Tomáš Lošák, Luděk Páleníček

Tom XXVI

Department of Agrochemistry, Soil Science, Microbiology and Plant Nutrition Mendel University of Agriculture and Forestry Brno, Czech Republic

Using nitrogen and sulphur for the poppy (*Papaver somniferum* L.) nutrition^{*}

Wykorzystanie azotu i siarki w nawożeniu maku (Papaver somniferum L.)

Key words: nitrogen, sulphur, poppy, yield seed, capsules per plant, morphine

The pot trial was carried out to estimate the effect of interaction of nitrogen and sulphur fertilization on seed yield, number of capsules per plant and concentration of morphine in the poppy straw (i.e. empty capsule + 15 cm of the stem) of the variety Opál. The natural content of sulphate sulphur in the soil was 18.3 mg $S-SO_4^{-2}\cdot kg^{-1}$ of soil. The respective variants were supplied with ammonium sulphate (20,5% N and 24% S) to the level of 40 mg $S-SO_4^{-2}\cdot kg^{-1}$ of soil. Nitrogen fertilization was applied in three doses: 0.3, 0.6 and 0.9 g N-pot⁻¹. Nitrogen was made up to the appropriate level with ammonium nitrate (34.5% N). Fertilizers were applied in one single application after the emergence of the plant.

The increase in seed yields in all the fertilized variants was statistically highly significant, i.e. by 54.8–143.7%, compared to the unfertilized control, and the yields increased with the higher doses of nitrogen. The increase in the level of sulphate sulphur in soil from 18.3 mg $S-SO_4^{-2}$ (control) to 40 mg $S-SO_4^{-2}$ resulted in a statistically significant increase in yields by 12.6%, but only in conjunction with the lowest dose of nitrogen (0.3 g N·pot⁻¹). In the other variants the joint application of nitrogen and sulphur had no statistically significant effect compared to the same variants without sulphur.

A statistically significant increase in the number of capsules per plant was seen with increasing N doses, in the fertilized variants ranging between 1.0 and 2.2 capsules per plant, compared to 1.0 per control plant. Irrespective of sulphur content in the soil, the highest number of capsules per plant (2.11 and/or 2.20) was noticed in the variants fertilized with the highest dose of nitrogen (0.9 g N·pot⁻¹).

The concentration of morphine alkaloid of in the poppy straw of the fertilized variants varied only a little, i.e. 0.63–0.76%, compared to 0.61% in the control. The highest dose of N resulted in the highest morphine concentration; sulphur showed no major effects.

Slowa kluczowe: azot, siarka, mak, plon, liczba makówek na roślinie, morfina

Celem doświadczenia wazonowego było określenie wpływu współdziałania nawożenia azotem i siarką na plon nasion, liczbę makówek na roślinie i zawartość morfiny w słomie makowej (pusta makówka + 15 cm łodygi) odmiany Opal. Zawartość siarki siarczanowej w glebie wynosiła 18,3 mg $S-SO_4^{-2}\cdot kg^{-1}$ gleby. Poszczególne warianty doświadczenia nawieziono siarczanem amonu (20,5% N i 24% S) do poziomu 40 mg $S-SO_4^{-2}\cdot kg^{-1}$ gleby. Nawożenie azotem zastosowano w trzech dawkach: 0,3, 0,6 i 0,9 g N·wazon⁻¹. Azot uzupełniono do zaplanowanego poziomu saletrą amonową (34,5% N). Nawozy zastosowano po wschodach roślin.

^{*} The present study was conducted within grant NAZV indicated QF 3173

Tomáš Lošák ...

Plon nasion istotnie wzrastał wraz ze wzrostem dawek nawożenia azotowego (54,8–143,7%). Wzrost zawartości siarki siarczanowej w glebie z 18,3 do 40 mg $S-SO_4^{-2} \cdot kg^{-1}$ przyczynił się do statystycznie istotnego wzrostu plonu — 12,6% tylko w warunkach najniższego poziomu nawożenia azotem (0,3 g N·wazon⁻¹). W pozostałych wariantach doświadczenia nawożenie siarką nie modyfikowało istotnie plonu nasion.

Liczba makówek na roślinie wzrastała od 1 do 2,2 wraz z wzrostem nawożenia azotem. Bez względu na zawartość siarki w glebie najwięcej makówek na roślinie odnotowano w wariantach doświadczenia, w których zastosowano największą dawkę azotu (2,11 i 2,20).

Wpływ nawożenia na zawartość morfiny w słomie makowej był nieznaczny i wzrastał od 0,63 do 0,76%. Rośliny nawożone najwyższą dawką azotu charakteryzowały się najwyższą zawartością morfiny w słomie.

Introduction

Poppy (*Papaver somniferum* L.) is an important oil plant grown in the Czech Republic along with winter rape (*Brassica napus* L.) and sunflower (*Heliantus annus* L.). It is grown on an area of 30-40,000 ha and the average yield is 0.6 t of seeds ha⁻¹; in 2004 we had a bumper crop of 0.92 t of seeds ha⁻¹ (Mottl 2005). The technology of cultivation has been thoroughly elaborated in the Czech Republic, which is now the main grower and exporter of poppy seed in Europe. Nowadays poppy is grown not only for the seeds but also for the poppy straw, which is the source of the alkaloid morphine used in the pharmaceutical industry; this increases and stabilizes the economics of poppy cultivation (Vašák et al. 2005).

Essential in poppy nutrition is the content of macro- and micro-biogenic elements in soil, and in plant, which is positively reflected both in yields and quality of production. Nitrogen nutrition is crucial for poppy; it is necessary to select a suitable dose, form and date of application (Yadav et al. 1984). According to Jain (1990) split nitrogenous nutrition is better as it results in higher yield and content of alkaloids.

Sulphur came to the fore of interest of breeders in the past ca 10 years when sulphur applications were considerably reduced, particularly by reducing the use of mineral and organic fertilizers, fungicides and reducing SO₂ emission below 10 kg S·ha⁻¹ (Lošák 2003). The plant takes up sulphur from soil solution especially in the form of sulphates (SO₄²⁻) and the first stable organic compound is the amino acid cysteine (Marschner 1995). In plant sulphur there is a component of various primary and secondary metabolites, among which we number the essential amino acids methionine, glutathione (a precursor of phyto-chelatins, which bound risk elements into chelate complexes), ferredoxin, sulpholipids, vitamin B₁ and vitamin H, enzymes and coenzymes or essential oils of the alliine and allicine type (Haneklaus et al. 1997, De Kok et al. 1993). Not only have secondary S — metabolites a high nutritional value, but they are also important in terms of resistance of the plants against diseases and pests (Schnug 1990).

262

With a deficiency of sulphur, increasing doses of nitrogen increase shortage of sulphur (Janzen, Bettany 1984). Deficient sulphur nutrition may also be the cause of lower nitrogen utilization and the final consequences are lower yields, poorer quality of production and the possibility of undesirable washing out of nitrates (NO_3^-) into the hydrosphere (Schnug 1993). Every kilogram of sulphur, which limits plant growth, leads to losses in nitrogen ranging between 4 and 15 kg (Schnug 1991). Joint application of N and S should be conducted in spring in order to reduce the risk of being washed out during winter if applied in autumn (Schnug 1993). According to Richter (2001) optimal nutrition with nitrogen and sulphur increases not only nitrogen utilization, which is reflected in higher yields, but also stabilizes the content of oil in oil plants. Jasinska et al. (1997) and Wielebski et al. (1998) described increased production of protein and reduced oil content under increasing doses of nitrogen. Tomar et al. (1993), Subrahmanyam et al. (1992) described the use of sulphur in poppy nutrition with regard to growth, seed yields, oil content, and the content of protein and of the alkaloid morphine.

Material and methods

One year vegetative pot trial with poppy was conducted in 4 replications (pods) in the outdoor vegetation hall of the Department of Agrochemistry, Soil Science, Microbiology and Plant Nutrition of Mendel University of Agriculture and Forestry in Brno. In the autumn of 2003 plastic pots were filled with 9.5 kg of medium heavy soil characterized as fluvial soil. The soil was characterized by good content of P and K and high content of Ca. Content of Mg was very high and soil reaction was slightly acid (Tab. 1).

Table 1

pH/KCl	Р	K	Ca	Mg	$S-SO_4^-$
5.85	103	239	3769	366	18.3
slightly acid słabo kwaśny	good dobry	good dobry	high wysoki	very high bardzo wysoki	

Agrochemical characteristics of the soil in mg·kg⁻¹ Agrochemiczna charakterystyka gleby (Mehlich III)

During winter the soil in the pots settled naturally. On 5th April shallow seeding of the poppy variety Opál into pots was carried out. This is the most widely spread poppy variety in the Czech Republic with a medium content of morphine in the poppy straw.

The uniform emergence of plants was noticed 10 days after seeding. On 16th April 2004 fertilizers were applied in the form of water solution. The natural

content of sulphate sulphur in soil was 18.3 mg·kg⁻¹ of soil (S₀). In variants 5–7 the sulphur was increased in the form of ammonium sulphate (20.5% N and 24% S) to the level of 40 mg·kg⁻¹ of soil (S₁) (Tab. 2). In variants 2–7, nitrogen was balanced to the appropriate level with ammonium nitrate (34.5% N). Nitrogen fertilization was applied in three doses: 0.3, 0.6 and 0.9 g N·pot⁻¹ (N₁, N₂ and N₃).

Tabl	e	2
1 uoi	v	-

Variant — Wariant		N [g·pot ⁻¹ — g·wazon ⁻¹]	$S-SO_4^{-2}$ [mg·kg ⁻¹]	
1	control	0	18.3	
2	N_1S_0	0.3	18.3	
3	N_2S_0	0.6	18.3	
4	N_3S_0	0.9	18.3	
5	N_1S_1	0.3	40.0	
6	N_2S_1	0.6	40.0	
7	N_3S_1	0.9	40.0	

Pattern of the trial — Schemat eksperymentu

On 29th April, in the stage of 2 leaves, the plants were thinned to 7 plants per pot. During vegetation the plants were watered with de-mineralized water on a regular basis, the plants were sampled for analyses, insecticides against beet aphid (*Aphis fabae*) were applied and the pots were kept free of weeds.

In the stage of full maturity on 16th August 2004 4 plants from each pot were harvested. The alkaloid morphine was determined polarographically in the Research Institute of Oilplants Opava. Results of cropping were elaborated statistically using the method of variance analysis expressing minimal significant differences ($Dt_{0.05}$ and $Dt_{0.01}$) at 95% and 99% level of probability, respectively.

Results and discussion

Nitrogen fertilization caused significant increase of seed yield (Tab. 3). The Yield increased with the rise of nitrogen doses from 54.8 to 143.7%. These results correspond with findings of Yadav et al. (1984) where doses of nitrogen increasing 50, 100, 150 and 200 kg N·ha⁻¹ increased seed yields compared to the control. Also Jain et al. (1990) detected yields increased by 37.5% when nitrogen doses increased from 30 to 90 kg N·ha⁻¹. Kharwara et al. (1988) noted a significant stimulation in seed yields when the nitrogen dose was increased from 75 kg N·ha⁻¹ to 150 kg N·ha⁻¹.

Variant Wariant	Seed yield Plon nasion		Number of capsules per plant Liczba makówek na roślinie		Content of morphine
	[g·pot ⁻¹] [g·wazon ⁻¹]	[%]	pcs	[%]	Zawartość morfiny [%]
1 control	5.58	100.0	1.00	100.0	0.61
$2 N_1 S_0$	8.64	154.8	1.00	100.0	0.65
$3 N_2 S_0$	11.46	205.4	1.75	175.0	0.63
$4 N_3 S_0$	13.60	243.7	2.20	220.0	0.76
5 N_1S_1	9.34	167.4	1.25	125.0	0.64
$6 N_2 S_1$	11.79	203.2	1.50	150.0	0.66
7 N_3S_1	13.44	240.9	2.11	211.0	0.70
Dt _{0.05}	0.68		0.24		
Dt _{0.01}	1.14		0.36		

Influence of nitrogen and sulphur fertilization on seed yield, number of capsules per plant and morphine content in poppy straw — *Wplyw nawożenia azotem i siarką na plon nasion, liczbę makówek na roślinie i zawartość morfiny w słomie makowej*

The positive effect of sulphur was statistically significant ($P_{0.05}$) only when applied together with the lowest dose of nitrogen (var. 5); then the yields increased by 12.6% compared to the same variant not fertilized with sulphur (var. 2). These results correspond with the conclusions of Subrahmanyam et al. (1992) who described the 18.6% increase in poppy seed yields when 60 kg S·ha⁻¹ was applied. Tomar et al. (1993) compared four levels of sulphur fertilization (0, 15, 30 and 45 kg S·ha⁻¹) in the form of elementary S and elementary super-phosphate. The highest dose of sulphur resulted in the highest seed yields.

The number of capsules per plant (Tab. 3) increased statistically significantly with the increase of nitrogen dose and in fertilized variants fluctuated between 1.0 and 2.2 capsules per plant compared to 1.0 in the control. With the highest dose of nitrogen (0.9 g N·pot⁻¹) the number of two capsules per plant was exceeded under both levels of sulphur (2.11 and 2.20; and variant 7 and 4, respectively).

The content of morphine in poppy straw (Tab. 3) after harvest (empty poppy head + 15 cm of stem) increased under the influence of nitrogen fertilization and markedly exceeded the average 0.56% of morphine given for the variety Opál. This is in accordance with a number of literary sources, which indicated that the increasing concentration of morphine is dependent on the increasing level of N fertilization (Yadav et al. 1984). Kharwara et al. (1988) detected that under a higher level of nitrogen it increased from 75 kg N·ha⁻¹ to a double value. Schrodter (1965) confirmed a higher content of morphine with a dose of 50–60 kg N·ha⁻¹ and sufficient supply of P and K.

Table 3

Tomáš Lošák ...

The effect of sulphur on the morphine content was ambiguous. If the level of nitrogen fertilization was lower (N₁ a N₂) the differences between variants with sulphur and without were not significant (variant 2 compared with variant 5 and variant 3 compared with variant 6). The highest dose of nitrogen (N₃) in combination with sulphur reduced the content of morphine to 0.70% (var. 7) compared with 0.76% of the variant without sulphur application (var. 4). This finding is at variance with the conclusions of Subrahmanyam et al. (1992) who noted that the content of alkaloids, including morphine, increased with higher doses of sulphur (30, 60 and 90 kg S·ha⁻¹). The content of morphine is primarily affected by the concrete variety and the effect of other factors is irregular references.

Conclusions

Seed yield increased with the rise of nitrogen fertilization. Sulphur fertilization caused significant increase of seed yield when it was applied with the lowest dose of nitrogen. Nitrogen fertilization increased the number of capsules per plant. The highest dose of nitrogen resulted in the highest morphine concentration in poppy straw.

References

- De Kok L.J., Stulen L. 1993. Functions of glutathione in plants under oxidative stress. In: De Kok L.J., Stulen L., Rennenberg H., Brunold C., Rauser W.E. Sulphur Nutrition and Assimilation in Higher Plants, The Hague: 125-138.
- Haneklaus S., Hoppe L., Bahadir M., Schnug E. 1997. Sulphur nutrition and alliin concentrations in Allium species. Sulphur metabolism in Higher Plants, The Hague: 113-118.
- Jain P.M. 1990. Effect of Split Application of Nitrogen on Opium Poppy. Indian J. Agron., 35 (3): 240-242.
- Jain P.M., Gaur B.L., Gupta P.C. 1990. Response of Opium Poppy Varieties to Nitrogen. Indian J. Agron., 35 (3): 243-245.
- Janzen H.H., Bettany J.R. 1984. Sulfur nutrition of rapeseed: I. Influence of fertilizer nitrogen and sulfur rates. Soil Sci. Soc. Am. J., 48: 100-107.
- Jasinska Z., Kotecki A., Kozak M. 1997. Residual effect of leguminous crops and nitrogen fertilizer application on the development and yield of winter oilseed rape (*Brassica napus*). Rośliny Oleiste Oilseed Crops, XVIII (1): 187-198.
- Kharwara P.C., Awasthi O.P., Sing C.M. 1988. Effect of Sowing Dates, Nitrogen and Phosphorus Levels on Yield and Quality of Opium Poppy. Indian J. Agron., 33 (2): 159-163.
- Lošák T. 2003. Studium utilizace dusíku při hnojení sírou u ozimé řepky. Doktorská disertační práce, Brno, s. 168.
- Marschner H. 1995. Mineral Nutrition of Higher Plants. Academic Press Limited, London, s. 889.

Mottl V. 2005. Pěstování, ekonomika a trh máku setého. In: Sborník odborných seminářů "Mák v roce 2005", 4. makový občasník: 5-11.

Richter R., Hřivna L., Cerkal R. 2001. Výživa a hnojení ozimé řepky. SPZO Praha, MZLU Brno, s. 41.

Schnug E. 1990. Sulphur nutrition and quality of vegetables. Sulphur in Agriculture, 14: 3-7.

- Schnug E. 1991. Sulphur nutritional status of European Crops and consequences for agriculture. Sulphur in Agriculture, 15: 7-12.
- Schnug E. 1993. Ökosystemare Auswirkungen des Einsatzes von N\u00e4hrstoffen in der Landwirtschaft. Berichte \u00fcber Landwirtschaft. In: BML, Bonn (Germany) (Ed.): N\u00e4hrstoffe und Pflanzenschutzmittel in Agrar\u00f6kosystemen. M\u00fcnster-Hiltrup, Germany: Landwirtschaftsverlag, 25 (1): 25-48.
- Schrodter H. 1965. Untersuchungen über Veränderung des Morphingehalts reifender Mohnkapseln. Pharmazie, 20: 169.
- Subrahmanyam K., Verma RK., Naqvi AA., Singh DV. 1992. The effect of forms of sulphur on yield and quality of seed, oil and alkaloids of opium poppy (*Papaver somniferum* L.). Acta Horticulturae, 306: 431-435.
- Tomar SS., Mohd-Abbas Nigam KN. 1993. Effect of sulphur on growth and yield of opium poppy (*Papaver somniferum* L.), Indian J. Agron., 38 (2): 346-347.
- Vašák J., Cihlář P., Roubal T., Vlk R. 2005. Některé využitelné výsledky z výzkumu pěstování jarního máku (*Papaver somniferum* L.). In: Sborník odborných seminářů "Mák v roce 2005", 4. makový občasník: 14-17.
- Yadav R.L., Mohan R., Singh R., Verma R.K. 1984. The effect of application of nitrogen fertilizer on the growth of opium poppy in north central India. J. of Agricultural Science, Camb. 102: 361-366.
- Wielebski F., Wojtowicz M. 1998. Response of winter rape varieties to high nitrogen fertilisation in rye soils in Experimental Station Zielecin. Rośliny Oleiste – Oilseed Crops, XIX (2): 507-514.