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Effects of fertilisers on pulse crop productivity and nitrogen assimilation on acid soil

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Abstract: This research aimed to compare the productivity of three species of pulse plants (*Vicia faba* L., *Pisum sativum* L., *Lupinus angustifolium* L.) in acidic sandy loam soil and to determine the influence of NPK (N0P0K0, N0P27K73, N30P27K73) fertilisers on grain yield and nitrogen assimilation. The experiments were conducted Voke Branch of Lithuanian Research Centre for Agriculture and Forestry (Baltic Sea region, 54°33'49.8"N, 25°05'12.9"E) in 2016–2018. Fertilisation with PK increased the grain yield of *Vicia faba* L. and *Pisum sativum* L. ($P < 0.05$) and had no effect on the yield of *Lupinus angustifolium* L. ($P > 0.05$). Nitrogen fertilisers only increased the yield of *Pisum sativum* L. grain, reduced the yield of *Lupinus angustifolium* L. grain and did not affect *Vicia faba* L. The chlorophyll concentration in the leaves was significantly dependent on the species of plant. In acidic soil, *Vicia faba* L. and *Lupinus angustifolium* L. assimilated similar amounts of total nitrogen in the grain – 146.8–230.0 and 160.1–220.5 kg N/ha, respectively. *Pisum sativum* L. nitrogen assimilation was lower – 93.0–128.8 kg N/ha. The assimilation of total nitrogen in *Pisum sativum* L. was stimulated by the application of P27K73 and N30, and in *Vicia faba* L. – only by fertilisation with PK.

Keywords: peas; faba bean; lupine; mineral fertiliser; photosynthetic pigment; Fabaceae

Pulses traditionally play a significant role in agriculture as they not only produce good quality protein production but also have an important ecological function – in symbiosis with root nodule bacteria they assimilate atmospheric nitrogen, which reduces the use of mineral fertilisers in agriculture (Allito et al. 2014). The grain of pulse crops is highly valued for the quality of its nutritional properties. In addition, the grain of certain plants has antioxidant properties as well (Akond et al. 2011, Dalaram 2017). Soil and climatic conditions in Europe are favourable for the cultivation of pulse crops. According to the data from FAOSTAT in 2017 in Europe, bean for grain was cultivated on 434 185 ha, pea – on 2 739 646 ha, and lupine – on 274 394 ha and, compared to 2010, the areas increased by 79, 45 and 34%, respectively (http://fenix.fao.org/faostat/internal/en/#data/QC).

The susceptibility of pulse plants to soil fertility determines their growing habitats in Europe. Most of the pulse species, such as bean and *Pisum sativum* L., produce higher grain yields in fertile, neutral soils. Others (lupine and forage pea) can also grow in infertile acidic soils. In addition to soil reaction, the concentration of nutrients is also important for pulses. Mostly they are susceptible to phosphorus deficiency as the development of nitrogen-fixing bacteria requires phosphorus, which is a structural constituent of ATP (adenosine 5'-triphosphate) and ADP (adenosine diphosphate) (Lapinskas 1998, Sanz-Saez et al. 2017). In poor soils, phosphorus deficiency often limits the yield of pulse plants; therefore, it could have a profound significance for improving agricultural sustainability (Amba et al. 2013, Allito et al. 2014).

The prevalence of nitrogen-fixing bacteria (rhizobia) also depends on soil reaction. Their development in the roots of pulse plants and the effectiveness of nitrogen fixation are greatly inhibited by available aluminium ions (Al^{3+}), which dominate in acidic soil. Jaiswal et al. (2018) found that Al^{3+} has lethal effects on many aspects of the rhizobia/legume symbiosis, which include a decrease in root elongation, root hair

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formation and suppression of nitrogen metabolism. Soil pH also influences the accumulation of proteins in the grain of pulse crops (Księżak et al. 2018).

The objective of this work was to compare the productivity of three species of pulse plants (*Vicia faba* L., *Pisum sativum* L., *Lupinus angustifolium* L.) in acidic sandy loam soil and to determine the influence of mineral fertilisers on grain yield and nitrogen assimilation.

MATERIAL AND METHODS

The experiments were conducted at Vokė Branch of Lithuanian Research Centre for Agriculture and Forestry (Baltic Sea region, 54°33'49.8"N, 25°05'12.9"E) in 2016–2018. The soil was sandy loam haplic luvisol (according to by FAO classification system, 2015) (World Reference 2015). Agrochemical characteristics of the arable layer (0–20 cm) were as follows: pH_{KCl} 4.8–5.0, mobile Al^{3+} 20–35 mg/kg, available phosphorus 52–55 mg P/kg, potassium 80–95 mg K/kg and organic carbon (C_{org}) 0.89–1.01%.

The experiment investigated plants that tolerated soil acidity differently. *Lupinus angustifolius* L. is classified as belonging to a group of acidic soil plants as they grow well in very acidic soils with high concentrations of mobile aluminium. *Vicia faba* L. and *Pisum sativum* L. grow better in slightly acidic and neutral soils; growing in acidic soil can result in a significant reduction of yield and nitrogen fixation.

Experimental scheme: Factor A – Fabaceae species: 1. Fodder bean (*Vicia faba* L.); 2. pea (*Pisum sativum* L.); 3. narrow-leaved lupine (*Lupinus angustifolium* L.); Factor B – fertilisation with mineral fertilisers: 1. N0P0K0; 2. N0P27K73; 3. N30P27K73.

Plant seeding rate was 0.8 million seeds per ha for *Vicia faba* L. and 1.0 million seeds per ha for *Pisum sativum* L. and *Lupinus angustifolium* L. Before planting, mineral NPK fertilisers were broadcasted using ammonium nitrate, granular superphosphate and potassium chloride at rates of 30 kg N/ha, 27 kg P/ha and 73 kg K/ha. The experiment was performed in three replications. The size of the plot was 36 m^2 (3 × 12 m), and the observation plot was 22 m^2 (2.20 × 10 m). The recording of crops grain yield was carried out at full maturity stage.

To determine chlorophyll content in plants, spectrophotometer atLEAF+ (Wilmington, USA) was used, which measures the absorption of chlorophyll at the wavelength ranges of blue (400–500 nm) and red (600–700 nm) light. Based on this data, the device calculates a numerical value corresponding to the chlorophyll content in leaves (μg/cm^2) (Zhu et al. 2012, Novichonok et al. 2016). Ten measurements (in 3 replications) were performed at the development of plant stage BBCH 73–77 per each plot. Total nitrogen in grain (N_{tot}, g/kg) was determined by the Kjeldahl method.

Soil samples for agrochemical analysis were taken from the arable (0–20 cm) layer before the establishment of the experiment. The following parameters were analysed: pH by the potentiometric method in 1 mol/L KCl suspension, plant-available phosphorus and potassium by the Egner-Riehm-Domingo (A-L method), organic carbon after dry combustion – Dumas method, mobile Al^{3+} by Sokolov method.

Statistical evaluation of experimental data. The data were evaluated by two factors dispersion analysis of variance (ANOVA) using Fisher‘s test ($P < 0.05$). A correlation-regression analysis of data was performed. The relationship between individual indicators was estimated by using correlation coefficient R at a probability level of 95%.

RESULTS AND DISCUSSION

The basis of all plant growth is photosynthesis. Plant growth, development and metabolic processes depend on the activity of the photosynthetic system and its products.

According to our data, chlorophyll concentration was significantly dependent on the species of plant (Table 1). At the development of fruit stage (BBCH 73–77), the largest content of chlorophyll was found in the leaves of *Lupinus angustifolium* L. – 49.1 μg/cm^2, and the lowest content – 38.8 μg/cm^2 – was found in *Vicia faba* L. There were no significant differences identified in the influence of fertilisation on chlorophyll concentration.

In acidic soil, the *Lupinus angustifolium* L. had the highest content of chlorophyll compared to *Vicia faba* L. and *Pisum sativum* L. ($P < 0.05$). Results show that the applied fertilisation rates were sufficient for intensive photosynthesis and sufficient productivity. The study showed that, regardless of fertilisation, plant genetic properties had a greater impact on the accumulation of photosynthesis pigments. The same conclusion that photosynthetic pigment accumulation was influenced by plant genetic properties was found by Adams et al. (2016), Nguyen et al. (2016) and Rubiales et al. (2018).

The *Lupinus angustifolium* L. demonstrated the highest yield in acidic unfertilised soil compared

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Table 1. Chlorophyll concentration (μg/cm^2) in leaves of plants

Factor	2017	2018	Average 2017–2018
Factor A			
Vicia faba L.	33.8	43.8	38.8
Pisum sativum L.	44.5	40.3	42.4
Lupinus angustifolium L.	52.9	45.3	49.1
$LSD_{0.05}$ (A)	0.81	0.97	0.89
Factor B			
N0P0K0	47.0	43.1	45.0
N0P27K73	46.0	41.4	43.7
N30P27K73	45.6	41.3	43.4
$LSD_{0.05}$ (B)	1.51	1.92	1.73
$LSD_{0.05}$ A × B	2.03	2.57	2.32

LSD – least significant difference

to *Vicia faba* L. and *Pisum sativum* L. According to the data of 2016–2018, its grain yield was 4.9 t/ha, those of *Vicia faba* L. and *Pisum sativum* L. were lower – 3.8 and 2.7 t/ha, respectively (Figure 1). Such advantage of *Lupinus angustifolium* L. was determined by its root system architectonics, its ability to absorb phosphorus from low solubility compounds and tolerance to higher aluminium concentrations in soil (Gresta et al. 2017).

Fertilisation with nitrogen, phosphorus and potassium had a different effect on the grain yield. The yield of *Lupinus angustifolium* L. grain did not significantly change due to fertilisation with PK fertilisers ($P > 0.05$), while N fertilisers reduced the yield by 22.3% ($P < 0.05$) on average compared to unfertilised soil. N fertiliser did not increase the *Vicia faba* L. grain yield ($P > 0.05$), it was similar to that in unfertilised soil, but fertilisation with PK was effective – grain yield increased by 36.8% ($P < 0.05$). It means that phosphorus and potassium were not sufficiently available for *Vicia faba* L. plants in poor acidic soil to fully satisfy the need for these elements. Adak and Kibritci (2016) publish conflicting research results. They found that the highest weight of plant and 100-kernel weight of *Vicia faba* L. was obtained from 80 kg/ha phosphorus and 30 kg/ha nitrogen combination. However, both phosphorus deficiency and high phosphorus fertiliser rates can reduce the yield of *Vicia faba* L. grain. According to Turuko and Mohammed (2014) data, the application of 20 kg P/ha significantly increased dry matter yield and all growth parameters, while, the application of 40 kg P/ha declined plant height and leaf area as compared to the control. It means that the efficiency of phosphorus fertiliser and its effect on the yield of *Vicia faba* L. grain depends on soil properties. Potassium, like phosphorus, is a necessary element in plant development and its deficiency in soil can reduce plant yield. However, as in the case with phosphorus, potassium fertiliser efficiency depends on soil conditions. Symanowicz et al. (2017) argue that both potassium deficiency and surplus potassium may create inappropriate conditions for field *Pisum sativum* L. development. They say high potassium fertiliser rates (166–208 kg K/ha) significantly reduce the content of nitrogen in seeds, straw and pods.

Pisum sativum L. grown in acidic soils, unlike *Vicia faba* L. and *Lupinus angustifolium* L., responded positively to mineral fertilisation. After fertilisation with PK, the *Pisum sativum* L. yield increased by 26.2% and after fertilisation with NPK – by 36.1%. Compared to *Vicia faba* L. and *Lupinus angustifolium* L., the *Pisum sativum* L. root system is less developed; therefore, they are not able, as *Lupinus angustifolium* L. roots, absorb nutrients from lower soil horizons and fertilisation with NPK was effective. The positive effect of fertilisers on the yield of *Pisum sativum* L. is also noted in work by Kanižai Šarić et al. (2016). After studying the effects of N fertilisation and inoculation on *Pisum sativum* L. yield and N fixation, they found that the highest yields of *Pisum sativum* L. were achieved by seed inoculation

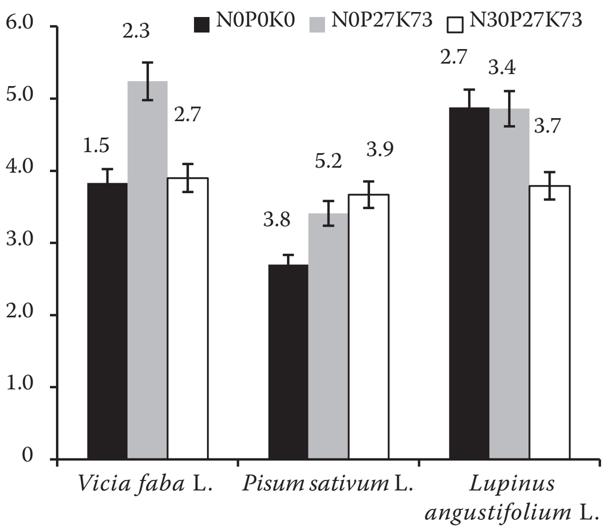

Figure 1. Grain yield (t/ha) (2016–2018) average. Error bars indicate ± $LSD_{0.05}$ ($LSD_{0.05}$ 0.87); *LSD* – least significant difference

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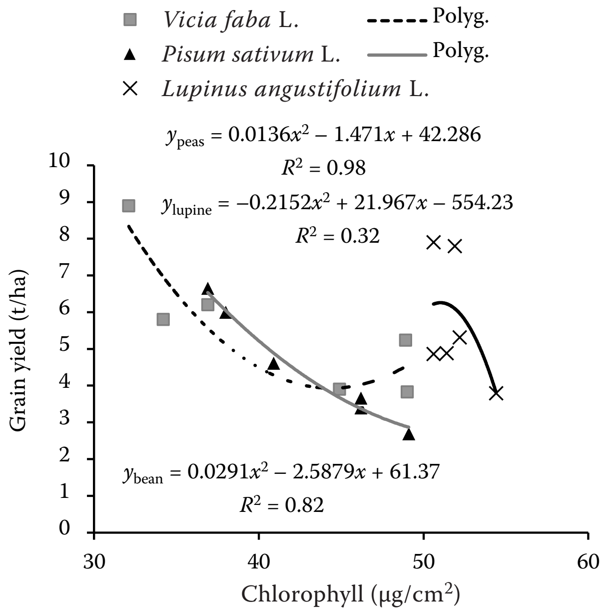

Figure 2. Chlorophyll concentration in leaves and grain yield correlations

with *R. leguminosarum* sv. *viciae* together with N fertilisers (30 to 60 kg N/ha).

The correlation analysis of the grain yield and the chlorophyll concentration in the leaves of pulse crops showed that the yield of the grain yield negatively correlated with this indicator (Figure 2). The particularly strong correlation between these indicators was found for *Vicia faba* L. (R = –0.91) and *Pisum sativum* L. (R = –0.99). Adams et al. (2016) compiled a global dataset and showed that photosynthesis was

Table 2. Nitrogen accumulation in crops (factors A × B interaction)

Plant	Fertiliser	Nitrogen accumulation in grain	
		N (mg/kg)	N_{total} (kg/ha)
Vicia faba L.	N0P0K0	39.3	146.8
	N0P27K73	43.1	230.5
	N30P27K73	42.4	169.4
Pisum sativum L.	N0P0K0	33.9	93.0
	N0P27K73	33.6	118.9
	N30P27K73	33.6	128.8
Lupinus angustifolium L.	N0P0K0	44.0	220.5
	N0P27K73	40.4	196.4
	N30P27K73	42.3	160.1
$LSD_{0.05}$ AB		1.01	8.24

LSD – least significant difference

not related to leaf nitrogen for nitrogen-fixing plants if respective of climate or growth form.

Fertilisation changed both crop yield and N concentration in grain. Changes of N concentrations in *Lupinus angustifolium* L. and *Pisum sativum* L. grain due to fertilisation with PK or NPK were insignificant; however, N content in *Vicia faba* L. grain increased by 9.5% and 7.9%, respectively (Table 2). Changes in crop yield and N concentration affected the accumulation of total nitrogen in plants. While cultivating pulse crops in unfertilized acidic soil, it was observed that the highest total nitrogen content was accumulated in the grain of *Lupinus angustifolium* L. – an average of 220.5 kg N/ha; the content of total nitrogen in *Vicia faba* L. grain was 33.3%, and that in *Pisum sativum* L. grain was 57.3% lower. It is associated with a higher yield of *Lupinus angustifolium* L. grain and higher N concentration in *Lupinus angustifolium* L. grain compared to *Vicia faba* L., and *Pisum sativum* L. Variations in N accumulation confirm that it is more effective to grow infertile soil crops in acidic soils – such crops are more adapted to lower nutrient concentrations and tolerate higher concentrations of mobile aluminium.

Compared to non-fertilised soil, fertilisation with P and K fertilisers mostly increased the total nitrogen accumulation in *Vicia faba* L. grain (+57%), N accumulation in *Pisum sativum* L. increased less – by 27.8%, and fertilisation of *Lupinus angustifolium* L. with PK had a negative impact – total nitrogen accumulation decreased by 18.5%. Only *Pisum sativum* L. responded positively to fertilisation with N fertilisers – total nitrogen accumulation increased by 8.3% compared to PK fertilisation, while N fertilisation of *Vicia faba* L. and *Lupinus angustifolium* L. reduced total nitrogen accumulation in yields by 26.5% and 18.5%, respectively. The results of these studies are analogous to the research data by Pampana et al. (2018), who investigated variations of N fixation in 4 pulse crop species (lupine, chickpea, bean and pea), depending on N fertilisers, and found that the legume species and the N rate were critical factors in determining symbiotic N_2-fixation responses to N fertilisation. Nitrogen fixation in lupine and chickpea linearly decreased with increasing N supply.

In conclusion, fertilisation with phosphorus and potassium fertilisers increased the yield of bean and pea (P < 0.05) and had no effect on the yield of lupine (P > 0.05). Nitrogen fertilisers only increased the yield of pea grain (+36.1%), reduced the yield of lupine grain (–22.8%) and did not affect bean. In

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acidic soil, more of total nitrogen was assimilated in the grain yields of bean (147–230 kg N/ha) and lupine (160–220 kg N/ha). The assimilation of nitrogen in pea plants was stimulated by the application of PK (P27K73) and nitrogen (N30) fertilisers, and in bean plants – only by fertilisation with PK fertilisers.

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