^{32}P – RADIOACTIVE TRACER FOR THE EVALUATION OF FERTILIZERS INFLUENCE ON NUTRIENTS TRANSLOCATION PROCESS FROM SOIL TO THE PLANTS

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This paper presents a study over the influence of some fertilizers with amino acids and proteins on the absorption capacity of the phosphorous (^{32}P) present in the soil. In order to evaluate the ^{32}P quantity translocated from the soil into the plant under the influence of three types of fertilizers, we inoculated in the soil from the pots in which the sunflower plants were cultivated certain quantities of ^{32}P. After the inoculation, we applied the three fertilizer solutions in three successive foliar treatments (one solution/pot). The vegetal material was harvested, conditioned and the quantity of ^{32}P translocated from the soil to the air organs of the plants (leaves, stems and flowers) was measured. The results showed that the applied fertilizer solutions increased the absorption of ^{32}P from the soil, favoring the translocation of ^{32}P to the flowers. We observed that the ^{32}P translocation degree was different depending on the composition of the three fertilizer solutions.

Key words: radionuclidic labeling, extra-radicular fertilizers, agrochemical efficiency, radiochemical analytical methods.

1. INTRODUCTION

There are two categories of nutrients required for plant development: macronutrients – requested in relatively large quantities (carbon, hydrogen, oxygen, nitrogen, phosphorus, potassium, calcium, magnesium, sulphur) and micronutrients – requested in small quantities (iron, chlorine, manganese, boron, zinc, copper, molybdenum).

The first reports on foliar application of mineral nutrients dates in the second half of the XVIII^{-th} century. Many studies were performed on foliar fertilizers formulation, on mineral nutrients absorption through leaves and their translocation to other plant organs. The extremely low use of foliar fertilizers was due to the opinion of some specialists that considered these fertilizers less efficient than the classic ones. This opinion was based on foliar fertilizers composition that contained much smaller quantities of nutrients, in a time when only the quantity of major nutrients – nitrogen, phosphorus, and potassium – was considered of interest [1].

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After a time, including in the foliar fertilizers of micronutrients like iron, zinc and manganese with an essential role in plant respiration, activation of enzymes involved in metabolism, photosynthesis, chloroplasts formation, chlorophyll synthesis and natural hormones synthesis [2], led to solutions able to correct deficiencies, fortify weak or impaired crops, accelerate the growth process, facilitate the development of stronger plants.

Nowadays, the foliar fertilizers used together with the classic (solid) fertilizers, constitute a solution for rapid correction of nutrient unbalances and to stimulate the soil nutrients absorption by the plants roots.

The proven efficiency of the foliar fertilizers for numerous crops makes the researches in this domain very intense, aiming to include in the fertilizer solutions formulas, beside macro and micronutrients, some organic substances [3], proteins and protein extracts, amino acids [4], seaweed extracts [5], animal products extracts, microbial cultures [6] etc.

Nuclear techniques and radioisotopes as radioactive tracers were used in a large number of studies on fertilizers of interest for agriculture. The labelling with radioisotopes of different components of the fertilizer solutions, allowed the following up of the translocation routes and mechanisms for these components, from the fertilizer solution applied in the soil or on the leaves to all plant's organs. The labelling with radioisotopes such as ^{32}P, of fertilizers differing by composition, is also allowing the evaluation of fertilizers agrochemical efficiency [7].

2. EXPERIMENTAL

Phosphorous is one of the major nutrients necessary for plants growth and development. The existing in soil phosphorous quantity and it's availability for the plants is an important factor for the crops quality and quantity. Providing of the optimal phosphorous quantity can be achieved by fertilization, applying either solid fertilizers to the soil or solutions of foliar fertilizers on the plants. Beside the phosphorous delivered directly to the plants, foliar fertilizers, depending on their composition, can increase the absorption of the phosphorous from the soil by the plants roots.

In the present study, we used ^{32}P as radioactive tracer to follow the absorption of phosphorous from the soil into sunflower plants under the influence of three types of foliar fertilizer solutions.

^{32}P is an adequate radionuclide for use in plant physiology and fertilization studies due to its nuclear features: pure beta emitter with the maximum β^- radiation energy E_{max}=1.7 MeV and a half-life of 14.3 days.

The decay scheme of ^{32}P is presented in Figure 1.

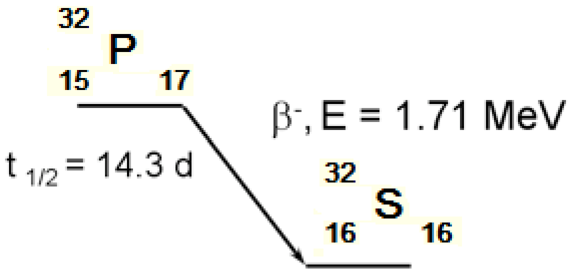

Fig. 1 – 32P Decay scheme.

For the experiments performed in the present study, we used 4 Mitscherlich type pots, each containing 20 kg of dry soil. In each pot were planted 3 sunflower plants.

The three types of fertilizers used in the pots are presented in Table 1, together with the pH value of the fertilizer solutions.

Table 1

The fertilizer solutions used in each pot; denomination and the pH value of the solutions

Pot No.	Denomination of the fertilizer solution	pH value
Pot # 1	WATER	Control
Pot # 2	IZO	6.0-6.2
Pot # 3	AMINO FERT 2 H	8.5
Pot # 4	AMINO FERT 1 H	8.5

The fertilizer solutions had an average phosphorous content of 19 x 10^{-5} g P/mL. All fertilizer solutions used were prepared at ICPA [8–10], with a similar content of microelements, but with different chemical forms of nitrogen. The solutions of AMINO FERT 1 H and AMINO FERT 2 H were supplemented with 1 mL respectively 2 mL collagen hydrolyzed at 100 mL fertilizer solution.

Our study included two steps: applying of a radioactive solution containing ^{32}P in the soil from every pot and, after three days, applying of the fertilizer solutions (by spraying) all over the plants.

For the preparation of the ^{32}P radioactive solution for soil application, we used a stock solution of ^{32}P-orthophosphate, carrier free, in diluted HCl, pH: 2-3, with the radioactive concentration of 370 MBq/mL (IZOTOP, Hungary). For every pot, the solution for soil application contained 200 mL water and 125 µL of ^{32}P-orthophosphate stock solution. In order to determine the radioactivity of the solution thus prepared, a sample of 100 µL was measured both by liquid scintillation method and with a gas flow proportional counter connected to an alpha/beta portable monitor (developed in IFIN-HH) [11]. The results obtained were: 2009 Bq/100 µL for the liquid scintillation method and 463 counts/s/100 µL with the proportional counter leading to a conversion factor of 4.34 between the activities measured in Bq and the ones measured in counts/s.

The radioactive solution applied in every pot had the activity of 4018 kBq and pH: 6.5–6.7.

Three days after the application of radioactive solutions to the soil started the second stage of the experiment consisting of treatments with foliar fertilizers solutions. There were performed three successive treatments at three days from each other. For every treatment, 50mL of fertilizer solution were sprayed over the plants from each pot. The total volume of fertilizer solution applied on the plants from every pot was of 150 mL. The plants from pot # 1, considered the control plants, were sprayed with the same volume of water instead of fertilizer solution. So, in the second stage of the experiment, a total quantity of 2850 x 10^{-5} g non-radioactive phosphorous was applied on the plants from every pot (except pot # 1).

One day after the last foliar fertilizers application, all plants were harvested separating the plants air organs (flower, stem, leaves). For each plant, were determined the mass (with an analytical balance) and activity of every plant organ. The activity measurements were performed using the same gas flow proportional counter, the determined values, counts/second, being lately converted to Bq, using the converting factor of 4.34, determined earlier. The reference moment chosen for all activity results reporting was the harvesting date.

3. RESULTS AND DISCUSSIONS

Each vegetal sample received a coded name of "aXY" type, meaning: a – pot number (1-4), X – the plant from the pot (A-C) and Y – the plant organ (I – inflorescence, T – stem, F – leaves). In the following tables (2.1. – 2.4.), are presented the plants mass, the translocated ^{32}P activity and the specific activity of translocated ^{32}P from the soil to the plants in the pots 1 – 4.

Table 2.1

Pot # 1/ Spraying solution: Water (control plants)

Sample code	Mass (g)	Translocated ^{32}P activity (Bq)	Specific activity of translocated ^{32}P (Bq/g)
1AI	53.164	59937	1127.398
1AT	113.231	67397	595.216
1AF	83.738	67991	811.949
	250.134	**195325**	**780.881**
1BI	39.301	70032	1781.939
1BT	110.209	81838	742.571
1BF	74.725	108049	1445.955
	224.235	**259919**	**1159.137**
1CI	51.918	80022	1541.315
1CT	115.041	86363	750.715
1CF	80.661	83588	1036.930
	247.570	**249973**	**1009.706**
Total	**721.939**	**705217**	**976.837**

Table 2.2

Pot # 2/ Spraying solution: IZO (fertilizer solution)

Sample code	Mass (g)	Translocated ^{32}P activity (Bq)	Specific activity of translocated ^{32}P (Bq/g)
2AI	38.419	90850	2364.715
2AT	99.127	91280	920.839
2AF	72.600	82983	1143.017
	210.146	**265113**	**1261.566**
2BI	43.300	110025	2540.993
2BT	109.858	118306	1076.899
2BF	74.316	122348	1646.321
	227.474	**350679**	**1541.622**
2CI	45.549	89434	1963.468
2CT	121.933	116940	959.051
2CF	81.609	153015	1874.977
	249.091	**359389**	**1442.802**
Total	**686.711**	**975181**	**1420.075**

Table 2.3

Pot # 2/ Spraying solution: AMINO FERT 2H (fertilizer solution)

Sample code	Mass (g)	Translocated ^{32}P activity (Bq)	Specific activity of translocated ^{32}P (Bq/g)
3AI	53.468	27383	512.138
3AT	111.641	51325	459.733
3AF	78.310	81105	1035.691
	243.419	**159813**	**656.535**
3BI	33.749	68278	2023.112
3BT	81.965	119955	1463.491
3BF	51.880	120042	2313.840
	167.594	**308275**	**1839.415**
3CI	38.092	140965	3700.646
3CT	121.653	234463	1927.310
3CF	85.342	215014	2519.439
	245.087	**590442**	**2409.112**
Total	**656.100**	**1058530**	**1613.367**

Table 2.4

Pot # 2/ Spraying solution: AMINO FERT 1H (fertilizer solution)

Sample code	Mass (g)	Translocated ^{32}P activity (Bq)	Specific activity of translocated ^{32}P (Bq/g)
4AI	43.434	207230	4771.147
4AT	143.504	160126	1115.830
4AF	75.061	180621	2406.323
	261.999	**547977**	**2091.523**
4BI	46.516	115646	2486.155
4BT	132.517	147257	1111.231

Table 2.4 (continued)

4BF	73.189	202498	2766.782
	252.222	**465401**	**1845.204**
4CI	23.890	42439	1776.434
4CT	67.494	43705	647.539
4CF	57.990	77531	1336.972
	149.374	**163675**	**1095.740**
Total	**663.595**	**1177053**	**1773.752**

The results show that fertilizers increase the ^{32}P translocation mainly in the inflorescence, followed by the leaves and stem, excepting plants 3A, 3B and 4B where the ^{32}P translocation is slightly higher in the leafs *versus* the inflorescences.

The variation of plants masses in every pot is presented in Figure 2.

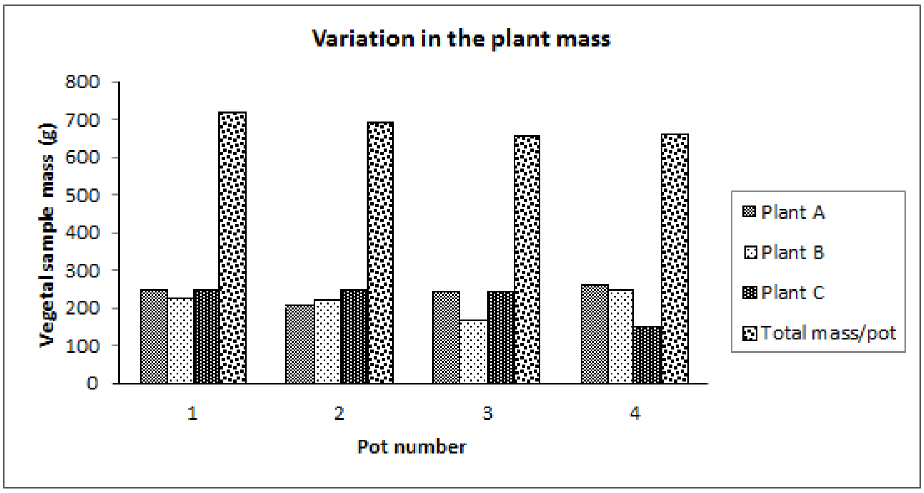

Fig. 2 – Variation of plants masses in every pot.

The higher vegetal material mass was obtained in the pot # 1, followed by pot # 2, pot #3 and pot #4.

The specific activity of ^{32}P translocated from the soil in the plants depends on the plant mass. Thus, the specific activity will decrease with the increasing of the plant mass. In order to compare the efficiency of the three types of fertilizer solutions used was necessary to apply a “mass correction factor” to the obtained results. This correction factor was calculated as a mass ratio between every plant mass and the mass of the smallest plant (149.375 g, plant C in pot #4). For the total plant mass of each pot, the correction factor was calculated as the mass ratio between the total plant mass of a pot and the smallest total plant mass (656.100 g, obtained in the pot #3). The correction factors calculated for each plant, for the total plant mass in each pot and the corrected specific activity of translocated ^{32}P are presented in Table 3.

Table 3

Correction factors and the corrected specific activity of translocated ^{32}P

Pot and Plant	Correction factor	Corrected specific activity of translocated ^{32}P (Bq/g)
1A	1.675	1307.975
1B	1.501	1739.864
1C	1.658	1674.093
Pot # 1	**1.100**	**1074.521**
2A	1.407	1775.023
2B	1.523	2347.891
2C	1.668	2406.594
Pot # 2	**1.047**	**1486.818**
3A	1.630	1070.151
3B	1.122	2063.824
3C	1.641	3953.353
Pot # 3	**1.000**	**1613.367**
4A	1.754	3668.532
4B	1.689	3116.549
4C	1.000	1095.740
Pot # 4	**1.011**	**1793.263**

The highest specific activity of ^{32}P translocated from the soil into the plants was observed in pot #4, followed by pot #3, pot #2 and pot #1. The specific activity (corrected with the mass correction factors) of ^{32}P translocated from the soil into the plants from the four pots is presented in Figure 3.

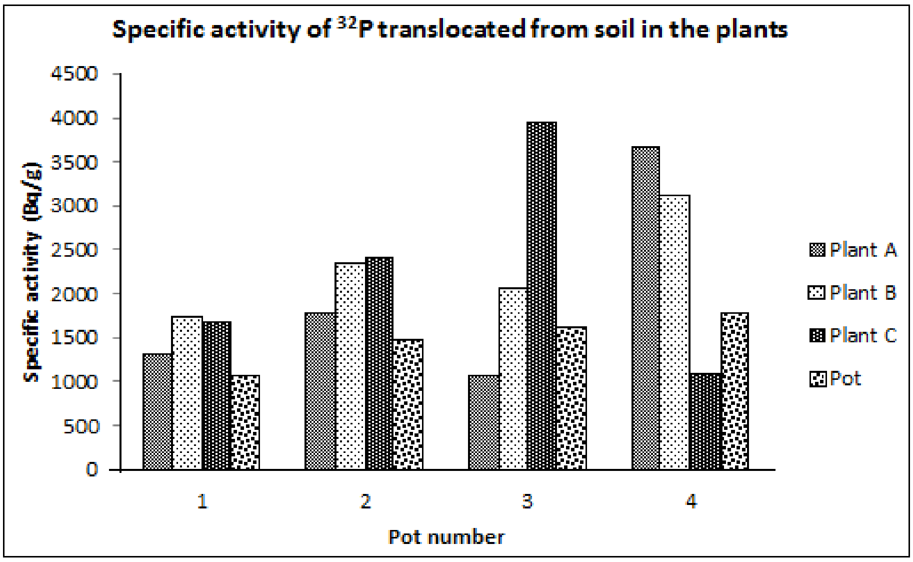

Fig. 3 – Specific activity (corrected with the mass correction factors) of ^{32}P translocated from the soil into the plants.

The percentage of the ^{32}P applied in every pot (4018 kBq) translocated into the plants depends on the fertilizer solution composition (Table 4). Thus, the higher translocation percentage was obtained in the pot #4, followed by pot #3, pot #2 and pot #1.

Table 4

Translocated ^{32}P dependence on the chemical composition of the fertilizer solution

Pot number / Fertilizer	^{32}P activity applied in the soil (Bq)	^{32}P activity translocated into the plants (Bq)	% ^{32}P translocated into the plants
Pot # 1 /Water	4018000	705217	17.551 %
Pot # 2 / IZO	4018000	975181	24.270 %
Pot # 3 / Amino Fert 2 H	4018000	1058530	26.345 %
Pot #4 / Amino Fert 1 H	4018000	1177053	29.294 %

4. CONCLUSION

The use of foliar fertilizers favored the translocation of ^{32}P mainly in the inflorescence.

The higher efficiency in ^{32}P translocation from the soil into the plants was observed for AMINO FERT 1H fertilizer, followed by AMINO FERT 2H fertilizer and IZO fertilizer.

Using of the ^{32}P radioisotope allowed the monitoring of the phosphorous translocation from the soil into the plants and the influence of the three types foliar fertilizers on the translocation process. The obtained results demonstrated that the use of foliar fertilizers significantly increased the phosphorous translocation from the soil into the plants.

The composition of the foliar fertilizers also influenced the percentage of translocated ^{32}P. Thus, the presence of collagen hydrolyzed led to the best ^{32}P translocation percentages (29.294% for AMINO FERT 1 H and 26.345% for AMINO FERT 2 H fertilizer solution). The maximum ^{32}P translocation percentage was obtained for AMINO FERT 1 H containing 1% collagen hydrolyzed.

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