Nitrogen content in yield of dry aboveground and root mass of forage lucerne (*Medicago sativa* L.) after mineral nitrogen fertilization and water deficiency stress

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Abstract. The effect of mineral nitrogen fertilization and water deficiency stress on nitrogen content in yield in dry aboveground and root mass of forage lucerne was tested at the Institute of Forage Crops, Pleven, Bulgaria (2003-04). Mineral nitrogen fertilization at the doses of 40, 80, 120 and 160 mg N kg⁻¹ soil was tested in pot trials. Nitrogen fertilizer was applied as ammonium nitrate. Ten-day water deficiency stress at the budding stage of lucerne was imposed by stopping the irrigation till soil moisture dropped to 37–40% FC. It was found that mineral nitrogen fertilization increased nitrogen in dry aboveground mass yield – for doses of 120 and 160 mg N kg⁻¹ soil in the conditions of optimal moisture, by 21 and 37%; for doses of 120 and 80 mg N kg⁻¹ soil and water deficiency stress, by 12 and 14%. Mineral nitrogen fertilization had a stronger effect on nitrogen in dry root mass yield compared with that in dry aboveground mass. The negative effect of water deficiency stress on nitrogen in dry aboveground and root mass was lowest when 80 mg N kg⁻¹ was applied to the soil.

Key words: nitrogen in yield, lucerne, mineral nitrogen fertilization, water deficiency stress

INTRODUCTION

Lucerne has the ability to utilize significantly more nitrogen than other legumes through its deep rooting characteristics; it derives 40–70% of total plant nitrogen through symbiosis (Heichel et al., 1981; Jarvis, 2005). Because of its nitrogen-fixing ability the issue of the additional introduction of nitrogen is debatable in the literature (Oliveira et al., 2004; Werner& Newton, 2005).

The question becomes even more acute under conditions of water deficiency stress (Busse & Bottomley, 1989; Vasileva et al., 2006a; Kirova, 2006). The aim of this work was to study the effect of mineral nitrogen fertilization and water deficiency stress on nitrogen in the yield in dry aboveground and root mass of lucerne grown for forage.

MATERIALS AND METHODS

An experiment with lucerne variety 'Victoria' was carried out at the Institute of Forage Crops, Pleven, Bulgaria (2003–04), using pots of 10 L capacity and soil subtype of leached chernozem. Sowing was conducted by hand at a depth of 2–3 cm with about 20 germinable seeds. After emergence four well developed plants per pot remained. The following treatments were tested with four replications.

Under optimum water supply – (75-80% of Field Capacity) (FC): 1. Control 1- unfertilized- N0PK (C1); 2. Soil + 40 mg N kg⁻¹ soil (N40PK); 3. Soil + 80 mg N kg⁻¹ soil (N80PK); 4. Soil + 120 mg N kg⁻¹ soil (N120PK); 5. Soil + 160 mg N kg⁻¹ soil (N160PK). Under 10-day water deficiency stress (37-40% FC): 6. Control 2unfertilized- N0PK (C2); 7. Soil + 40 mg N kg⁻¹ soil (N40PK); 8. Soil + 80 mg N kg⁻¹ soil (N80PK); 9. Soil + 120 mg N kg⁻¹ soil (N120PK); 10. Soil + 160 mg N kg⁻¹ soil (N160PK).

For treatments 1 to 5, 75–80% of FC was maintained, and for 6 to 10, water deficiency stress was provoked at the stage of budding of the lucerne by interrupting the irrigation until soil moisture was reduced to 37–40% of FC. Mineral nitrogen as ammonium nitrate equivalent was applied to the tested doses (a triple super phosphate and potassium chloride - as a background). Two cuts for forage were harvested each year.

Nitrogen in dry aboveground and root mass was calculated as a product of dry aboveground and root mass yield and total nitrogen content (determined by standard method) in dry mass. The total content of nitrogen in yield was calculated as the sum of that in aboveground and root mass (dried at 60 °C). Reduction in percentage of nitrogen in yield after water deficiency stress was determined using the following formula:

Reduction in N in yield $(\%) = \frac{N \qquad N \text{ in yield after}}{N \text{ in yield in control} - nitrogen fertilization} \times 100$ N in yield in control

The data from two experimental years were averaged and statistically processed using the SPSS 10.0 computer program.

RESULTS AND DISCUSSION

Nitrogen fertilization is essential to create optimal conditions for intensive fixation and nitrogen accumulation in plants (Evstigneeva &Pushkin, 1983). The accumulation of nitrogen in yield is an important indicator of legumes and in lucerne, in particular (Bliss, 2003).

Data in Table 1 show that nitrogen in the dry aboveground mass at optimal moisture increased as doses of mineral nitrogen fertilization increased. When 120 and 160 mg N kg⁻¹ soil were applied, nitrogen in the dry aboveground mass, relative to that of the unfertilized control, was increased by 21 and 37%, respectively.

Mineral nitrogen fertilization at the doses of 40 and 160 mg N kg⁻¹soil under water deficiency stress did not affect nitrogen in the dry aboveground mass yield. The

increase compared to unfertilized control was 12 and 14% at the doses of 120 and 80 mg N kg⁻¹soil, respectively. Given that the effect of mineral nitrogen localized on the roots, rather than on the whole nitrogen nutrition of the plant, nitrogen in the dry root mass yield increased significantly. Nitrogen in the dry root mass yield relative to that of the unfertilized control varied between 15 and 58% for the conditions of optimal moisture, and between 42 and 81% for water deficiency stress.

Treatments	Nitrogen in yield			
	Dry aboveground mass	To C1C2	Dry root mass	To C1C2
	mg N kg ⁻¹ dry mass	%	mg N kg ⁻¹ dry mass	%
	Optimal moisture (75-80% FC)			
Control1 PK	417.8	-	170.0	-
N40 PK	436.1	+ 4	195.1	+ 15
N80 PK	449.6	+ 8	200.3	+ 18
N120 PK	505.1	+ 21	261.9	+ 54
N160 PK	571.1	+ 37	268.8	+ 58
SE ($p = 0.05$)	27.9		19.5	
	W	Water deficiency stress (37-40% FC)		
Control2 PK	329.7	-	128.6	-
N40 PK	327.7	- 1	182.2	+ 42
N80 PK	376.2	+ 14	189.7	+ 47
N120 PK	368.3	+ 12	233.0	+ 81
N160 PK	337.3	+ 2	209.3	+ 63
SE ($p = 0.05$)	10.1		17.3	

Table 1. Nitrogen in dry aboveground and root mass of lucerne for forage after mineral nitrogen fertilization (on average for 2003–04).

With increasing the doses of nitrogen fertilization under optimal moisture, nitrogen in the dry aboveground and root mass increased progressively, as the differences for all treatments were significant (Fig. 1). Exceeding to unfertilized control varied from 7 to 43%.

According to Antolin et al. (1995) crops treated with nitrogen which have been subjected to water deficiency stress had better growth and better productivity as compared to those depending only on nitrogen from symbiotic N_2 -fixation.

In our study the relationship between mineral nitrogen quantities and nitrogen in yield under optimal moisture (R^2 =0.976) was greater, compared to the same under water deficiency stress (R^2 =0.923). Nitrogen in yield under water deficiency stress increased with increasing doses of fertilization to 120 mg N kg⁻¹soil.

Although lucerne is more tolerant for drought than most forage legumes, its dry mass productivity, as well as other characteristics, such as nitrogen in yield, is influenced negatively by water deficiency stress, particularly in the year of establishment (Frame et al., 1998; Zahran, 1999, 2001; Humphries & Auricht, 2001).



Figure1. Curve trend, equation and prognostic effect of the relationship between mineral nitrogen fertilization and nitrogen yield in lucerne for forage.

Treatments	Nitrogen in yield				
	Dry aboveground mass	Dry root mass	Total (dry aboveground + dry root mass)		
Control1 PK	21	24	22		
N40 PK	25	7	19		
N80 PK	16	5	13		
N120 PK	27	11	22		
N160 PK	41	22	35		

Table 2. Percent of reduction in nitrogen in dry mass yield after water deficiency stress.

The results of our study show that nitrogen in the dry aboveground mass under water deficiency stress decreased to 41% for the highest trial dose (Table 2). When 80 mg N kg⁻¹soil was applied the decrease was lowest (16%). Nitrogen in the dry root mass decreased less compared to that in the dry aboveground mass: it was 5 and 7% for the doses of 80 and 40 mg N kg⁻¹soil, and 11 and 22%, for 120 and 160 mg N kg⁻¹soil, respectively. For the untreated plants the decrease was by 24%. Nitrogen in dry aboveground and root mass decreased by 13% at the dose of 80 mg N kg⁻¹soil, which corresponds to the whole development of lucerne (Vasileva et al., 2006b).

CONCLUSIONS

Mineral nitrogen fertilization increased nitrogen in dry aboveground mass yield – for doses of 120 and 160 mg N kg⁻¹ soil in the conditions of optimal moisture, by 21 and 37%; for doses of 120 and 80 mg N kg⁻¹soil and water deficiency stress, by 12 and 14%. Mineral nitrogen fertilization had a stronger effect on nitrogen in dry root mass yield, and nitrogen in the dry root mass yield relative to that of the unfertilized control varied between 15 and 58% for the conditions of optimal moisture, and between 42 and 81% for water deficiency stress.

Water deficiency stress had a stronger effect on nitrogen in dry aboveground mass compared to that in dry root mass. For the doses of 80 and 40 mg N kg⁻¹soil, the decrease in nitrogen in dry root mass yield was by 5 and 7%, and for 120 and 160 mg N kg⁻¹soil, by 11 and 22%, respectively. Water deficiency stress had the strongest effect on unfertilized plants, reducing nitrogen in dry root mass yield by 24%. Mineral nitrogen, applied at the dose of 80 mg Nkg⁻¹soil, was optimal for the lucerne development where water deficiency stress effects had the lowest impact on nitrogen in dry aboveground and root mass.

REFERENCES

- Antolin, M.C., Yoller, J. & Sanches-Diaz, M. 1995. Effects of temporary drought on nitrate-fed and nitrogen-fixing alfalfa plants. *Plant Science***107**,159–165.
- Bliss, F.A. 1993. Breeding common bean for improved biological nitrogen fixation. *Plant Soil* **152**,71–79.
- Busse, M. D.& Bottomley, P. J.1989. Growth and nodulation responses of *Rhizobium meliloti* to water stress induced by permeating and nonpermeating solutes. *Appl. Environ. Microbiol.* 55, 2431–2436.
- Evstigneeva, Z.G. & Pushkin, A.V. 1983. In: Kretovich, W.L. (ed.) Molecular mechanisms of nitrogen assimilation in plants, pp. 198–234.
- Frame, J.J., Charlton, J.F. & Laidlow, A.S. 1998. *Temperate Forage Legumes*. Cab International, Wallingford, UK. 107–159.
- Heichel, G., Barnes, D.& Vance, C. 1981. Nitrogen fixation of alfalfa in the seeding year. *Crop Sci.* **21**, 330–335.
- Humphries, A. & Auricht, G. 2001. Breeding lucerne for Australia's southern dryland cropping environments. *Aust. J. Agric. Res.* **52**, 153–169.
- Jarvis, S. C. 2005. N flow and N efficiency in legumes based systems: a system overview. In: Wachendorf, M., Helgadottir, A. & Parente, G. (eds) Sward dynamics, N-flows and Forage Utilisation in Legume-Based Systems. Proc. of the 2ndCOST 852 Workshop, Grado, Italy 10–12 November 2005, pp. 187–198.
- Kirova, E. 2006. Response of nitrate-fed and nitrogen-fixing legume plants to water stress. *Plant Science* **44**, 13–17 (in Bulgarian).
- Oliveira, W., Oliveira, P., Corsi, M., Duarte, F.& Tsai, S. 2004. Alfalfa yield and quality as function of nitrogen fertilization and symbiosis with *Sinorhizobium meliloti*. *Sci. Agric.* (Piracicaba, Braz.) **61**, 433–438.
- Vasileva, V., Kostov, O. & Vasilev, E. 2006. (a) Development of lucerne (*Medicago sativa* L.) treated with mineral fertilizer and manure at optimal and water deficit conditions. *Commun. Agric. Appl. Biol.Sci.* **71**(4), 5-17.

- Vasileva, V., Vasilev, E. & Kostov, O. 2006. (b) Effect of water deficiency stress on nitrogen yield of lucerne under mineral and organic fertilisation. In: Wachendorf, M., Helgadottir, A. & Parente, G. (eds) Sward dynamics, N-flows and forage utilisation in legume-based systems. Proceedings of the 2nd COST 852 Workshop held in Grado, Italy 10–12 November 2005, pp. 173–175.
- Werner, D. & Newton, W.E. 2005. Nitrogen fixation in agriculture, forestry, ecology, and the environment. Springer. 349 pp.
- Zahran, H.H. 1999. *Rhizobium*-legume symbiosis and nitrogen fixation under severe conditions and in an arid climate. *Microbiol. Mol. Biol. Rev.* 63, 968–989.
- Zahran, H.H. 2001. Rhizobia from wild legumes: diversity, taxonomy, ecology, nitrogen fixation and biotechnology. *J. Biotechnol.* **91**, 143–153.