NITROGEN USE BY STANDARD HEIGHT AND SEMI-DWARF BARLEY ISOTYPES[1]

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ABSTRACT- Considering that a substantial amount of N applied in cereal production may be lost by leaching or erosion, the development of cultivars with increased efficiency to utilize N and more efficient N management practices are desirable. The objective of this experiment was to study the effect of 30, 60, 90 and 120 kg of N/ha on N use by four standard height semi-dwarf normal-mutant malting barley isotype pairs (Morex, Hazen, Norbert, Andre). Genotypic differences in N remobilization in the grain, remobilization efficiency, utilization efficiency and uptake efficiency and N use efficiency were observed. The standard isotypes with few exceptions had higher values for all of these N traits. Nitrogen remobilization in the grain increased with increasing N fertilization while N utilization and N use efficiency decreased in both standard and semi-dwarf isotypes. Nitrogen use and grain yield relations are discussed.

Index terms: *Hordeum vulgare*, nitrogen remobilization, nitrogen efficiency.

EFICIÊNCIA NO USO DE NITROGÊNIO DE GENÓTIPOS DE CEVADA DE ALTURA NORMAL E SEMI-ANÃ

RESUMO- Considerando que uma quantidade substancial do nitrogênio utilizado para produção de grãos de cereais pode ser perdida por lixiviação ou erosão, o desenvolvimento de cultivares com maior eficiência na utilização deste nutriente bem como um melhor manejo do mesmo são, sem dúvida, práticas desejáveis em uma agricultura auto-sustentável. O objetivo do presente trabalho foi de estudar o efeito de 30, 60, 90 e 120 kg/ha de N na eficiência de uso de nitrogênio de oito genótipos de cevada cervejeira (quatro com altura normal e quatro semi-anões, mutantes de cada um dos normais). Foram observadas diferenças entre os genótipos na remobilização e na eficiência da remobilização de nitrogênio para o grão na eficiência de utilização, absorção e uso de nitrogênio. De modo geral, os genótipos de estatura normal apresentaram valores maiores para todos os parâmetros de uso de N. A remobilização de nitrogênio para o grão aumentou com as doses crescentes de N enquanto a utilização e eficiência de uso decresceram, tanto nos genótipos de estatura normal como nos semi-anões. São discutidas as relações entre o uso de nitrogênio e o rendimento de grãos.

Termos para indexação: *Hordeum vulgare*, remobilização de nitrogênio.

INTRODUCTION

Efficiency of nitrogen use has been a major concern subsequent to the rise in price of N fertilizers in the early 1970's and increased contamination of ground water by nitrate. The development of cultivars with enhanced ability to utilize N to increase grain yield and more efficient N management are strategies of interest. Nitrogen uptake, nitrate reduction, and N remobilization are traits with potential to affect grain yield, and consequently are suitable for genetic improvement (Cregan & Berkum, 1984). Factors affecting N uptake include the degree of association between roots and soil, supply of N, requirement for N in the plant, efficiency of uptake by the roots and water status of

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the soil (Huffaker & Rains, 1978). The shoot probably determines the N requirement when N supply is not limiting and the uptake mechanism in the roots would be important. To the extent that root development, shoot growth and plant phenology are under genetic control, so too would nitrate uptake be genetically determined (Cox et al., 1985). There are contradictions in the literature concerning the relationship between root volume and plant height (Mackey, 1973; Lupton et al., 1974; Irvine et al., 1980; Pommer & Opptiz, 1986). Shorter or smaller volume of roots may affect a reduction in N uptake capability and smaller root volume has been implicated with semi-dwarf cereals (Mackey, 1973; Cholick et al., 1977; Pommer & Opptiz, 1986). Differences in nitrate uptake among genotypes within species has been shown (Rao et al.,1977; Chevalier & Schrader, 1977; Bloom & Chapin, 1981; Jackson et al., 1986; Pan et al., 1987; Konesky et al., 1989).

Much field research has been done on wheat trying to characterize genotypes with increased ability to use N to produce more grain (Cox et al., 1985; Sanford & Mackown, 1987; Dhugga & Waines, 1989). However, very little has been done with barley.

The objective of this experiment was to study the effect of N supply on N use by standard height and semi-dwarf barley isotypes.

MATERIAL AND METHODS

Field experiments were conducted in 1987 and 1989 at the Spillman Agronomy Farm, near Pullman Washington on a Palouse silt loam soil (fine-silty, mixed mesic Pachic Ultic Haploxeroll). The two experimental crop years were preceded by typical Palouse crops; peas (*Pisum sativum* L.) in 1986 and barley in 1988.

The design was a randomized complete block arranged in split-plots with four replications. The treatments were 4 N levels (30, 60, 90 and 120 kg N/ha) as main plots, and, 10 spring barley genotypes as sub-plots. The barley genotypes included 4 standard height semi-dwarf (SD) normal-mutant malting isotype pairs (Aydin, 1985; Ullrich & Aydin, 1988): (Morex and Morex SD (6-row); Hazen and Hazen SD (6-row); Norbert and Norbert SD (2-row); Andre and Andre SD (2-row); and 2 check cultivars: Steptoe (6-row) as a yield check and Klages (2-row) as a malting quality check. Morex also served as a 6-row malting quality check. The plot size was of 4 x 1.28 m and 4.8 x 1.28 m in 1987 and 1989, respectively.

Fertilizer was applied during planting with an experimental plot planter. Most of the N was placed 10 cm below the seed level but 10 kg N/ha was applied with the seed as starter with 12.5 kg P/ha and 8.7 kg S/ha. Ammonium nitrate was the N source. In 1989 an additional 45 kg P/ha, as triple superphosphate, and 20 kg S/ha, as calcium sulfate were broadcasted and incorporated into the soil prior to planting.

Plant samples were taken at growth stage 10.4 (3/4 of the heads emerged) and at stage 11.3 (kernel dough) (Large, 1954). The plants harvested at stage 11.3 were divided into vegetative tissue (culm+leaves+chaff) and grain. Dry weight was obtained after drying for 72 h at 55°C in a convection dryer. The vegetative samples were ground through a 1 mm sieve in a Wiley mill. The grain samples were ground through a 0.5 mm sieve in a Udy mill. Total N in the vegetative tissue and in the grain was determined by the micro-Kjeldahl method (Association of Official Agricultural Chemists, 1965).

From the parameters measured the following variables were derived:

N remobilization (mg/pl) = pre-anthesis N accumulation - shoot (leaf+culm+chaff) N yield at maturity.

Remobilization efficiency (%) = (N remobilization/pre--anthesis N accumulation) x 100.

The variation in N use efficiency (NUE) was evaluated according to the method of Moll et al. (1982):

NUE = N uptake efficiency x N utilization efficiency.

N uptake efficiency = total N in the plant/N supply to the plant (fertilizer + residual + mineralized).

N utilization efficiency = grain dry weight/total N yield at maturity.

Residual nitrogen was estimated by summation of NO_3^- and NH_4^+ available up to 0.90 m depth, before sowing. In 1987 and 1989 these values were 83 and 33 kg/ha, respectively. Mineralized nitrogen was estimated according to the Fertilizer guide (1975), which considers the erosion level, annual rainfall, yield and kind of the preceding crop. The mineralized N values were 114 and 74 kg/ha, respectively, in 1987 and 1989.

There was no effect of N or genotype on the stand (plant population) within years. This allowed for an estimate of the number of plants per area, which was used to calculate the amount of N supplied as fertilizer per plant, by averaging the stand of all plots in each year.

Taking logarithms of the NUE expression and denoting the first term of the equation (NUE) as Y; the second term (N uptake efficiency) as X_1 and the third term (N utilization efficiency) as X_2, the following equation results: $Y = X_1 + X_2$. The contribution of each X_i term to the sum of squares was calculated according to Moll et al. (1982): $Sx_iy/Sy^2 = (R_{YXi})\, St_{Xi}/St_Y$, in which R_{YXi} is the correlation coeficient between Y and X_i and St_{Xi} and St_Y are the standard deviation for the X_i component and for log N use efficiency, respectively, and $Sx_iy/Sy^2 = 1$.

Analyses of variance over years as well for individual years were conducted with MSTAT microcomputer statistical program (Michigan Statistic-C, 1989). As means comparisons of the various paramenters of interest were between two means, either isotypes of a pair or overall standard vs semi-dwarf isotypes the independent t-test was used.

RESULTS AND DISCUSSION

The analysis of variance over years indicated significant effects of N treatments and genotypes for N utilization, uptake and use efficiency. The treatment (N levels, genotypes) by year interactions for most of the variables measured were significant, indicating effects of one or more environmental factors. Most of the significant interactions between treatments and year were probably due to the differences in the initial NO_3^- levels in the soil. The incorporation of the pea crop as green manure in the summer 1986 and the higher spring temperatures in 1987 were factors that likely contributed to increased availability of N in the soil from N fixed by peas, by increased N mineralization potential and increased rate of mineralization through the season. El-Haris et al.(1983) reported a significant increase in the N mineralization potential of the soil after green-manured peas. Varvel & Severson (1987) largely attributed the variation observed in barley response across three years to the difference in the initial NO_3^-. Due to the significant interactions (treatment x year) the results are presented by year.

Morex SD and Hazen showed significantly higher N remobilization than their respective isotype pairs (Table 1) besides that Morex SD had a significantly lower N uptake after anthesis in relation to its isotype pair (Nedel et al., 1997). The other two isotype pairs were not different. No significant differences in remobilization were detected between the means of the four standard and semi-dwarf genotypes (Table 1). Nitrogen remobilization was lowest in Steptoe and so was N concentration in the grain (Nedel et al., 1997). This cultivar is known to have low grain protein (Tillman et al., 1991). The low remobilization capability may be a factor contributing to the characteristically low grain N or protein level in this cultivar. By increasing N fertilizer levels, there was a corresponding increase in N remobilization for both standard and semi-dwarf isotypes (Table 2). These results were similar to those reported by Tillman et al. (1991). Semi-dwarf and standard isotypes differences in N remobilization efficiency were observed in this study (Table 1). Morex, Hazen and Andre SD showed significantly higher N remobilization efficiency than their respective isotypes. When the mean of the four standard isotypes was compared with the mean of the semi-dwarf isotypes no difference between them was observed (Table 1). The larger remobilization efficiency of Andre SD over Andre contributed substantially to reducing the difference between the overall means of standard and semi-dwarf isotypes. By increasing the N availability a decrease on N remobilization efficiency was observed in both standard and semi-dwarf isotypes (Table 2).

There was a significant difference between isotypes within pairs in N utilization efficiency (Table 1). In 1987 only the standard isotype Morex did not have a significantly higher N utilization efficiency than its derived semi-dwarf mutant. In 1989 Hazen and Andre SD showed a significantly higher N utilization efficiency than their respective isotypes. The extraordinarily high N utilization efficiency of Andre SD and Steptoe was mainly due to the low total N accumulation of these two genotypes at the low N level without a corresponding reduction in the grain yield per plant. This caused an increase in the grain yield per plant/total N in the plant ratio. The N utilization efficiency mean over all standard isotypes was significantly higher than for the semi-dwarfs in both years (Table 1). A decrease in the N utilization efficiency with increasing N fertilizer was observed (Table 2).

TABLE 1. Mean values for N remobilization, remobilization efficiency, utilization efficiency, uptake efficiency and N use efficiency by genotypes averaged over N treatments.

Genotypes	Remobilization		Remobilization efficiency		Utilization efficiency		Uptake efficiency		N use efficiency	
	1987	1989	1987	1989	1987	1989	1987	1989	1987	1989
	mg/pl		%		Gw/Nt		Nt/Ns		Gw/NtxNt/Ns[1]	
Morex	-	30.4**	-	78.7**	37.4	47.8	0.73	0.57	27.3	26.9
Morex SD	-	38.1	-	74.8	36.8	45.0	0.75	0.62	27.6	27.2
Hazen	-	37.8**	-	80.7**	37.1**	49.3**	0.97**	0.60	35.9**	28.8**
Hazen SD	-	31.6	-	75.7	31.5	38.5	0.67	0.55	21.0	20.5
Norbert	-	40.5	-	78.5	33.5*	45.2	0.79*	0.70	26.4**	30.8**
Norbert SD	-	39.6	-	76.1	31.8	43.8	0.69	0.63	21.9	26.9
Andre	-	36.0	-	75.5**	35.3**	43.8**	0.58**	0.70**	20.2**	31.7**
Andre SD	-	36.3	-	83.0	33.5	51.7	0.46	0.51	15.3	25.7
Klages	-	40.4	-	76.6	35.0	45.9	0.66	0.68	23.2	30.4
Steptoe	-	27.6	-	75.3	43.6	56.2	0.75	0.58	32.7	32.1
Standard	-	36.2	-	78.4	35.8**	47.3**	0.77**	0.63*	27.5**	29.5**
Semi-dwarf	-	36.4	-	77.4	33.4	44.7	0.64	0.57	21.5	25.1

[1] Gw= grain weight; Nt= total N accumulation; Ns= N supplied per plant (fertilizer + residual + mineralizable).
*, ** Significant difference between isotypes within a genotype according to t-test at P= 0.05 and 0.01, respectively.

TABLE 2. Mean values combined over standard and semi-dwarf genotypes for N parameters for four N treatments[1].

Genotypes	1987 N_1	1987 N_2	1987 N_3	1987 N_4	Sig.	1989 N_1	1989 N_2	1989 N_3	1989 N_4	Sig.
				N remobilization (mg/pl)						
Standard	-	-	-	-		27.4	31.7	40.6	45.1	LC
Semi-dwarf	-	-	-	-		26.3	31.6	37.4	50.3	LQ
				N remobilization efficiency (%)						
Standard	-	-	-	-		80.3	78.0	77.1	78.2	LQ
Semi-dwarf	-	-	-	-		78.7	77.7	76.9	76.6	L
				N utilization efficiency (Gw/Nt)						
Standard	37.9	36.4	33.9	35.0	LQ	52.6	49.6	44.1	42.8	LQ
Semi-dwarf	33.6	34.8	32.9	32.3	LQ	50.3	46.9	44.1	37.6	LQ
				N uptake efficiency (Nt/Ns)						
Standard	0.79	0.74	0.77	0.77	NS	0.61	0.64	0.70	0.60	NS
Semi-dwarf	0.76	0.76	0.61	0.52	LQC	0.57	0.55	0.58	0.61	NS
				N use efficiency (Gw/Nt)x(Nt/Ns)						
Standard	29.9	27.2	25.9	26.8	LQ	31.1	30.8	30.4	25.5	LQC
Semi-dwarf	25.6	23.5	20.0	16.7	L	27.6	24.9	25.2	22.5	LC

[1] N_1, N_2, N_3 and N_4= 30, 60, 90 and 120 kg/ha of N, respectively; L= Linear, Q= Quadratic, C= Cubic; Gw= grain weight; Nt= total N assimilation; Ns= N supplied as fertilizer.

Nitrogen remobilization in the standard isotypes was less dependant on the total N accumulation as the N in the soil increased than the semi-dwarf isotypes (Table 1). Nitrogen remobilization efficiency was moderately negatively associated with total N accumulation, indicating that a smaller proportion of the N in the vegetative tissue at anthesis was remobilized when total N accumulation increased. Similar results were reported by Cox et al. (1986) in spring wheat.

All standard isotypes, except Morex, had significantly higher N uptake efficiency (total N in the plant/N supplied as fertilizer + residual + + mineralized per plant) than their derived semi--dwarfs (Table 1). With the exception of Morex, there was a strong tendency for the standard isotypes to have a higher N uptake efficiency than their respective mutants in 1989. N uptake efficiency was higher in the standard vs semi-dwarf isotypes overall in both years, indicating that a larger proportion of the N available in the soil was absorbed by the standard than by the semi-dwarf isotypes. The highest values observed in 1987 vs 1989 for N uptake efficiency, are an indication that those genotypes are more efficient in N uptake when the N availability in the soil is higher. The standard isotypes did not respond to N fertilization in N uptake efficiency (Table 2). The semi-dwarf isotype response was not consistent between years. They had a decrease in N uptake efficiency with increasing N fertilization in 1987 and no response in 1989. This decrease observed in 1987 was related with their relatively stable total N accumulation in the shoot across all N levels (Nedel et al., 1997).

Nitrogen use efficiency results were consistent between years. All standard isotypes, except Morex, had significantly higher N use efficiency (Table 1). Also the mean over all standard isotypes was significantly higher than over all semi-dwarf isotypes. In 1989, as in 1987, there was a reduction in N use efficiency with an increase in N fertilizer levels (Table 2). This reduction is explained mainly by the decrease in N utilization efficiency, one of the N use efficiency components, with increasing N fertilization (Table 2). These results were similar to those obtained by Moll et al. (1982) with maize.

N uptake efficiency contributed substantially more than N utilization efficiency to the sum of squares of N use efficiency at all N fetilization levels in 1987 and at the first three N fertilization levels in 1989 (Table 3). In maize, Moll et al. (1982) observed a very high contribution of N utilization efficiency at low N levels and very low contribution at higher N levels. In winter wheat using one N fertilizer level Sanford & MacKown (1987) observed a very small contribution of N utilization efficiency to the N use efficiency sum of squares. In contrast the contribution of N uptake efficiency was 60% in one year and 81% in the other. Dhugga & Waines (1989) observed a weak contribution to N utilization efficiency and no differences in the contribution among the three N fertilizer regimes used in spring wheat. It seems that to some degree these two N traits are affected by environmental influences and may

TABLE 3. Contribution of N utilization efficiency and N uptake efficiency to the N use efficiency sum of square.

Component ration	Log_{10}	Fraction of genotypes SS (Sx_1y/Sy^2)									
		1987					1989				
		N level (kg/ha)					N level (kg/ha)				
		30	60	90	120	Combined	30	60	90	120	Combined
N use efficiency (Gw/Nt)x(Nt/Ns)[1]	Y	-	-	-	-	-	-	-	-	-	-
N utilization efficiency (Gw/Nt)	X_1	0.179	0.150	0.124	0.158	0.174	-0.138	0.051	-0.067	0.554	0.190
N uptake efficiency (Nt/Ns)	X_2	0.821	0.850	0.876	0.844	0.826	1.138	0.949	1.067	0.446	0.810

[1] Gw= Grain weight; Nt= Total N accumulation; Ns= Nitrogen supplied per plant (fertilizer + residual + mineralizable).

vary among species. The analysis presented in the Table 3 indicates that genotype selection based on N uptake efficiency, will probably be more effective to improve the overall N use efficiency than selection based on N utilization efficiency, at least under the conditions the study genotypes were developed (relatively high N).

CONCLUSIONS

1. There are genotypic differences for N traits in barley.
2. N remobilization to the grain increases with increasing level of N fertilizer.
3. N remobilization efficiency, N utilization efficiency and N use efficiency decrease at the same rate with increasing N fertilizer levels.

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