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EVALUATION OF NITROGEN FERTILIZATION EFFECT ON THE CONTENT AND ACCUMULATION OF PHOSPHORUS IN YELLOW LUPINE IN SOIL CONDITIONS WITH VERY HIGH AVAILABLE FORMS OF THIS ELEMENT

Andrzej Wysokiński✉, Beata Kuziemska, Monika Kożuchowska

Department of Soil Science and Agricultural Chemistry, Siedlce University of Natural Sciences and Humanities
B. Prusa 14, 08-110 Siedlce, **Poland**

ABSTRACT

Background. In view of the possibility to ensure the crop's demand for phosphorus from soils with a high content of available forms of this element, as well as to increase the amount of mobile forms of phosphorus after the effect of nitrogen fertilization, a study was undertaken on the effect of different of nitrogen doses applied into soil on the phosphorus content and accumulation in yellow lupine roots, stems, leaves, flowers, pods and seeds.

Material and methods. In two-years field experiment the influence of development stages (BBCH 65 and BBCH 90) at which harvest was performed as well as nitrogen doses (0, 30, and 120 kg·ha^{-1}) introduced into the soil prior to sowing were the factors under study.The experiment was set up on slightly acid soil with very high concentration of available phosphorus content for plant.

Results. A 10.7% higher average content of phosphorus was obtained in the whole mass of lupine harvested during the full maturity stage than during the flowering stage. The phosphorus content in the seeds was more than four times higher than in the roots and stems, more than three times higher in the pods and more than two times higher than in the leaves. Differentiated nitrogen fertilization in the form of mineral fertilizer had no significant effect on the phosphorus content of the seeds or, on average, in the whole mass of lupine. Lupine fertilized with 120 kg·ha^{-1} N took up more phosphorus in total than it did without nitrogen fertilization and following the application of 30 kg·ha^{-1} N. The amount of phosphorus accumulated in the biomass harvested at the full maturity stage was two times higher than that in the flowering stage.

Conclusion. Irrespective of the applied nitrogen fertilization and non-use of phosphorus fertilization, soil with a high content of P in forms available for plants, ensured appropriate supply of yellow lupine to phosphorus, which is important in the protection of non-renewable resources of phosphorites.

Key words: growth stage, *Lupinus luteus* L., nitrogen doses, phosphorus uptake, soil richness

INTRODUCTION

Most frequently, the sources of phosphorus in cultivable soils are fertilizers, plant residues and wastes from agricultural and industrial production. Under conditions of intensive agricultural productions, phosphorus fertilizers produced from non-renewable resources of phosphorites are the main source of phosphorus for plants (Sapek, 2007; Balemil and Negisho, 2012; Van Kawenbergh *et al*., 2013; Sapek,

✉ andrzej.wysokinski@uph.edu.pl, beata.kuziemska@uph.edu.pl, monika.kozuchowska@uph.edu.pl

2014; Wolski *et al*., 2017). Phosphorus accumulated in the soil, depending on the soil conditions and on the form in which it is found, can be transformed into forms that can be taken up by plants but also leached downward in the soil profile, or subjected to surface and subsurface run-off to soil water and open water bodies (Djodjic *et al*., 2004; Vadas *et al*., 2005). Leaching phosphorus is less than other basic macronutrients (Pondel *et al*., 1991; Wierzbowska *et al*., 2016). The mechanisms of transformations leading to the release and leaching of phosphorus from the soil are complex and determined by many factors (Sapek, 2014).The most important factors are the pH of soils, organic matter content and nitrogen fertilization (Li *et al*., 2009; Sapek, 2010; Tate and Salcedo, 1988). Nitrogen introduced into a soil contributes to the formation of a soluble form of phosphorus which penetrates into the deeper layers of the soil profile and poses the threat of enriching groundwater with this component (Sapek and Wolicka, 2007; Wesołowski and Durkowski, 2004). The introduction of mineral nitrogen into soils can contribute to the intensification of the organic matter mineralization process and lead to an increase content of mobile forms of phosphorus in the soil. The need for reasonable phosphorus management in the environment, taking account of prudent exploitation of non-renewable resources and skilful use of soil reserves of this element, was the cause of undertaking a study into quantitative determination of phosphorus uptake from soil very rich in this nutrient by yellow lupine.

The aim of the study was to determine the effect of various doses of nitrogen fertilisation and of the development stage on the content and accumulation of phosphorus in particular organs of yellow lupine (*Lupinus luteus* L.) cultivated on soil with very high contents of plant-available phosphorus forms.

MATERIAL AND METHODS

The field experiment was conducted in Siedlce (52°10' N; 22°17' E) in 2008 – 1st year and 2011 – 2nd year. The experiment as conducted on slightly acidic soil with granulometric composition of loamy sand (by PTG). The content of selected macro- and micronutrients in the soil before the experiment was set up is shown in Table 1.

Table 1. Selected properties of soil in humus layer prior on which the field experiments were carried out

Soils properties	Unit	Years of foundation experiment	
		2008	2011
pH_{KCl}	–	5.90	5.80
C_{tot}	$g \cdot kg^{-1}$	25.7	23.8
N_{tot}		2.04	1.92
P_{tot}		1.10	1.15
K_{tot}		0.85	0.81
Mg_{tot}		0.96	0.93
S_{tot}		0.448	0.56
P_{av}	$mg \cdot kg^{-1}$	369.0	314.0
K_{av}		67.0	59.0
Mo_{tot}		0.12	0.09
Mn_{tot}		155.2	157.9
Cu_{tot}		20.3	18.0
Fe_{tot}		5243	5135
Zn_{tot}		219.1	176.0

P_{av}, K_{av} – available forms for plants
X_{tot} – total content

1 m^2 plots were delineated in a field of yellow lupine of the 'Mister' cultivar. Two-factorial experiment was set up in the randomised split-block design, in three replications. Nitrogen fertilisation was the first factor: a) control, with no nitrogen fertilisation; b) with nitrogen applied at a rate equivalent to 30 kg $N \cdot ha^{-1}$; c) with nitrogen applied at a rate equivalent to 120 kg $N \cdot ha^{-1}$. The time of harvest was the second factor (determined as per Bleinholder *et al*., 2001): a) full flowering stage, 65 BBCH (date I, marked A in Table 3–6); b) full maturity stage, 90 BBCH (date II, marked B in table 3–6). Mineral nitrogen was introduced to the soil as ammonium sulphate $(NH_4)_2SO_4$ before yellow lupine was sown. The amounts of phosphorus and potassium were established on the basis of the amounts of the available element forms in soil. Potassium was introduced to the soil in all plots at 100 kg $K \cdot ha^{-1}$ as potassium salt. Because of a very high amount of phosphorus as available forms

(Table 1), no phosphorus fertilisation was applied. Before sowing, seeds of yellow lupine were inoculated with a vaccine containing *Rhizobium lupini*. Sowing was performed in early April at 100 germinating seeds per 1 m^2. Soil was sprayed with the herbicide Stomp 330 EC at a rate of 4 $dm^3 \cdot ha^{-1}$ on the day following the sowing of lupine. Lupine plants was sprayed with Amistar 250 SC at 1.0 $dm^3 \cdot ha^{-1}$ against anthracnose at the beginning of the budding phase; this procedure was repeated after 10 days. Plants harvested manually during the flowering stage were divided into roots, steams, leaves and flowers, whereas those harvested during the full maturity stage were divided into roots, steams, leaves, pods and seeds.

The content of phosphorus in the plant material was determined by the ICP-AES method in the bulk solution obtained by mineralisation of samples at 450°C. The ash obtained by mineralisation was dissolved in HCl 6 $mol \cdot dm^{-3}$ in order to degrade carbonates and evaporated to dryness on a sand bath. A 10% solution of HCl was used to transfer chlorides to volumetric flasks (Krzywy-Gawrońska, 2007).

The results were worked out statistically with an analysis of variance. Conclusions regarding the significance of an effect of the factors under study on individual features were based on the Fisher-Snedecor F-test, and the $HSD_{0.05}$ for comparison of the calculated means were calculated by the Tukey test. Moreover, linear correlation coefficients for content, uptake of phosphorus and amount and percentage of nitrogen taken up by yellow lupine from the atmosphere were (published by Wysokiński, 2013) calculated. To these calculations the Statistica 12 PL software package (StatSoft, Tulsa, USA) and MS Excel was used.

The total rainfall in individual months and mean monthly air temperature during the growing season for yellow lupines shown in Table 2. It shows that both growing seasons were rather favourable for the growth, development and yielding of yellow lupine. The total rainfall during the 2008 and 2011 growing seasons satisfied the plants needs in full. However, it was not properly distributed over the months of growing. The amount of rainfall in June 2008 and in May and June 2011 was lower than required for yellow lupine, as reported by Dzieżyc *et al*. (1987). In addition to the greater water deficit during the period of intensive growth of lupine (May-June) in 2011, higher temperatures were recorded during the period than in 2008, which probably exacerbated the water deficit and decreased the yield of tested plant, presented in publication of Wysokiński (2013).

Table 2. Rainfall and air temperatures during the test crop (data from Hydro-Meteorological Station in Siedlce IMGW PIB in Warsaw)

Weather parameter	Month	Study period		Multiyear (1981-2007)
		2008	2011	
Monthly rainfall, mm	April	43.5	38.1	32.9
	May	72.7	55.6	54.2
	June	56.7	44.3	68.8
	July	108.8	204.2	64.9
	August	85.1	55.4	61.8
Averages monthly temperatures, °C	April	8.7	9.8	7.9
	May	12.5	13.5	13.7
	June	17.0	18.1	16.1
	July	18.1	18.1	18.3
	August	18.3	18.1	17.6

RESULTS AND DISCUSSION

Phosphorus content of the whole mass of yellow lupine ranged from 3.23 to 4.51 g·kg^{-1} P in DM (Table 3). The average content of this element found in the whole mass of lupine harvested during the full maturity stage was higher by 10.7% compared to that harvested during the flowering stage (Table 4). This dependency was due to the highest concentration of phosphorus in the seeds as compared to other organs of the test plant. The average phosphorus contents of particular organs of lupine harvested during the flowering stage can be presented in the following series in descending order: the flowers, roots, stems and leaves, while for that harvested during the stage of full maturity: the seeds, leaves, pods, roots and stems. The content of this macroelement in the roots and stems was nearly two times higher when lupine was harvested during the flowering stage as compared to the harvest following the reaching of full maturity. Phosphorus concentration in the leaves was similar during both development stages. Differentiated nitrogen fertilization had no significant effect on the phosphorus content of the roots, leaves, flowers, pods or seeds or (on average) in the entire biomass of yellow lupine (Table 4). Following the application of 30 kg·ha^{-1} N, the content of this macronutrient was only significantly lower in the stems than following the introduction of 120 kg·ha^{-1} N and without nitrogen fertilisation.

Table 3. Phosphorus content in yellow lupine, g·kg^{-1} P in dry mass (DM)

Year	Growth stage	Nitrogen dose kg·ha^{-1} N	Part of plant					Meanly in plant
			roots	stems	leafs	flowers/ stripped pods[1]	seeds	
1st	A	0	4.75	4.54	3.64	5.35	–	4.21
		30	3.55	3.39	2.82	4.87	–	3.23
		120	3.81	3.72	3.13	4.89	–	3.51
	averages		4.04	3.88	3.20	5.04	–	3.65
	B	0	1.90	1.75	3.02	2.77	8.86	3.79
		30	1.88	1.38	2.86	2.58	8.08	3.66
		120	2.15	1.95	3.68	2.69	8.76	4.42
	averages		1.98	1.70	3.19	2.68	8.56	3.96
Averages			3.01	2.79	3.19	–	–	3.80
2nd	A	0	4.60	4.48	2.72	5.18	–	3.76
		30	4.89	4.11	3.08	6.20	–	3.89
		120	4.52	4.23	3.02	5.53	–	3.82
	averages		4.67	4.27	2.94	5.63	–	3.82
	B	0	1.83	2.33	3.89	1.89	8.67	4.19
		30	2.12	1.80	3.38	3.51	8.90	4.26
		120	1.90	2.21	3.48	2.40	9.75	4.51
	averages		1.95	2.11	3.58	2.60	9.11	4.32
Averages			3.31	3.19	3.26	–	–	4.07
$HSD_{0.05}$ for year			0.28	0.24	ns	0.39/ns	ns	ns

[1] in dependence from yellow lupine's growth stage: for blooming stage (A) the tower concerns the flower, ut for full maturity stage (B) concerns the stripped pods
ns – non-significant

Table 4. The averages for investigated factors of phosphorus contents in yellow lupine, g·kg^{-1} P in DM

Investigated factor		Part of plant						Meanly in plant
		roots	stems	leafs	flowers	stripped pods[1]	seeds	
Nitrogen dose kg·ha^{-1} N	0	3.27	3.28	3.32	5.27	2.33	8.77	3.99
	30	3.11	2.67	3.04	5.54	3.05	8.49	3.76
	120	3.10	3.03	3.33	5.21	2.55	9.26	4.07
$HSD_{0.05}$		ns	0.35	ns	ns	ns	ns	ns
Growth stage	A	4.35	4.08	3.07	5.34	–	–	3.74
	B	1.96	1.90	3.39	–	2.64	8.84	4.14
$HSD_{0.05}$		0.28	0.24	n.s.	–	–	–	0.31
$HSD_{0.05}$ for interaction								
Growth stage/year		0.40	ns	ns	–	–	–	ns
Nitrogen dose/year		0.59	ns	ns	0.84	ns	ns	ns
Nitrogen dose/growth stage		ns	ns	ns	–	–	–	ns

[1] in dependence from yellow lupine's growth stage: for blooming stage (A) the tower concerns the flower, ut for full maturity stage (B) concerns the stripped pods
ns – non-significant

Phosphorus content of the roots, stems and flowers of yellow lupine was lower in 2008, which was characterised by a lower temperature during the vegetation period as compared to 2011 (Table 3). Ercoli et al. (1996) in studies on Sorghum proved that at too low or too high temperature caused the content of phosphorus in individual organs of this plant is lower than at the optimum temperature (27°C). During both years of the study, no significant differences were obtained for the content of the tested macronutrient in the leaves, pods, seeds or (on average) in the entire biomass of the tested species.

The total amount of phosphorus taken up by lupine, and the amounts of this element accumulated in the roots, stems and seeds, were not significantly different depending on the year of the study (Table 5). In the leaves and pods, a lower amount of phosphorus was accumulated by lupine in 2011, which was characterised by a higher temperature during the growing season, despite the less favourable distribution of precipitation than in 2008.

The amount of phosphorus taken up by the entire yellow lupine plant harvested during the full maturity stage was over two times higher than during the flowering stage (Table 6). During the full maturity stage, the amount of phosphorus accumulated in the roots and stems was lower than that during the flowering stage, while for the leaves, the dependencies provided were reverse. The test plant harvested at the full maturity stage accumulated approx. half of the total amount of taken up phosphorus in the seeds. Lupine fertilized with 120 kg Na·ha^{-1} accumulated the largest amount of phosphorus in the stems, leaves, seeds and, in total, in the entire biomass. The amounts of phosphorus accumulated in particular organs, and the total amounts in the entire mass of lupine, were similar in general following fertilisation with 30 kg N·ha^{-1} and in the control object. The calculated linear correlation coefficients indicate that the amount of phosphorus taken up by lupine was especially dependent on the amount of the harvested mass provided in a study by Wysokiński (2013), r = +0.978 and to a lesser extent on the average content of this element in its mass, r = +0.638 (Table 7).

Phosphorus deficiency exerts a great impact on white lupine metabolism, leading to a wide range of

metabolic and physiological adjustments. Protein anabolism is reduced under P-deficiency, but the formation of secondary metabolites is enhanced (Müller *et al*., 2015). Lupine utilises soil phosphorus from a normally non-labile pool of soil P that is not utilised by soybean (Braum and Helmke, 1995). In order to prevent a reduction in plant mass production following phosphorus deficiency, it is necessary to ensure the proper availability of this element to plants. In the author's own study conducted under conditions of a very high content of phosphorus forms being available to plants, with the fertilisation of this macronutrient omitted, irrespective of the applied nitrogen fertilisation, the level of phosphorus content of yellow lupine biomass was similar to its content obtained in the conditions of fertilization with this macroelement (Szostek and Ciećko, 2015; Jarecki *et al*., 2017).The obtained results confirm the thesis about the possibility of abandoning phosphorus fertilization of plants cultivated in conditions with very high content available forms of this element in the soil.

Table 5. The amount of phosphorus taken up by yellow lupine, kg·ha^{-1} P

Year	Growth stage	Nitrogen dose kg·ha^{-1} N	Part of plant					Sum
			roots	stems	leafs	flowers/ stripped pods[1]	seeds	
1st	A	0	2.58	4.10	4.57	0.57	–	11.83
		30	2.10	3.84	4.12	0.63	–	10.71
		120	2.40	4.79	5.16	0.55	–	12.92
	averages		2.36	4.24	4.62	0.58	–	11.82
	B	0	0.59	2.16	5.94	3.04	10.05	21.77
		30	0.71	1.84	5.51	3.42	12.17	23.67
		120	08.3	3.06	8.10	4.10	18.17	34.26
	averages		0.71	2.35	6.52	3.52	13.46	26.57
Averages			1.53	3.30	5.57	–	–	19.19
2nd	A	0	2.36	4.12	3.30	0.63	–	10.41
		30	2.60	3.89	3.94	0.80	–	11.22
		120	2.50	4.26	3.91	0.72	–	11.41
	averages		2.49	4.09	3.72	0.72	–	11.01
	B	0	0.79	2.95	6.00	1.77	11.64	23.12
		30	0.85	2.27	5.29	3.60	11.68	23.71
		120	0.68	2.88	5.63	2.38	14.30	25.88
	averages		0.77	2.70	5.64	2.59	12.54	24.24
Averages			1.63	3.40	4.68	–	–	17.63
$HSD_{0.05}$ for year			ns	ns	0.59	0.10/0.52	ns	ns

[1] in dependence from yellow lupine's growth stage: for blooming stage (A) the tower concerns the flower, ut for full maturity stage (B) concerns the stripped pods
ns – non-significant

Table 6. The averages of phosphorus amount taken up by yellow lupine, kg·ha^{-1} P

Investigated factor		Part of plant						Sum
		roots	stems	leafs	flowers	stripped pods	seeds	
Nitrogen dose kg·ha^{-1} N	0	1.58	3.33	4.95	0.60	2.41	10.85	16.78
	30	1.56	2.96	4.72	0.72	3.51	11.92	17.33
	120	1.60	3.75	5.70	0.64	3.24	16.24	21.12
$HSD_{0.05}$		ns	0.78	0.88	ns	0.78	3.07	3.49
Growth stage	A	2.42	4.17	4.17	0.65	–	–	11.42
	B	0.74	2.53	6.08	–	3.05	13.00	25.40
$HSD_{0.05}$		0.23	0.53	0.59	–	–	–	2.36
$HSD_{0.05}$ for interaction								
Growth stage/year		ns	ns	ns	–	–	–	ns
Nitrogen dose/year		ns	ns	ns	ns	1.10	ns	ns
Nitrogen dose/growth stage		ns	ns	ns	–	–	–	ns

[1] in dependence from yellow lupine's growth stage: for blooming stage (A) the tower concerns the flower, ut for full maturity stage (B) concerns the stripped pods
ns – non-significant

Table 7. The values of correlation coefficient between the content, uptake of phosphorus and total yield of yellow lupine biomass, $P < 0.05$

Investigated parameters	Content of P (meanly in plant)	Total uptake of P
Content of P (meanly in plant)	–	0.638*
Total mass of lupine	0.473	0.978*

* the value of correlation coefficient is important, $P < 0.05$

Koper and Lemanowicz (2007) demonstrated an increase in the phosphorus content of red clover only for doses of up to 60 kg N·ha^{-1}. After the application of a higher dose of nitrogen, phosphorus content decreased. Knapowski *et al.* (2001) in studies on wheat and Bąk *et al.* (2016) in studies on maize noted that the application of high nitrogen doses decreased the plant phosphorus content. The author's own study did not confirm the above observations, as different doses of nitrogen had no effect on the average phosphorus content of the whole mass of yellow lupine. The obtained results and the data provided in the literature (Ciećko and Wyszkowski, 1996) indicate that increased nitrogen fertilisation increases the amount of phosphorus taken up by crops. The correlation coefficients calculated show that it is mainly related to the greater amount of obtained mass of plants at higher doses of nitrogen introduced into the soi.

CONCLUSIONS

Averages content of phosphorus in whole yellow lupine's biomass was not significantly determined by the applied various nitrogen fertilisation. More phosphorus was accumulated in whole yellow lupine's biomass after 120 kg·ha^{-1} N application than non nitrogen fertilisation and 30 kg·ha^{-1} N application. Yellow lupine harvested at the full maturity stage contained and took up more phosphorus than at the flowering stage. The highest concentration and the largest amount of phosphorus was accumulated in the lupine's seeds (about half of total amounts taken up by whole plant). The averages concentration of phosphorus in whole biomass of lupine and the amount of this element taken up by whole plant were not significantly differentiated in both years of study. In the cultivation of yellow lupine in soil conditions with a high content of available phosphorus, fertilization with this macronutrient can be omitted.

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OCENA WPŁYWU NAWOŻENIA AZOTEM NA ZAWARTOŚĆ I AKUMULACJĘ FOSFORU W ŁUBINIE ŻÓŁTYM W WARUNKACH GLEBY O BARDZO WYSOKIEJ ZAWARTOŚCI PRZYSWAJALNYCH FORM TEGO PIERWIASTKA

Streszczenie

W doświadczeniu polowym określono zawartość oraz ilość zakumulowanego fosforu w korzeniach, łodygach, liściach, kwiatach, strączynach i nasionach łubinu żółtego. Badanymi czynnikami były faza rozwojowa (65 BBCH i 90 BBCH), w której nastąpił zbiór oraz ilość azotu (0, 30 i 120 kg·ha^{-1} N) wprowadzona do gleby przedsiewnie. Większą o 10,7% średnią zawartość fosforu uzyskano w całej masie łubinu zbieranego w fazie pełnej dojrzałości niż w fazie kwitnienia. Zawartość tego pierwiastka w nasionach była ponad czterokrotnie większa niż w korzeniach i łodygach, ponad trzykrotnie większa niż w strączynach oraz ponad dwukrotnie większa niż w liściach. Zróżnicowane nawożenie azotem w formie nawozu mineralnego nie miało istotnego wpływu na zawartość fosforu w nasionach i średnio w całej masie łubinu. Łubin nawożony 120 kg·ha^{-1} N pobrał ogółem więcej fosforu niż bez nawożenia azotem i po zastosowaniu 30 kg·ha^{-1} N. Ilość fosforu zgromadzonego w biomasie zbieranej w fazie pełnej dojrzałości była ponad dwukrotnie większa niż w fazie kwitnienia. Niezależnie od zastosowanego nawożenia azotem gleba o wysokiej zawartości fosforu w formach dostępnych dla roślin zapewniła odpowiednie zaopatrzenie łubinu żółtego w ten makroelement.

Słowa kluczowe: dawki azotu, faza rozwojowa, *Lupinus luteus* L., pobranie fosforu, zasobność gleby