant	Difference		Signification
		g	%	
Average	212.73	-107.27	66.48	000
Unpolluted soil, V0	320.00	0.00	100.00	Mt
Polluted soil, V1	53.67	-266.33	16.77	000
Polluted soil + 25% perlite, V2	132.00	-188.00	41.25	000
Polluted soil + 50% perlite, V3	251.00	-69.00	78.44	000
Polluted soil + 75% perlite, V4	307.00	-13.00	95.94	000
DL5% = 4.060	DL5% in % = 1.2687			
DL1% = 5.920	DL1% in % = 1.8500			
DL01% = 8.860	DL01% in % = 2.7687			

Fteoretic = (001%= 14.72574); (0.1%= 7.089267); (5%= 3.861993)

**Table 2**  
THE AVERAGE AMOUNT OF TUBES PER PLANT, IN g, OBTAINED ON DIFFERENT SOIL VARIANTS

capacity of absorption and aeration. From productivity point of view the best results concerning total production per plant were obtained for the variant of soil plus 75% Perlite. This production is close to that obtained on unpolluted soil.

Until now, there has been little studies related to influence of substrates on different compounds of potatoes (*Solanum tuberosum*). In this context, in our study we followed the content of protein and sodium. We choose to analyze these compounds because sodium is in excess in polluted soil and protein is an important compound representing mainly enzymes.

At the harvested plants, we conducted analyzes of protein and sodium both in leaves and in tubers (fig. 4 and 5, tables 3, 4).

Protein content varies both in relation to substrate on which plants grew as well as between the leaves and tubers for the plants on the same crop. Leaf protein content is relatively high compared to the content in tubers. The content of protein in tubers is close to that found in Colette potatoes grown on unpolluted soil, namely an average of 2.08%. So, the variations in protein content in tubers are not very large (fig. 4, tables 3a, 3b) and it decrease in the order 2.4g/100g (V0), 2.2g/100g (V4), 2.1g/100g (V3), 1.9/100g (V2), 1.8g/100g (V1). In the leaves, protein concentrations were the highest for all crops, decreasing as follows: 4.2g/100 g (V4), 4.0g/100g (V0), 3.5g/100 g (V3), 2.5g/100 g (V2), and 2.2g/100 g (V1).

Tuber protein content obtained on the polluted soil was 72.97% lower than the one obtained from tubers obtained on unpolluted soil. From the statistic point of view to this variant (V1) a very significant negative difference was obtained. Through soil conditioners with 75% perlite the protein content was 89.19% of V0 variant (unpolluted soil). From the statistic point of view it can still noticed a very significant negative difference.

Regarding the protein content of the leaves, it can be appreciated that at improved polluted soil with 75% perlite,

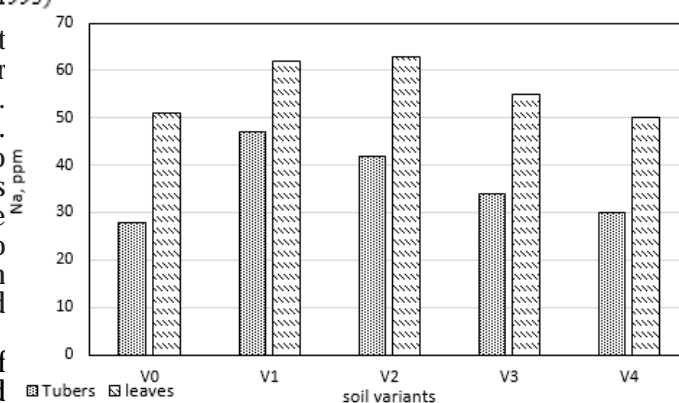


Fig. 5. Influence of culture substrates on potato leaves and tubers sodium concentrations

from a statistic point of view a significant increase compared to V0 (soil unpolluted soil) was obtained.

Relatively high concentrations of protein in the leaves could mean the presence of various enzymes in these tissues. So, further study on the influence of soil characteristics on various possibly to be present enzymes, it is necessary, especially because such studies are relatively few so far, especially by comparing the protein content according to different cultivation substrates. In addition, the highest protein content in leaves is normal taking into account the fact that in the leaves most metabolic processes take place.

The excess of sodium in soil as a result of oil pollution is a current issue. Consequently sodium contents in potato samples were evaluated in order to see whether or not, the nature of substrate influence the absorption and retention of sodium in tissues. Figure 5 and tables 4a, 4b presents the results obtained concerning the sodium concentrations found in leaves and tubers harvested from greenhouse grown potatoes on the 5 variants of soil.

Sodium content of tubers was the lowest at control variant of 28.00 ppm and highest in polluted soil of 47.07

Variant	Protein content, g/100g vegetal material	Difference		Signification
		g	%	
Average	2.09	-0.37	84.86	OOO
Unpolluted soil, V0	2.47	0.00	100.00	Mt
Polluted soil, V1	1.80	-0.67	72.97	OOO
Polluted soil + 25% perlite, V2	1.90	-0.57	77.03	OOO
Polluted soil + 50% perlite, V3	2.10	-0.37	85.14	OO
Polluted soil + 75% perlite, V4	2.20	-0.27	89.19	OO
DL5% = 0.170 DL5% in % = 6.8919 DL1% = 0.250 DL1% in % = 10.1351 DL01% = 0.370 DL01% in % = 15.0000				

Fteoretic = (001%= 14.72574); (0.1%= 7.089267); (5%= 3.861993)

Variant	Protein content, g/100g vegetal material	Difference		Signification
		g	%	
Average	3.28	-0.72	82.00	OOO
Unpolluted soil, V0	4.00	0.00	100.00	Mt
Polluted soil, V1	2.20	-1.80	55.00	OOO
Polluted soil + 25% perlite, V2	2.50	-1.50	62.50	OOO
Polluted soil + 50% perlite, V3	3.50	-0.50	87.50	OO
Polluted soil + 75% perlite, V4	4.20	-0.20	105.00	*
DL5% = 0.200 DL5% in % = 5.0000 DL1% = 0.290 DL1% in % = 7.2500 DL01% = 0.440 DL01% in % = 11.0000				

Fteoretic = (001% = 14.72574); (0.1% = 7.089267); (5% = 3.861993)

Variant	Na content, ppm	Difference		Signification
		ppm	%	
Average	36.21	8.21	129.33	***
Unpolluted soil, V0	28.00	0.00	100.00	Mt
Polluted soil, V1	47.07	19.07	168.10	***
Polluted soil + 25% perlite, V2	42.00	14.00	150.00	***
Polluted soil + 50% perlite, V3	34.00	6.00	121.43	***
Polluted soil + 75% perlite, V4	30.00	2.00	107.14	**
DL5% = 0.940 DL5% in % = 3.3571 DL1% = 1.360 DL1% in % = 4.8571 DL01% = 2.050 DL01% in % = 7.3214				

Fteoretic = (001% = 14.72574); (0.1% = 7.089267); (5% = 3.861993)

Variant	Na content, ppm	Difference		Signification
		ppm	%	
Average	56.20	5.20	110.20	***
Unpolluted soil, V0	51.00	0.00	100.00	Mt
Polluted soil, V1	62.00	11.00	121.57	***
Polluted soil + 25% perlite, V2	63.00	12.00	123.53	***
Polluted soil + 50% perlite, V3	55.00	4.00	107.84	***
Polluted soil + 75% perlite, V4	50.00	-1.00	98.04	OO
DL5% = 0.610 DL5% in % = 1.1961 DL1% = 0.890 DL1% in % = 1.7451 DL01% = 1.330 DL01% in % = 2.6078				

Fteoretic = (001% = 14.72574); (0.1% = 7.089267); (5% = 3.861993)

ppm, with 19.07 ppm more, increasing being by 68.10% above control. From statistic point of view for this variant the sodium content is significant very positive.

Through perlite polluted soil conditioners (V4) it was achieved a decrease in the content of Na in potato tubers at 30.00 ppm relative to V1 (polluted soil) and close to control V0. The difference was 7.14% higher than the variant V0 (control) and of statistic point of view it was very significant positive.

Through the analysis of content of Na in potato leaves, it was found that at oil polluted soil as well as at Perlite ameliorated variants with a percentage of 25% and 50%, a

high concentration of Na it was maintained. Differences being statistically significant very positive. Through soil conditioners with perlite in 75%, Na content of potato leaves dropped very significantly.

The content of sodium is about 1.68 times higher in tubers and 1.22 times higher in the leaves of potatoes grown in polluted soil compared to potatoes grown on unpolluted soil.

In the case of plants grown on the substrate that contained 75% perlite, sodium content both for tubers and leaves are virtually the same as that found in plants grown in unpolluted soil, for tubers 30 ppm towards 28 ppm and

**Table 3a**  
THE PROTEIN CONTENT IN POTATO TUBERS OBTAINED ON DIFFERENT SOIL VARIANTS

**Table 3b**  
THE PROTEIN CONTENT IN POTATO LEAVES OBTAINED ON DIFFERENT SOIL VARIANTS

**Table 4a**  
THE SODIUM CONTENT IN POTATO TUBERS OBTAINED ON DIFFERENT SOIL VARIANTS

**Table 4b**  
THE SODIUM CONTENT IN POTATO LEAVES OBTAINED ON DIFFERENT SOIL VARIANTS

in leaves 50 ppm towards 51 ppm. The difference of 1 ppm found in leaves could be considered as an error method.

It should be noted that the sodium light higher content in tubers, practically do not change their taste and properties.

## Conclusions

Crop productions and concentration of protein and sodium in their leaves and tubers largely depend on soil nature. In 2011 the agricultural lands in the Icoana village, Olt county, were severely polluted with oil on a surface of 3.5 ha.

The extent of oil pollution in the fall of 2014 was virtually zero but the soil has a salt concentration of about 3.8 times higher than unpolluted soil. An important aid in the process of phytoremediation using potatoes is adding to the polluted soil of Perlite. Due to its properties, namely, its high pore volume, with good water retention ability, and being chemically inert and totally environmentally friendly, perlite is an ideal substrate not only in soilless crops but to phytoremediation too. In addition, in Romania, due to its availability and the lowest price compared to other substrates Perlite is the most convenient substrate for use. However, given the diversity of existing perlite in trade, it is necessary to choose the best Perlite choice for a crop, by taking into account the behaviour of the plant that will be cultivated. In our experience we choose Perlite 4 mm because this gave the best results in soilless culture of some vegetables like tomatoes, potatoes, cucumbers and peppers.

Results of the current study showed a positive impact of salty soil mixed with Perlite 4mm in concentrations of 50 to 75% on growth of potatoes. So, as a simple and safe method, the inclusion of a specific quantity of Perlite in polluted soil before sowing of potatoes can be used to improvement plant growth and soil remediation. It appears that the utilization of Perlite can led at least to potato production and quality obtained on unpolluted soil. Generally, using Perlite treatment of polluted soil in phytoremediation processes and appropriate plants is an affordable and friendly way for regeneration of polluted areas.

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