ANNALES
UNIVERSITATIS MARIAE CURIE – SKŁODOWSKA
LUBLIN – POLONIA
VOL. VI SECTIO EEE 1998

Department of Cultivation and Fertilization of Horticultural Plants
Agricultural University in Lublin

ZENIA MICHAŁOJĆ

Influence of Substrate, Nitrogen and Potassium Fertilization on Yield and Chemical Composition of Broad-Bean

Wpływ podłoża oraz nawożenia azotem i potasem na plon i skład chemiczny bobu

Papilionaceous plants, for example broad-bean, can, like all others, use the mineral nitrogen. However, it is assisted by the decrease of nitrogen fixation from the atmosphere in the amount almost same as the up-taken mineral nitrogen (Scherer et al. 1980).

Binding of the atmospheric nitrogen is performed by the symbiotic system: papilionaceous plant — *Rhizobium*. It starts with receiving appropriate signals exchanged between *Rhizobium* and the master plant. Roots of the papilionaceous plants extrude the complex of metabolites, mainly flavonoids, which are taken up by the bacteria and cause the activation of *Rhizobium* (Peters et al. 1986).

Incubation of an appropriate plant and *Rhizobium* leads to hair bending, which creates the conditions for bacteria to penetrate. After penetrating the hair the bacteria create the infection thread. Mitotic activity of primary cortex cells leads to the increase of nodule primordium. In the final phase the cells of *Rhizobium* penetrate the cytoplasm of primordium cells, which grow to mature nodules. Bacteria in cytoplasm divide and differentiate reaching irregular shapes of bacterioids, which bind nitrogen. They supply plants with NH_4^+ and get carbohydrates instead. This process is conducted by specific genes, which induct both partners in various states of symbiotic interaction (Ronson et al. 1987, Adams et al. 1984, Głowacka 1992).

Papilionaceous plants can, in optimum conditions, bind from several dozen up to several hundred kg of N/ha/year, which surely is a positive feature of these plants. However, for the proper growth and development, despite nitrogen, they need other nutritional components such as potassium, which can be supplied to the plants as KCl or K_2SO_4.

Therefore the aim of the present study was to determine the influence of KCl, K_2SO_4, mineral nitrogen and substrate type on the yield of seeds, green tops and nodules in broad-bean.

MATERIAL AND METHODS

The research was performed in a green house from the beginning of March till the middle of May in the years 1989-1991. Two substrates were used: soil+low peat and soil+sand, which were mixed together in 1:1 proportion (by volume). Soil was taken from the surface arable layer of the field, on which broad-bean had been cultivated. Low peat of pH 6.8 contained 30% of organic substance, and the sand of pH 7.3 contained 3% of fine particles.

The experiments were carried out according to the triple cross classification system, in Mitscherlich pots of 5 dm^{-3} capacity. During the period of vegetation of broad-bean the following fertilisation was used (in mg dm^{-3}):

— phosphorus 500 P in the form of 46% superphosphate;

— microelements: 1.8 Mn, 0.6 B, 0.08 Cu, 0.07 Zn, 0.08 Mo, 9.0 Fe. Both phosphorus and microelements were used while filling the pots;

— potassium 1000 K in the form of KCl and K_2SO_4;

— magnesium 100 Mg as $MgSO_4 \times 7\ H_2O$. Potassium and magnesium were used in four doses, every 7 days, starting from the fourth week after sowing.

The test plant was broad-bean (*Vicia faba* var. *major* L), White Windsor variety. Three plants were cultivated in each pot.

Cropping was performed in the period of milk maturity of the seeds after 72 days. Yields of green tops, seeds, and the mass of nodules were measured. Nodule mass was determined twice: during flowering and at the time of harvest. The results were statistically analysed according to the experiment method (triple cross classification), and the confidence half-intervals were calculated using Tukey's test.

In leaves and seeds of broad-bean the following were determined: organic nitrogen using Kjeldahl's method; P, K, Ca — after dry combustion, using common methods; and in the 2% extract of CH_3COOH — sulphur with barium chloride and chlorine with silver nitrate.

RESULTS AND DISCUSSION

The research was carried out for three years in a greenhouse, where the temperature was about 15°C at night, and 22°C during the day, and the water capacity was kept at the level of 60%. Broad-bean vegetation period, from sowing till cropping in the phase of milk maturity, lasted for about 10 weeks.

Considering that similar results had been received concerning the investigated features, they are presented here as average values for three years of the study.

Significant differences in yield of green tops and seeds in plants which grew in the substrate soil+peat in comparison with the soil+sand (which was the bed with higher content of organic substance) were noticed (tab. 1).

In Prat's research (1960) it was proved that humic acids extracted from low peat stimulated the symbiosis of papilionaceous plants with *Rhizobium leguminosarum*. These acids were also buffering the substrate, which regulated the reaction and the concentration of nutritive components, which positively influenced the growth and development of plants.

Fertilisation with mineral nitrogen was the second of the investigated factors. Small doses were used every other day to try to imitate the binding of atmospheric nitrogen. The results showed that the nitrogen added in the amount of 0.3 g N dm^{-3} of the substrate had an important influence on the yield of seeds and green tops (tab. 1).

Bendycka et al. (1995) used the nitrogen doses from 0 to 240 kg N ha^{-1}. However, the only doses which significantly influenced the growth of broad-bean seeds were those from 20 to 120 kg N ha^{-1}. Michałojć (1997) showed significantly higher yield of broad-bean seeds after using 40 kg N ha^{-1}.

The mass of nodules was determined twice: in full flowering period and during the period of milk maturity of the seeds. Significant influence of mineral N and the substrate on noduling was stated. The nitrogen used in the form of NH_4NO_3 since the beginning of flowering till cropping significantly limited the mass of formed nodules (tab. 1). The results are similar to those received by Richards et al. (1979) and Scherer et al. (1980). They stated that the used mineral nitrogen limited the uptake of atmospheric nitrogen from air and noduling in bean, broad-bean, and small-grained broad-bean.

In the substrate with higher content of organic substance (soil+peat) it was found out that the mass of nodules was lower, during flowering, than in soil+sand. During the second term that correlation was not observed. The positive influence of the soil+sand substrate on the development of nodules

Tab. 1. Yield of fresh mass of green tops, seeds and nodules of broad-bean in g/pot. Average values from 1989-1991

Substrate	Soil + peat				Soil + sand				$LSD_{0.05}$
N dose [g N dm^{-3}]	0		0.3		0		0.3		
K fertiliser	KCl	K_2SO_4	KCl	K_2SO_4	KCl	K_2SO_4	KCl	K_2SO_4	
Top-parts	217.58	211.66	244.63	221.56	138.40	124.70	150.93	138.13	
mean for N dose	214.61		233.09		131.55		144.53		10.08
mean for substrate	233.85				138.04				57.49
Seeds	33.40	41.50	44.33	45.76	17.53	18.30	24.23	26.00	
mean for N dose	37.45		44.89		17.91		25.11		6.05
mean for substrate	41.25				21.52				7.57
Nodules I*	6.30	5.05	1.85	2.15	9.25	6.90	4.50	4.55	
mean for N dose	5.67		2.00		8.07		4.52		2.17
mean for substrate	3.85				6.30				1.38
Nodules II*	7.90	6.76	3.10	3.76	6.40	5.53	3.96	3.26	
mean for N dose	7.33		3.43		5.96		3.61		2.13
mean for substrate	5.38				4.79				n.s.

*I — harvest at full flowering

*II — harvest at milk maturity of seeds

in the initial period of vegetation should be explained by a better possibility for the air to reach the root zone, which means better conditions for the nodules' development.

Maliszawska et al. (1971) emphasize the positive influence of the physical properties of soil on the development of *Rhizobium*. The results concerning the mass of nodules in the period of cropping are also interesting. Close or higher mass of the nodules during seeds' milk maturity than during the period of flowering was stated. These results prove that broad-bean keeps the possibility of noduling, and therefore of binding the atmospheric nitrogen until the cropping of seeds. Schilling (1982) stated that in peas and whi-

Tab. 2. Content of dry mass, N, P, K, Ca, Cl, $S-SO_4$ (in % d. m.) in leaves and seeds of broad-bean. Average values from 1989-1991

Substrate		Soil + peat				Soil + sand			
K fertiliser		KCl		K_2SO_4		KCl		K_2SO_4	
Dose g N dm^{-3}		0	0.3	0	0.3	0	0.3	0	0.3
D. mass	leaves	15.86	16.01	16.14	16.91	15.14	15.39	17.04	16.66
	seeds	22.61	24.91	24.50	24.77	23.38	24.20	20.50	24.12
N-total	leaves	3.36	3.64	3.56	3.61	3.77	3.86	3.82	4.05
	seeds	4.39	4.45	4.45	4.47	4.25	4.38	4.27	4.36
	leaves	0.08	0.08	0.10	0.08	0.19	0.19	0.21	0.21
P	seeds	0.37	0.40	0.44	0.41	0.47	0.40	0.48	0.46
	leaves	3.23	3.72	3.16	3.49	3.76	4.74	4.14	4.42
K	seeds	1.69	1.60	1.51	1.65	1.81	1.71	1.77	1.73
	leaves	3.58	3.05	2.55	2.46	3.28	2.69	1.46	1.27
Ca	seeds	0.03	0.03	0.03	0.02	0.03	0.01	0.01	0.01
	leaves	1.76	2.43	0.21	0.27	2.59	3.10	0.30	0.41
Cl	seeds	0.12	0.17	0.06	0.06	0.20	0.20	0.08	0.08
	leaves	0.14	0.23	0.32	0.36	0.25	0.25	0.62	0.57
$S-SO_4$	seeds	0.03	0.03	0.05	0.05	0.02	0.02	0.06	0.07

te lupine the bonding of N_2 ended after flowering, while in broad-bean it lasts also after flowering, which means that there is a possibility of using the easily available source of nitrogen much longer.

Differentiated potassium fertilisers were the third of the investigated factors. Lack of the influence of KCl and K_2SO_4 on yield of seeds, green tops, and nodule mass in broad-bean was stated.

Chemical composition of plants was determined by analysing seeds and leaves during the period of cropping. Trace quantities of mineral forms of nitrogen in leaves and their lack in seeds were found. That is why the content of $N-NH_4$ and $N-NO_3$ has not been presented in table 2. The amount of organic nitrogen was differentiated to a small degree by the investigated experimental factors (tab. 2).

Many authors emphasize close correlation between the nitrogen dose and its content in a plant. Considering the higher yield of green tops and seeds after using mineral nitrogen and very close content of nitrogen in plants which used the atmospheric N and the mineral N, we should come to a conclusion that the plants being able to use the easily available mineral nitrogen — took it, but at the same time they limited noduling. It was also noted that after using mineral nitrogen the content of potassium, chlorine, and sulphur in leaves was higher, while the content of calcium was lower than in the ones that used atmospheric nitrogen. That correlation, although not so significant, was also observed in seeds. The influence of the mineral nitrogen on the content of dry mass was not noticed univocally. It is a widely known fact that the increasing doses of nitrogen cause the decrease of dry mass content. But this dependence was not confirmed by this research. It should be explained by very small doses of nitrogen (20 mg N/dm^3 every other day), which were applied during the vegetation period, to try to imitate the binding of the atmospheric nitrogen (tab. 2).

According to Benedycka et al. (1995), a dose of 0.5 g N/pot stimulated the growth and the accumulation of nutrients in broad-bean. It was also confirmed by this research.

Differentiated substrate decided only upon the content of phosphorus in broad-bean leaves. Its concentration in the plants growing on the substrate: soil+sand was stated twice as high (tab. 2). It is obviously related to the amount of calcium in the substrate: in the soil+peat it was about 2200 mg Ca dm^{-3}, while in the soil+sand it was about 820 mg Ca dm^{-3} (tab. 3). It proved that high concentration of calcium in the substrate limited the amount of phosphorus easily available for the plants.

The potassium fertilisers used considerably differentiated the content of dry mass, calcium, chlorine, and sulphur in plants and, to a smaller degree, the content of nitrogen, phosphorus, and potassium (fig. 1). In plants fertilised with potassium in the form of KCl, higher content of calcium and chlorine was recorded, while in plants fertilised with K_2SO_4 higher contribution of dry mass and sulphur was found (tab. 2, fig. 1).

Tab. 3. Content of N-NO_3, P, K, Ca, Cl, S-SO_4 in mg dm^{-3} in the substrate, in the experiment with broad-bean. Average values from 1989-1991

Substrate	Soil + peat				Soil + sand			
K fertiliser	KCl		K_2SO_4		KCl		K_2SO_4	
Dose g N dm^{-3}	0	0.3	0	0.3	0	0.3	0	0.3
N-NO_3	30	45	31	67	15	29	18	28
P	154	168	172	165	175	178	193	181
K	545	605	615	608	556	662	675	690
Ca	2266	2056	2516	2143	818	860	759	899
Cl	486	560	56	56	456	516	68	89
S-SO_4	227	235	475	440	126	118	280	317

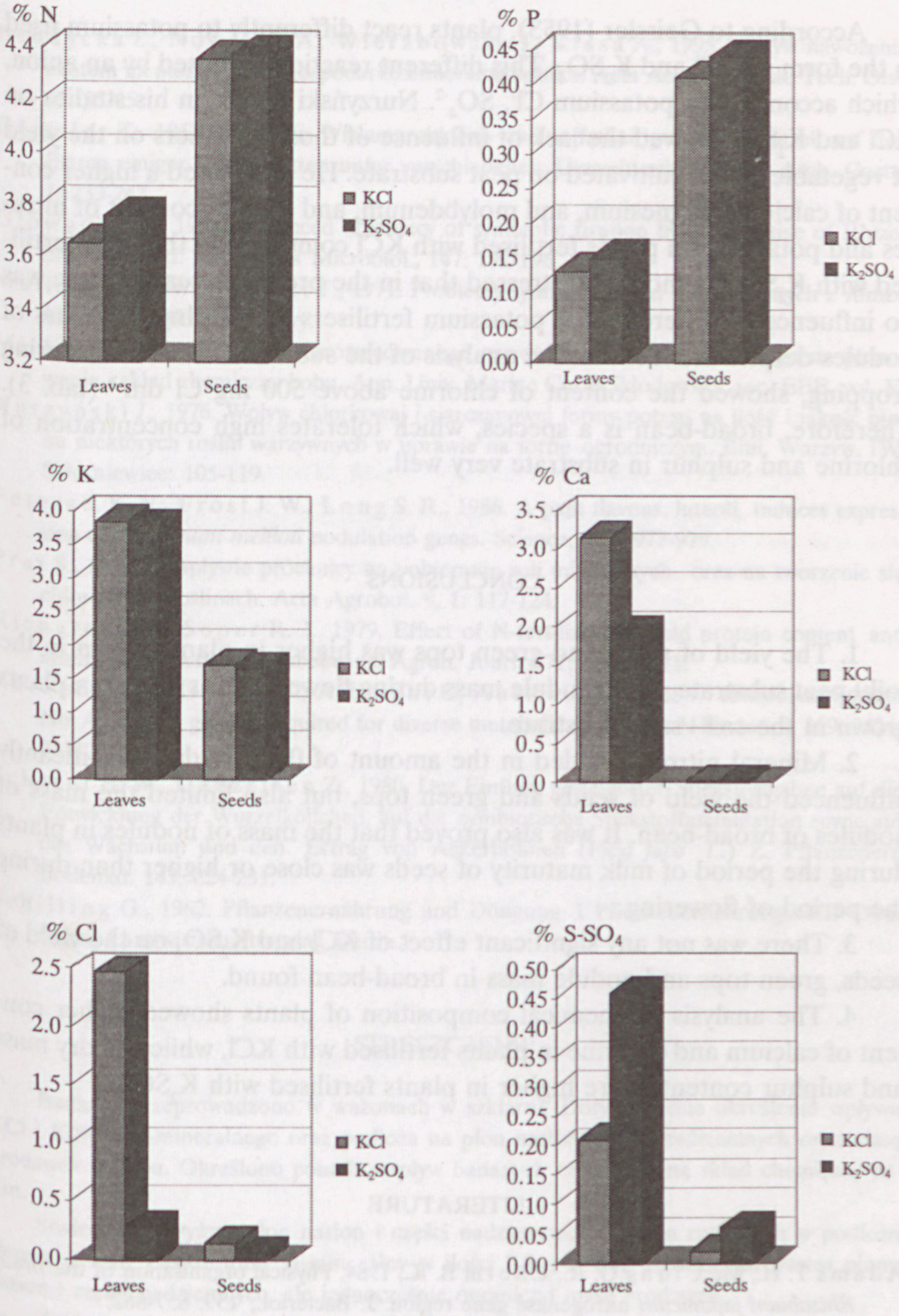

Fig.1. Influence of fertilising with KCl and K_2SO_4 on chemical composition of leaves and seeds of broad-bean

According to Geissler (1953), plants react differently to potassium used in the form of KCl and K_2SO_4. This different reaction is caused by an anion, which accompanies potassium Cl^-, SO_4^{-2}. Nurzyński (1976) in his studies on KCl and K_2SO_4 showed the lack of influence of those fertilisers on the yield of vegetable plants cultivated on peat substrate. He also stated a higher content of calcium, magnesium, and molybdenum, and a lower content of nitrates and potassium in plants fertilised with KCl compared to the ones fertilised with K_2SO_4. It should be stressed that in the present research there was no influence of differentiated potassium fertilisers on yielding and mass of nodules despite the fact that the analysis of the substrate carried out during cropping, showed the content of chlorine above 500 mg Cl dm^{-3} (tab. 3). Therefore, broad-bean is a species, which tolerates high concentration of chlorine and sulphur in substrate very well.

CONCLUSIONS

1. The yield of seeds and green tops was higher in plants grown in the soil+peat substrate, while nodule mass during flowering was higher in plants grown in the soil+sand substrate.

2. Mineral nitrogen added in the amount of 0.3 g N dm^{-3} significantly influenced the yield of seeds and green tops, but also limited the mass of nodules of broad-bean. It was also proved that the mass of nodules in plants during the period of milk maturity of seeds was close or higher than during the period of flowering.

3. There was not any significant effect of KCl and K_2SO_4 on the yield of seeds, green tops and nodule mass in broad-bean found.

4. The analysis of chemical composition of plants showed higher content of calcium and chlorine in plants fertilised with KCl, while the dry mass and sulphur contents were higher in plants fertilised with K_2SO_4.

LITERATURE

Adams T. H., Mc Clung G. R., Chelm B. K., 1984. Physical organisation of the Brady *Rhizobium japonicum* nitrogenase gene region. J. Bacteriol., 159: 857-862.

Bendycka Z., Nowak G. A., 1995. Wpływ żywienia azotem na produktywność i gospodarkę mineralną bobiku. Acta Acad. Agricult. Tech. Olst. 61: 45-55.

Bendycka Z., Nowak G. A., Wierzbowska J., Klasa A., 1995. Wpływ nawożenia azotem na plonowanie i gospodarkę mineralną bobiku. Acta Acad. Agricult. Tech. Olst. 61: 55-65.

Geissler T., 1953. Über die Wirkung chlorid- und sulfathaltiger Düngemittel auf den Ertrag einiger Gemüsearten unter verschiedenen Umweltverhältnissen. Arch. Gartn. 1: 233-243.

Głowacka M., 1992. Enhanced efficiency of symbiotic fixation by a derivative of Rhizobium meliloti. Zentralblat Microbiol., 147: 192-196.

Maliszewska W., Wróbel T., 1971. Problemy symbiozy roślin motylkowatych z *Rhizobium*. Wyd. IUNG Puławy.

Michałojć Z., 1997. Wpływ zróżnicowanego nawożenia azotowo-potasowego na plonowanie i skład chemiczny bobu. Ann. Univ. Mariae Curie-Skłodowska, sec. EEE, vol. V.

Nurzyński J., 1976. Wpływ chlorkowej i siarczanowej formy potasu na ilość i jakość plonu niektórych roślin warzywnych w uprawie na torfie ogrodniczym. Biul. Warzyw. IW, Skierniewice: 105-119.

Peters S. K. N., Frost J. W., Long S. R., 1986. A pant flavons, luteoli, induces expression of *Rhizobium meliloti* nodulation genes. Science, 223: 977-979.

Prat S., 1960. O wpływie próchnicy na pobieranie soli mineralnych oraz na tworzenie się chlorofilu w roślinach. Acta Agrobot. 9, 1: 117-124.

Richards J. E., Soper R. J., 1979. Effect of N-fertilizer on yield protein content and symbiotic N-fixation in fabebeans. Agron. Journ., 71, 5: 807-811.

Ronson C. W., Nixon B. T., Albright L., Auslibel F. M., 1987. *Rhizobium meliloti* Ntr A (rpo N) gene is required for diverse metabolic functions. J. Bacteriol. 169: 2424-2431.

Scherer H. W., Danzeisen Z., 1980. Der Einfluss gesteigerter Stickstoffgaben auf die Entwicklung der Wurzelköllchen, auf die symbiotische Stickstoffassimilation sowie auf das Wachstum und den Ertrag von Ackerbohnen (*Vicia faba* L.) Z. Pflanzenern Bodenkd. 143, 224-231.

Schilling G., 1982. Pflanzenernährung und Düngung. I Pflanzenernährung.VEB Deutscher Landwirtschaftsverlag Berlin.

STRESZCZENIE

Badania przeprowadzono w wazonach w szklarni. Dotyczyły one określenia wpływu KCl i K_2SO_4, N-mineralnego oraz podłoża na plon nasion, części nadziemnych oraz masę brodawek u bobu. Określono ponadto wpływ badanych czynników na skład chemiczny roślin.

Stwierdzono wyższy plon nasion i części nadziemnych u roślin rosnących w podłożu ziemia + torf. Zastosowany N-mineralny w ilości 0,3 g N dm^{-3} powodował wzrost plonu nasion i części nadziemnych, ale jednocześnie ograniczył masę brodawek.

Zróżnicowane nawozy potasowe (KCl, K_2SO_4) nie miały wpływu na wielkość plonu nasion, części nadziemnych i brodawkowanie u bobu, ale różnicowały zawartość suchej masy, wapnia, chloru i siarki w roślinach.