NITROGEN FERTILIZATION OF ALFALFA (*MEDICAGO SATIVA* L.) IN LATE WINTER IN MEDITERRANEAN ENVIRONMENTS

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SUMMARY

Fertilizing established alfalfa with nitrogen is a practice that is generally questioned. However, in cold springs, some alfalfa growers believe that N fertilization might increase the rate of spring regrowth. To evaluate the effects of N fertilization in spring, field experiments were conducted during four years under irrigation in a Mediterranean environment. The treatments were 0 and 30 kg ha^{-1} of N applied annually in late February on different alfalfa varieties. N fertilization increased the forage dry matter (DM) yields in the first spring harvest in one experiment, from 3.74 to 4.21 Mg ha^{-1}, whereas the proportion of weeds in the DM, in this harvest, rose from 22.8% to 32.8%. No DM yield differences between treatments were found in any of the other five harvest of the year or in the total annual DM yield. When weeds were controlled by herbicide, the N fertilization in late winter did not affect the herbage DM yields, weed proportion of the herbage, plant height or crude protein concentration of the forage. The results suggest that in the conditions of the experiments, the yield improvement with N fertilization was due to an increase in the weed content (from 22.8% with 0 N to 32.8% with 30 kg/ha N).

Key words: DM spring yields, weeds.

INTRODUCTION

Fertilizing established alfalfa (*Medicago Sativa* L.) stands with nitrogen is a practice that is generally questioned, especially if the plants are properly nodulated. Evidence in the literature does not appear to justify N fertilization to increase herbage yield (Lee and Smith, 1972; Hojjati *et al.*, 1978; Lamb et al., 1995), but there are few data available to support or contradict the use of this practice (Lee and Smith, 1972; Hannaway and Shuler, 1993), and some reports (Nuttall, 1985; Raun *et al.*, 1999) indicate yield improvement associated with N fertilization of established stands of alfalfa.

Hannaway and Shuler (1993) reported highly significant N and temperature interactions in alfalfa, and stated that the application of 18 kg N/ha was likely to provide benefits in low-N cool soils, when biological nitrogen fixation is reduced after harvest or if inoculation failure occurs. On the other hand Raun *et al.* (1999) in Oklahoma, reported alfalfa DM yield increases due to N fertilization but only in late season harvests, with the decrease in N_2 fixation capacity of the alfalfa, whereas Nuttall (1985) in Canada found that a single N application (45 kg N ha^{-1}) in early spring significantly increased alfalfa DM production compared with no N application, but that this practice was not economical.

The effect of N fertilizer on alfalfa in stimulating weed growth and competition has been reported (Fishbeck and Phillips, 1981). According to Hannaway and Shuler (1993) nitrogen fertilization of alfalfa has been a controversial practice. Many studies about the effects of N on alfalfa have been conducted in synthetic growth media and at optimal growth temperatures, but these results may not be directly applicable to field conditions where temperature, moisture, and nitrate levels are variable.

The effect of low temperatures in the spring regrowth of alfalfa has also been studied by Cralle and Heichel (1982). They reported that from a practical viewpoint, chilling injury of alfalfa nodules may be an important limitation to the onset of nitrogen fixation activity in the spring in overwintering alfalfa nodules, since it is known that low temperatures reduce nitrogenase activity.

Svenning and Macduff (1996), working with white clover, reported poor growth during spring due to low temperature, which is a constraint on the agronomic value of white clover at northern latitudes, and stated that physiological processes differ in their intrinsic sensitivities to temperature. For white clover there is evidence that N_2 fixation is less sensitive than shoot growth to root temperatures in the range 13-23^{O}C. In contrast, it may be more sensitive than the uptake of mineral N to inhibition at low root temperature.

At present, the Ebro Valley (Spain) is one of the main alfalfa producing areas of Europe, with about 145 000 ha, where the crop is produced for dehydration (Ollé, 2001). In this region it is a common practice of the dehydration industries to apply a small amount of N fertilizer in late winter in order to increase the rate of spring regrowth and thus increase alfalfa yields in the first harvest. They are concerned that cold spring temperatures might reduce spring regrowth (Alvaro and Lloveras, 2003).

The objective of this study was to evaluate the effects of small amounts of N fertilization in late winter on the yield and quality of alfalfa in the first spring harvest in irrigated continental Mediterranean conditions.

MATERIALS AND METHODS

Two field experiments were conducted in irrigated conditions over four growing seasons: Experiment 1 in 1997 and 1998 and Experiment 2 in 2000 and 2001 in the University of Lleida (UdL) - IRTA research fields (41°39'N, 0°51'E), in Ebro Valley (Spain) on Calcixerolic Xerochrept soils. Soil analysis of the two field experiments are presented in Table 1. Mean winter and annual temperatures and rainfall for the growing seasons and long-term averages are presented in Table 2.

TABLE 1

Initial soil analyses at two depths (0-30 and 30-60 cm).

Analisis de suelo al inicio de los ensayos en dos profundidades (0-30 y 30-60 cm).

	Experiment 1		Experiment 2	
	0-30 cm	30-60 cm	0-30 cm	30-60 cm
pH (water)	8.3	8.3	8.3	8.2
Organic matter (g kg^{-1})	19	12	25	16
N-NO_3- (mg kg^{-1})	6	7	56	56
Available P† (mg kg^{-1})	32	8	46	27
Available K‡ (mg kg^{-1})	226	73	424	146
$CaCO_3$ (g kg^{-1})	350	480	260	380

† Olsen method, ‡ Ammonium acetate method.

TABLE 2.

Mean (T_m) and maximum (T_{max}) air temperatures (°C) and rainfall (mm) during the experimental period.

Temperaturas medias (T_m) y máximas (T_{max}) y lluvia caída durante los ensayos.

	1996		1997		1998		1999		2000		2001	
Month	T_m	Rainfall	T_m	Rainfall	T_m	Rainfall	T_m	Rainfall	T_m	Rainfall	T_m	Rainfall
January	-	-	5.1	91.0	6.2	21.5	-	-	1.3	3.4	6.8	21.4
February	-	-	7.2	3.0	6.6	17.0	-	-	8.7	0.2	6.3	4.2
March	-	-	9.8	0.0	10.1	7.8	-	-	9.9	26.5	13.0	36.5
April	-	-	12.3	33.1	11.6	55.3	-	-	12.4	53.1	13.2	62.1
May	-	-	16.9	24.4	16.5	24.4	-	-	18.0	47.6	17.0	53.7
November	7.8	82.1	5.6	10.2	-	-	5.6	45.9	8.7	44.8	-	-
December	6.2	63.9	2.9	40.4	-	-	4.2	4.3	7.2	39.9	-	-

Long term mean annual temperature and rainfall for the area were 15.5 °C and 368 mm, respectively.

The experimental design was a split-plot in space and time (Steel and Torrie, 1980) with four replications. The N treatments were the main plots and the alfalfa cultivars the subplots. N was applied as NO_3NH_4, and consisted of one annual broadcast application of 30 kg N ha^{-1} once a year in late winter before the start of spring regrowth, and a control treatment without N fertilization. The dates of application were 3 March 1997, 25 February 1998, 28 February 2000, and 14 February 2001 (Table 3). The N treatment was broadcast on six alfalfa cultivars in experiment 1 ('Ampurdán', 'Aragón', 'Artal', 'BarMS82', 'Capitana', and 'Europe'), and on four cultivars in experiment 2 ('Altiva', 'Aragón', 'BarMS82', and 'Capitana'). The varieties had a dormancy ratings of 8 ('Altiva', 'Ampurdán', 'Aragón', 'Artal', 'Capitana') and lower, 7 ('BarMS82') and 4.5 ('Europe') (Lloveras et al., 1999; Montegano *et al.*, 2002).

TABLE 3

Dates of the N fertilizer applications and main determinations.

Fechas de la aplicación de N y de las principales determinaciones.

Determination	Year 1997	1998	2000	2001
N fertilization	3 March	25 February	28 February	14 February
Plant height	21 March	18 March	22 march	23 march
1st harvest	17 April	7 abril	13 April	19 April

At seeding, alfalfa was fertilized with 30 kg N ha^{-1}, 90 kg P ha^{-1} and 120 kg K ha^{-1} (Table 3). The alfalfa also received an annual maintenance application of 90 kg P ha^{-1}, and 120 kg K ha^{-1}. Experiment 1 was seeded on 2 October 1995, and Experiment 2 on 3 March 1999. At the time of the treatments, the alfalfa was in the second year and third year of production.

The seed rate of both experiments was 30 kg ha^{-1} in rows 20 cm apart. Subplots were 1.5 m x 6 m and were irrigated every 12 to 16 days from March to September receiving a total of about 900 mm of water per growing season.

Alfalfa yield was determined by harvesting the whole plot. Six cuttings were given each year at the mid to full-flowering state, except for the first and the last cut of each year in which the crop does not flower because of the short photoperiod. The first harvest was about mid-April, and this harvest is the objective of these experiments. The last cut was at the end of October with a period of about 30 days between harvests (Lloveras *et al.*, 1998).

Insects were controlled by spraying 0.1kg ha^{-1} a.i. fenvalerate [cyano(3-phenoxyphenyl)methyl4-cholo-alpha-(methylethyl)benzeacetate], two to four times per year and weeds were controlled in 2000 and 2001 in Gimenells (Experiment 2), by applying 1 kg ha^{-1}a.i. of hexazinone [3-cyclohexyl-6-dimethylamino-1-methyl-1,3,5-triazine-2,4 (1H,3H)-dione] in January.

Plant height was determined before the first spring harvest and before the second harvest (experiment 1) by taking three measurements per plot (Table 3). The proportions of alfalfa and weeds were evaluated just before harvesting, by cutting 2 (0.4 m x 0.6 m) squares for each individual plot, and separating alfalfa and weeds by hand in the laboratory, although the proportion of the different weed species was not determined.

A 200g wet sample of herbage was collected from each plot at each harvest for moisture determination and subsequent determinations. Samples were dried at 70°C and DM yields and proportions of alfalfa and weeds were calculated on these basis. Total N was analyzed by NIRS (Bran+Luebbe. InfraAlyzer 500) calibrated for alfalfa. Crude protein (CP) was calculated as N x 6.25.

Soil samples for the determination of the nitrates NO_3^-N were taken on 8 points of the field (2 per plot) the day of N application, and the average initial NO_3^-N contents were 6, 35, 56, and 32 mg NO_3^-N kg^{-1} in 1997, 1998, 2000, and 2001, respectively.

The results of each experiment were analyzed separately and subjected to analysis of variance with the General Linear Model procedure of the SAS procedures (SAS Institute, 1996). In the analyses of variance, varieties, treatments and years were considered fixed, and replications random. Data were analysed individually for each year according to the model:

$$Y_{i,j,k} = \grave{I}+V_i+R_j+\hat{A}_{(ij)}+T_k+TV_{ki}+\hat{A}_{(ijk)}$$

Were $\grave{I}$ is the overall mean, V_j is the variety, R_j the replication effect in each field, $\hat{A}_{(ij)}$ or Error a, T_k is the nitrogen Treatment effect, TV_{ki} is the interaction effect between nitrogen Treatments and alfalfa Varieties, and $\hat{A}_{(ijk)}$ is the error term. Finally the combined analysis of years was carried out according to the model:

$$Y_{i,j,k,l} = \grave{I}+V_i+R_j+\hat{A}_{(i,j)}+T_k+TV_{ki}+ \hat{A}_{(ijk)}+Y_l+YV_{il}+YT_{lk}+YVT_{ikl}+\hat{A}_{(ijkl)}$$

Were $\grave{I}$ is the overall mean, V_i is the variety effect, R_j is the replication effect, $\hat{A}_{(Ij)}$ or Error a. Moreover, T_k is the nitrogen treatment effect, $T\ V_{ki}$ is the interaction effect between Treatments and alfalfa Varieties, $\hat{A}_{(ijk)}$ or error term is the interaction between V_i, R_j, and T_k. Y_l *is the year* effect, $Y\ V_{il}$, is the interaction between years and alfalfa varieties, $Y\ T_{lk}$, is the interaction between years and N treatments, $Y\ V\ T_{ilk}$, is the interaction between Years, Varieties and N treatments and $\hat{A}_{(ijkl)}$ is the error term.

RESULTS AND DISCUSSION

Experiment 1

N fertilization at the end of winter increased the forage DM yields in the first spring harvest (Table 4). The average DM yields increased from 4.2 to 4.44 Mg ha^{-1} in 1997, and from 3.29 to 3.98 Mg ha^{-1} in 1998. The average dry matter yield of the varieties in the first harvest were 4.93, 4.61, 4.30, 4.19, 3.31, and 2.46 Mg ha^{-1} for 'Ampurdán', 'Artal', 'Capitana', 'Aragón', 'BarMS82' and 'Europe', respectively. No N x cultivar interaction was observed even if the varieties used in the experiment belonged to different alfalfa fall dormancy ratings and consequently have different rates of spring regrowth. No differences were found in plant height at any of the measurements and no interaction between N and cultivar was observed (Table 4).

TABLE 4

Forage dry matter (DM) yields, crude protein contents and plant heigth of the first and second harvest of the year, weed contents on the first harvest, and total annual dry matter yields. Average of six varieties. Experiment 1.

Producción de materia seca de forraje (DM), contenido en proteína bruta y altura de planta en el primer y segundo cortes del año, contenido de adventicias en el primer corte de cada año y producción total anual de materia seca. Media de seis variedades. Experimento 1.

		1st harvest								2nd harvest						Annual yield	
Treatment (kg ha^{-1})		DM yield (kg ha^{-1})		Crude protein (g kg^{-1} DM)		Weed contents (%)		Plant heigth (cm)		DM yield (kg ha^{-1})		Crude protein (g kg^{-1} DM		Plant heigth (cm)		Total DM yield (kg ha^{-1})	
		1997	1998	1997	1998	1997	1998	1997	1998	1997	1998	1997	1998	1997	1998	1997	1998
0 N		4.20	3.29	18.6	20.7	20	25	31.8	17.7	4.22	4.73	20.4	22.8	33.8	32.7	23.0	20.5
30 N		4.44	3.98	18.9	20.3	30	035	32.5	17.9	4.23	4.84	21.1	22.0	34.3	33.0	22.7	21.1
Significance		*	**	NS	NS	*	**	NS	NS	NS	NS	*	NS	NS	NS	NS	NS
ANOVA over years																	
	d.f.																
Variety	5	**		NS		*		**		**		**		**		**	
Replication	3	*		*		NS		NS		NS		NS		NS		*	
Error a	15	-		-		-		-		-		-		-		-	
N	1	**		NS		**		NS		NS		NS		NS		NS	
N x Var	5	NS		NS		NS		NS		NS		NS		NS		NS	
Error b	18	-		-		-		-		-		-		-		-	
Year	1	**		**		*		**		**		**		NS		**	
Year x Var	5	*		NS		NS		**		*		NS		NS		NS	
Year x N	1	NS		NS		NS		NS		NS		NS		NS		NS	
Year x N x Var	5	NS		NS		NS		NS		NS		NS		NS		NS	
Error	36	-		-		-		-		-		-		-		-	

On the other hand, N fertilization significantly increased the proportion of weeds in the DM which rose, on average, from 22.8% to 32.8% and beeing the most common weeds of the trials *Capsella bursa-pastoris* (L.) Med, *Taraxacum officinale* Weber, *Poa annua* L., *Rumex cripus* L., *Stellaria media* (L.) Vill., and *Lamium purpureum* L. These results suggest that the increase in herbage DM yields observed in the first harvest was primarily due to an increase in the proportion of weeds instead of an increase in the alfalfa. Weed growth increase when alfalfa is fertilized with N has been noticed by others in different climatic areas (Fishbeck and Phillips, 1981; Hannaway and Shuler, 1993).

No effects of the N fertilization were observed in any other harvest of the season and in the total annual DM production in any year. This type of response can be considered normal because, as suggested by Hannaway and Shuler (1993), the possible yield increases in spring could be mainly due to relatively cool (less than 15°C) low N soils (<15 mg of NO_3^- N kg^{-1}). In our case, the nitrate content of the soil was low in 1997 (6 mg of NO_3^- N kg^{-1}), and might have been sufficient in 1998 (35 mg of NO_3^- N kg^{-1}). N fertilization did not affect CP content of the herbage, in any of the two first harvests of the year.

Experiment 2

In this experiment, herbicide was applied in winter to control weeds, and as a consequence the proportion of weeds in the DM in the first spring harvest was low, between 2.5% and 3.7%, being the the most common weeds of the trials were *Capsella bursa-pastoris* (L.) Med, *Diplotaxis Erucoides* L., *Sonchus oleraceus* L., and *Rumex* cripus L.

In this experiment, N fertilization in late winter did not affect the herbage DM yields in the first harvest of the growing season and in the annual total DM yield (Table 5), did not affect either the weed content of the herbage, nor plant height or CP content of the forage. These results suggest that in our Mediterranean conditions, although average air temperatures in March and April are below 15°C (Table 1) they seem adequate, and the spring growth of alfalfa is not normally delayed. In this experiment the nitrate content of the soil at the beginning of the 2000 growing season was considered adequate (56 mg of NO_3^- N kg^{-1}), and might have been also sufficient in 2001 (31 mg of NO_3^- N kg^{-1}).

The average dry matter yield of the varieties in the first harvest were: 2.86, 2.65, 2.45, and 2.43 Mg ha^{-1}, for 'San Isidro', 'BarMS82', 'Aragón', and 'Altiva', respectively.

TABLE 5

Forage dry matter yield (DM), crude protein contents, plant heigth and weed contents on the first harvest of the year and total DM yields. Average of four varieties. Experiment 2.

Producción de materia seca de forraje (DM), contenido en proteína bruta, altura de planta y contenido de adventicias en el primer corte de cada año y producción total anual de materia seca. Media de cuatro variedades. Experimento 2.

Treatment ($kg\ ha^{-1}$)		DM yield ($kg\ ha^{-1}$)		Crude protein ($g\ kg^{-1}$ DM)		Weed contents (%)		Plant heigth (cm)		Total DM yield ($kg\ ha^{-1}$)	
		2000	2001	2000	2001	2000	2001	2000	2001	2000	2001
0 N		3.00	2.13	169	224	4	3	39.30	29.43	18.0	14.7
30 N		2.87	2.23	175	223	2	2	40.38	31.72	17.4	14.9
Significance		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
ANOVA over years											
	d.f.										
Variety	3	NS		NS		NS		NS		NS	
Replication	3	*		*		NS		NS		NS	
Error a	9	-		-		-		-		-	
N	1	NS		NS		NS		NS		NS	
N x Var	3	NS		NS		NS		NS		NS	
Error b	12	-		-		-		-		-	
Year	1	**		**		NS		**		**	
Year x Var	3	NS		NS		NS		NS		NS	
Year x N	1	NS		NS		NS		NS		NS	
Year x N x Var	3	NS		NS		NS		NS		NS	
Error	24	-		-		-		-		-	

CONCLUSIONS

In the Mediterranean conditions of the experiments the first spring harvest is performed in mid-April and the air temperature of the area rose from an average of 7.2°C in February to 10.7 °C in March and to 12.4 °C in April. With these conditions, the results suggest that the effects of the N fertilization on the DM yield increase of the first spring harvest of alfalfa was due to an increase in the weed content associated with the use of N, and not to the effect of N fertilizer to the alfalfa.

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EFECTO DE LA FERTILIZACIÓN NITROGENADA DE LA ALFALFA (*Medicago sativa* L.) AL FINAL DEL INVIERNO, EN CLIMAS MEDITERRÁNEOS

RESUMEN

La fertilización nitrogenada de la alfalfa es una práctica generalmente cuestionada. Sin embargo, en algunas zonas con primaveras frías, algunos productores creen que la aplicación de N puede incrementar la producción del primer rebrote de primavera. Para evaluar los efectos de la fertilización nitrogenada al final del invierno se llevaron a cabo, diversos ensayos de campo, en los que en algunos se aplicaron herbicidas para eliminar las adventicias y en otros no. Los ensayos se realizaron durante cuatro años en condiciones de regadío, en ambiente mediterráneo.

Los tratamientos que se estudiaron fueron 0 y 30 N kg ha^{-1}, aplicados anualmente a distintas variedades de alfalfa, al final del mes de febrero. La aplicación de N incrementó la producción de materia seca (MS) del primer corte en un ensayo pasando de 3,74 a 4,21 t/ha, mientras que la proporción de adventicias en este corte, aumentó del 22,8 % al 32,8 %. No se detectó ningún otro efecto en el resto de los cortes del año o en la producción total anual.

En los ensayos donde las adventicias se eliminaron, la aplicación de N al final del invierno no afectó a la producción de materia seca, ni a la proporción de adventicias, ni a la altura de planta, ni al contenido de proteína bruta.

Los resultados sugieren que, en las condiciones de los ensayos, el incremento de producción de MS debido a la fertilización nitrogenada fue debido al incremento del contenido y producción de adventicias (del 22,8 % con 0 N a 32,8 % con 30 N kg ha^{-1}).

Palabras clave: Adventicias, producción primaveral.