

Nitrogen Use Efficiency Assessment under Intra- and Inter-Specific Competitions Stress

Amir Aynehband^{1*}, Sara Asadi² and Afrasyab Rahnama³

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¹Professor, Agronomy & Plant Breeding Department, Agriculture Faculty, Shahid Chamran University, Ahvaz, Iran

²MSc Student, Agronomy & Plant Breeding Department, Agriculture Faculty, Shahid Chamran University, Ahvaz, Iran

³Assistant Professor, Agronomy & Plant Breeding Department, Agriculture Faculty, Shahid Chamran University, Ahvaz, Iran

*Corresponding author: E-mail: aynehband@scu.ac.ir

Abstract

Improving nitrogen use efficiency (NUE) is an important objective of the agroecosystem management. A field experiment was conducted to study the effect of different competition patterns on nitrogen balance in the wheat agroecosystem. Experimental design was split-plot based on randomized complete blocks with four replications. The main plots consisted of different nitrogen rates (i.e. 0, 50, 100, 150 kg N ha⁻¹) and sub-plots consisted of four different competitiveness patterns including: no competition, intraspecific competition, interspecific competition and intra- and inter-specific competitions. Our results showed that NUE decreased with increasing N level, especially under higher N rates. N uptake efficiency (NUPE), N harvest index (NHI), N remobilization efficiency (NRE), N harvest efficiency (NHE) and N fertilizer utilization efficiency (NFUE) were lowest in the intra- and inter-specific competition treatments than others. The differences between each intra- and inter-specific competition treatments were not significant for NUPE, N balance index (NBI), NHI, NRE and NHE. We suggested that greatly increased suppression of weeds through increased crop density for reducing the use of herbicide can play an important role in a change in both crop yield and crop N use efficiency. This strategy increased NUE, N utilization efficiency and N reliance index (NRI). Moreover, higher intraspecific competition in most cases reduced NUPE, NBI and NHE but, had no significant effect on NHI, NRI and NFUE.

Keywords: Nitrogen balances; Nitrogen uptake; Plant density; Weed

Introduction

The high yields of today's modern wheat cultivars require the use of mineral fertilizers and chemical herbicides and fungicides, all of which lead to both higher production costs and a greater risk of environmental pollution. Increasing public awareness of this, along with growing consumer demand for healthier products, has led on one hand to greater criticism of this type of production and on the other hand to a greater emphasis on crops grown under integrated-management and organic systems. As defined by Moll *et al.* (1982), NUE is grain yield (GY) per unit of N available in the soil, and it is the product of N uptake

efficiency (NUPE, total shoot N/soil N supply) and N utilization efficiency (NUTE, GY/total shoot N). It is stated that there are genetic differences for NUPE and NUTE in wheat (*Triticum aestivum* L.)(Barraclough *et al.* 2010). Similar results were observed using 12 winter wheat genotypes in a pot experiment, in which N uptake and utilization efficiency affected the genotypic variations for NUE at low N levels, while N uptake was the main influencing factor at high N levels. For a given crop or cultivar, NUE decrease if more N is put because of the law of diminishing marginal returns (Raun and Johnson 1999). Therefore, interventions to increase NUE

and reduce N inputs will play an important role not only in reducing environmental risk but also in lowering production costs (Wang *et al.* 2011).

An assessment of agroecosystem N use should include indicators that evaluate: (1) agronomic practices and their influence on major soil and plant physiological processes that effect N use; (2) economic factors and the optimization of agronomic inputs to achieve crop performance goals; and (3) environmental consideration including the sustainability of the agricultural resources base and the potential for resources degradation (Huggins and Pan 1993). In fact, cereal producers are under pressure to increase yields and maintain profitability against a background of environmental constraints and high fertilizer costs. High yields require high inputs of N and excessive N can lead to pollution of water resources (Semenov *et al.* 2007). Moreover, wheat crops with increased NUE will be of economic benefit to farmers and will help to reduce environmental contamination associated with excessive inputs of N fertilizers (Fageria 2009). Developing cropping systems and management practices that improve the ability of crops to absorb N could minimize the potential for N losses (Moll *et al.* 1982). The varieties suited for integrated weed management should combine high N use efficiency with superior competitive ability against weeds. To this end, it is also necessary to take into account the fact that N application can significantly affect the competitive interactions between the crop and the weeds and that N application often increases the competitiveness of weeds more than that of the

crop (Giambalvo *et al.* 2010). Managing the increased competitive ability of crops with weeds is an important component of the integrated weed management system; effective fertilizer management may be able to reduce weed interference with crops (O'Donovan *et al.* 2001). Many studies have only investigated the effects of N application on weed abundance, particularly density and dry matter (Blackshaw *et al.* 2001). In many situations, particularly those with higher weed densities, added nutrients favor weed growth, often providing little added benefit in crop yield.

There has been relatively little consideration of NUE in intra- and inter-specific competition. In this view, the objectives of this study were to determine the effects of intra- and inter-specific competition on grain yield and nitrogen efficiency indices of wheat under different nitrogen fertilizer levels.

Materials and Methods

The trial was conducted at the experimental field of the Agriculture Faculty of Shahid Chamran University of Ahvaz, Iran (31°20' N latitude and 48°41' E longitudes) at an elevation of 20 m above mean sea level during the 2010 and 2011 wheat growing seasons. The soil type was sandy loam with pH 7.9 and a 0.53% average organic matter concentration. The 0-30 cm soil layers contained 0.043 % nitrogen, 15 mg kg⁻¹ phosphorus rate and 165 mg kg⁻¹ potassium. Seeds of Chamran cultivar were planted on 23 Nov. 2010. Four competition types including: (1) No competition (i.e. D₁: optimum planting density and W₀: no weed

density), (2) Intraspecific competition (i.e. D_2 : high planting density), (3) Interspecific competition (W_1 : high weed density), (4) Intra- and interspecific competition (i.e. D_2W_1 : high plant and weed density) were considered under four N-input regimes of 0, 50, 100 and 150 kg N ha^{-1} in a randomized complete block design with a split plot arrangement and four replications. Each plot consisted of 8 rows of 4 m length. Inter-row spacing was 20 cm and interplant spacing was 3 cm. Optimum and high planting densities were ~ 450 and ~ 750 plants m^{-2} , respectively. At anthesis and maturity, a one m^2 portion was also collected from the center of each plot to determine the N content of straw and grain (total dry matter) by standard macro-Kjeldahl procedure. Soil samples were taken on all plots, prior to wheat sowing and after wheat harvest, at a depth of 60 cm, and analyzed for nitrate content.

Soil and plant data collected as inputs for the procedure were: Nitrogen supply (N_s) which was estimated according to Huggins and Pan (1993) by the summation of aboveground plant N (N_t) in the control plots, postharvest soil nitrate (N_h) in control plots, and applied N (N_f). Several N efficiency traits were calculated for each treatment as shown in Table 1 (Moll *et al.* 1982; Cox *et al.* 1986; Huggins and Pan 1993; Ortiz-Monasterio *et al.* 1997; Delogu *et al.* 1998; Fageria 2009).

Analysis of variance was carried out to test the main effects of nitrogen rates and cultivars and their interaction. The Fisher LSD test ($P \leq 0.05$) was used to test differences between treatment means. The SAS PROC REG Procedure was used to calculate linear regressions between the nitrogen efficiency indices with the cultivar yield.

Results and Discussion

Nitrogen efficiencies

N use efficiency (NUE): This is defined as grain production per unit of N supply. NUE was significantly affected by competition types of release and N fertilizer rates. Nitrogen use efficiency diminished as N fertilizer rates increased ($r = 0.61$, $p < 0.0001$), but the increase was not significant at higher rates (N_{100} , N_{150}) (Table 2). NUE at the maximum N fertilizer rate of 150 Kg N ha^{-1} was 20.22 to 32.54 kg kg^{-1} which was obtained at 0 Kg N ha^{-1} . This variation of NUE was 37.86% between N rates. The decrease in NUE with increase in N fertilizer rate is due to the fact that grain yield rises less than the N supply in the soil and fertilizer. In other words, wheat plants grown at lower N rates actually produced lower yield due to N deficiency. Huggins and Pan (1993) reported that variation of NUE ranged from 19 to 32% kg G_w per kg N_s and was significantly influenced by N level. They found that N management and climate interactions had a large and variable effect on NUE. However, Lopez-Bellido and Lopez-Bellido (2001) with comparing of different N levels reported that NUE reduced as N fertilizer rates increased. The NUE index was significantly ($r = 0.61$, $p = 0.004$) higher for intraspecific competition ($D_2W_0 = 31.52$ kg kg^{-1}) than for other competition types (Table 2), due to the lower N_s rather than to difference in grain yield. The competition \times N fertilizer rate interaction showed that mostly N_{50} plots prompted significant ($r = 0.67$, $p = 0.0014$) differences between competition types with regard to the NUE index. The most immediate goal of improving agricultural NUE is to improve the recovery of N from fertilizer, either organic or synthetic. Reductions in NUE with increasing N

supply could result from reductions in any of the components, including NUPE, NUTE and NRE (Dawson Julie *et al.* 2008). For example, Oritz-Monasterio *et al.* (1997) found that in all varieties evaluated, both uptake and translocation/

utilization efficiency were reduced at the higher N supplies, causing an overall reduction in NUE. Cox *et al.* (1986) showed a reduction in translocation efficiency at high N compared with low N supplies.

Table 1. Components of nitrogen use efficiency for annual crops

Component symbol and descriptor
N_f = Applied N fertilizer
N_g = Grain N
N_h = Postharvest inorganic N
N_s = Nitrogen supply
N_t = Aboveground plant N at physiological maturity
N_{ta} = Aboveground plant N at anthesis
NA = Total aboveground N at maturity minus total aboveground N at anthesis
G_w = Grain yield
Y = Total biomass
G_w / N_s = NUE: N use efficiency (kg kg^{-1})
N_t / N_s = NUPE: N uptake efficiency (kg kg^{-1})
G_w / N_t = NUTE: N utilization efficiency (kg kg^{-1})
N_f / N_s = NRI: N reliance index (kg kg^{-1})
N_g / N_f = NBI: N balance index (kg kg^{-1})
N_g / N_t = NHI: N harvest index (%)
$(N_g - NA) / N_t$ anthesis = NRE: N remobilization efficiency (%)
N_g / N_s = NHE: N harvest efficiency (kg kg^{-1})
G_w / N_f = NFUE: N fertilizer utilization efficiency (kg kg^{-1})

N uptake efficiency (NUPE): This reflects the efficiency of the crop in obtaining N from soil. NUPE also varied significantly according to N rates; the significant ($r= 0.5$, $p= 0.021$) interactions being shown in Table 2. This index behaved in a similar manner to the N use efficiency index for N levels. The lower value recorded for NUPE was in intra- and inter-specific competition ($D_2W_1= 0.68 \text{ kg kg}^{-1}$) which was significantly ($r= 0.5$, $p= 0.0009$) lower than other competition types. No significant differences ($r= 0.5$, $p \geq 0.05$) observed between intraspecific ($D_2W_0= 0.77 \text{ kg kg}^{-1}$) and interspecific ($D_1W_1= 0.75 \text{ kg kg}^{-1}$) treatments. Nitrogen uptake in the no competition treatment (D_1W_0) was more efficient in all N rates and the highest efficiency was

recorded by this treatment ($D_1W_0=0.95 \text{ kg kg}^{-1}$) in N_{50} . Differences among N rates with respect to grain yield, which is directly related to crop N uptake, account for the variation in the NUPE index. Dhugga and Waines (1989), on the hexaploid spring wheat and tetraploid spring wheat (*Triticum turgidum* L.), have shown that N uptake efficiency becomes more important than N utilization efficiency in determining N use efficiency as soil N supply increased. The major managerial causes for low NUPE are: (i) poor synchrony between fertilizer N and crop demand, e.g. associated with large application of N fertilizer early in the season, (ii) uniform field applications to spatially variable fields, (iii) use of yield based N recommendations assuming

constant efficiency of fertilizer recovery and (iv) failure to take account of year-to-year weather variations, e.g. the amount of soil N mineralized in warm and wet years (Raun and Johnson 1999).

N utilization efficiency (NUTE): This reflects the efficiency with which crops utilize N in the plant for the synthesis of grain yield. Increased N fertilizer rates prompted a fall in the NUTE index. Nitrogen utilization efficiency ($r= 0.56$, $p= 0.0024$) was lower for N_{150} than for other N rates (Table 2). The highest NUTE was observed in intraspecific competition ($D_2W_0= 41.5 \text{ kg kg}^{-1}$) ($p= 0.002$) and followed by intra- and inter-specific competition ($D_2W_1= 39.33 \text{ kg kg}^{-1}$) ($p= 0.002$). In both lowest (N_0) and highest (N_{150}) N rates, intraspecific competition (D_2W_0) ($r= 0.69$, $p= 0.0014$) had the best NUTE due to the lower N supply (Data not shown). In addition, differences in grain yield between N rates were eliminated with increased levels of applied N and resulted in greater NUTE under lower N rates (39.29 kg kg^{-1}) as compared by the higher N rate (29.33 kg kg^{-1}) (Table 2). Similar findings also reported by Delogu *et al.* (1998) and Lopez-Bellido and Lopez-Bellido (2001); they suggested that differences in N utilization efficiency among cultural practices are expressed when grain yields are either lower or higher at equivalent levels of aboveground N. Comparisons of yield at equal levels of aboveground N removes most yield effects of cultural practices due to differences in N supply, availability, or uptake. Under high N supply, several studies worldwide concluded that wheat breeding did not result in consistent improvements in NUPE but in improved NUTE associated with higher harvest index, e.g. in Mexico (Fageria 2009). Selection for higher HI may reduce the N storage capacity of the shoot

and reduction in protein N has been associated with high NUTE so the disadvantage of high NUTE may be a reduced nutrient value of the plant. In addition to the better expression of NUTE under either high soil N or low soil N, there are differences between aerial species in their NUE to NUTE relationship (Dhugga and Waines 1989).

Nitrogen reliance index (NRI): A greater NRI indicates increased dependency on external N supplies to alleviate crop N deficiencies. Furthermore, depletion of internal N contribution over time would indicate soil resource degradation and a greater necessity to rely on external N inputs to achieve crop performance goals. The NRI index was significantly ($r= 0.97$, $p < 0.0001$) affected by crop competition and N rates (Table 2). This index was significantly ($r= 0.97$, $p= 0.0006$) higher for intraspecific competition ($D_2W_0= 0.31 \text{ kg kg}^{-1}$) than other interspecific competition ($D_1W_1= 0.27 \text{ kg kg}^{-1}$). No significant differences was observed between intraspecific ($D_2W_0= 0.31 \text{ kg kg}^{-1}$) and both intra- and inter-specific competition ($D_2W_1= 0.31 \text{ kg kg}^{-1}$). NRI increased as N fertilizer rates increased, with significant differences among all rates. NRI value at maximum N fertilizer rate of 150 Kg N ha^{-1} was 0.50 to 0.25 kg kg^{-1} lower than those obtained at 50 Kg N ha^{-1} . The variation of NRI was 50%. The competition \times N fertilizer rate interaction was significant ($r= 0.98$, $p < 0.0001$) and showed that NRI increased with higher N rates and averaged 22% at the lowest N level for the no competition treatment. Similar results by Huggins and Pan (1993) showed that the N reliance index increased with higher N rates and

averaged 69% at the greatest N level under conventional tillage. Therefore, Huggins and pan (1993) noted that the N reliance index has likely increased over time in many agroecosystems as levels of soil organic matter have declined.

Nitrogen balance index (NBI): This is defined as the ratio of N removal via grain harvest to N fertilizer inputs and accounts for major N inputs and outputs directly related to N management.

NBI varied significantly according to N rates ($r=0.86$, $p<0.0001$) and competition type was shown in Table 2. This index declined as N fertilizer rates increased and no significant difference was observed at two highest N rates. NBI for the no competition treatment was significantly highest than the other competition types. On average, the lowest value recorded for NBI was in the highest

Table 2. Grain yield, N use efficiency (NUE), N uptake efficiency (NUPE), N utilization efficiency (NUTE), N reliance index (NRI) and N balance index (NBI) for competition types at different N rates

Treatments	Grain yield	NUE	UPE	UTE	NRI	NBI
	Ton ha ⁻¹	Kg Kg ⁻¹				
Nitrogen (N)						
N ₀	3.25 b+	32.54 a	0.84 a	39.85 a	-	-
N ₅₀	5.70 a	30.43 a	0.78 ab	39.29 a	0.25 c	2.35 a
N ₁₀₀	6.00 a	22.84 b	0.73 b	32.02 b	0.40 b	1.30 b
N ₁₅₀	6.15 a	20.22 b	0.70 b	29.33 b	0.50 a	1.19 b
Plant × Weed density						
D ₁ W ₀	5.74 a++	24.1 b	0.87 a	27.92 b	0.26 b	1.53 a
D ₁ W ₁	4.96 b	23.83 b	0.75 bc	31.74 b	0.27 b	1.16 b
D ₂ W ₀	5.53 ab	31.52 a	0.77 b	41.50 a	0.31 a	1.20 b
D ₂ W ₁	4.88 b	26.58 b	0.68 c	39.33 a	0.31 a	0.95 b
Third order interactions						
N ₀ D ₁ W ₀	3.9 de	25.80 c-f	0.91 a	28.52 fg	-	-
N ₀ D ₁ W ₁	2.81 e	32.67 a-c	0.80 a-e	41.3 a-e	-	-
N ₀ D ₂ W ₀	3.05 e	39.26 a	0.88 ab	45.33 a-c	-	-
N ₀ D ₂ W ₁	3.25 e	32.42 a-c	0.78 b-e	42.00 a-d	-	-
N ₅₀ D ₁ W ₀	6.53 bc	28.95 b-e	0.95 a	30.18 e-g	0.22 e	2.90 a
N ₅₀ D ₁ W ₁	5.13 cd	24.09 c-f	0.83 a-d	28.17 fg	0.23 e	2.05 c
N ₅₀ D ₂ W ₀	6.47 bc	36.22 ab	0.69 c-f	52.60 a	0.28 e	2.53 b
N ₅₀ D ₂ W ₁	5.89 bc	32.48 a-c	0.68 c-f	48.02 ab	0.28 e	2.03 cd
N ₁₀₀ D ₁ W ₀	7.15 a	19.36 d-f	0.8 a-e	25.75 g	0.36 d	1.53 c-e
N ₁₀₀ D ₁ W ₁	5.10 cd	19.02 f	0.7 b-f	27.82 fg	0.38 cd	1.34 ef
N ₁₀₀ D ₂ W ₀	5.47 cd	30.10 bc	0.86 a-c	36.11 c-g	0.44 b	1.42 d-f
N ₁₀₀ D ₂ W ₁	5.33 cd	22.87 d-f	0.60 f	38.37 b-f	0.43 bc	0.89 f
N ₁₅₀ D ₁ W ₀	7.32 a	22.27 f	0.85 a-c	27.24 fg	0.46 b	1.79 c-e
N ₁₅₀ D ₁ W ₁	6.35 bc	19.52 f	0.67 d-f	29.23 fg	0.47 b	1.25 ef
N ₁₅₀ D ₂ W ₀	5.69 bc	20.52 ef	0.65 d-f	31.94 d-g	0.54 a	0.85 f
N ₁₅₀ D ₂ W ₁	5.24 cd	18.56 f	0.64 ef	28.92 fg	0.53 a	0.88 f

+ In each section, means followed by the same letter within columns are not significantly different ($p\leq 0.05$) according to LSD test.

++ D₁: ~450 plants m⁻² (optimum wheat density), D₂: ~750 plants m⁻² (high wheat density), W₀: control (weed absence), W₁: weed presence (30 weed.m⁻²).

N fertilizer application (N_{150}) for all competition types. NBI was the lowest for intra- and inter-specific competition at the all N rates presumably because of the lowest grain N in this treatment (Data not shown). Similarly, Huggins and Pan (2003) reported that this index was decreased by increase in N levels and noted that values of N balance indices close to one occur for the first level of applied N and then decrease below one as levels of applied N increase. In this case, N requirement for obtaining wheat yield and grain protein goals, result in N balance indices that are less than one. Therefore, they suggested that N balance indices greater than one indicate the use of internal N sources and possible soil N resource degradation, whereas indices less than one indicate the potential for N losses from the agroecosystem. In fact, the combination of N balances and NUE offers a sound basis to adjust N fertilizer application to optimize N management for productivity and ecological sustainability of cropping systems (Arregui and Quemada 2008).

N harvest index (NHI): This is the proportion of aboveground N that is partitioned to the grain. NHI was significantly ($r=0.24$, $p=0.04$) influenced by just N fertilizer rates (Table 3) and competition \times N rate interaction were not significant ($r= 0.48$, $p= 0.083$). Application of N fertilizer at 100 and 150 Kg N ha⁻¹ prompted a lower NHI than the other rates (Table 3). The highest value for NHI corresponded to the no competition treatment ($D_1W_0= 73.68$ %). While no significant ($r= 0.26$, $p= 0.314$) differences were found among competition types. In addition, NHI in the intraspecific competition (D_2W_0) plots was higher than the interspecific competition

(D_1W_1) plots for all N application rates. Delogu *et al.* (1998) and Lopez-Bellido and Lopez-Bellido (2001) reported that application of fertilizer at 150 kg N ha⁻¹ prompted a lower NHI than 0 kg N ha⁻¹. Genetic variations for NHI exist with small grain species and high NHI values are associated with low rates of N fertilizer application (Muurinen *et al.* 2007). Moreover, significant differences in NHI occurred between cultural practices (i.e. tillage) at equal N rates. Therefore, increasing the annual stability of NHI in wheat may be an important management objective particularly if grain quality is a goal (Huggins and Pan 2003). In addition, under low N supply, genetic gains in yield were positively associated with AGN and NHI and negatively associated with GNC, whereas under high N supply, genetic gains in yield were positively associated with AGN (Ortiz-Monasterio *et al.* 1997).

N remobilization efficiency (NRE): This is estimated as the fraction of stored N at flowering that is not recovered in the vegetative parts at maturity. This index was significantly ($r= 0.30$, $p= 0.023$) affected by N fertilizer rate (Table 3). The competition \times N fertilizer rate interaction showed that only rates of 50 Kg N ha⁻¹ prompted more significant ($r= 0.48$, $p= 0.036$) differences between different competition treatment types with regard to the NRE index. The competition \times N fertilizer rate interaction was significant between intra- and inter-specific competition types only at the highest rate of N application (N_{150}). Nitrogen remobilization efficiency diminished as N fertilizer rates increased, with significant differences among lowest (N_0) and highest (N_{150}) rates (Table 3). In other words, NRE value at a minimum N fertilizer rate of 0 Kg

N ha⁻¹ was 77.21 to 68.61% which was obtained at 150 Kg N ha⁻¹. This variation of NRE was 11.14% between N rates. The competition × N fertilizer rate interaction showed that the no competition treatment at the highest N rate (N₁₅₀) had highest NRE. Muurinen *et al.* (2007) reported that the results from different N treatments in their study had supported the assumption that lower N supply (0 and 50 kg N ha⁻¹) would result

in a higher NRE than the 100 and 150 kg N ha⁻¹ treatment. This might indicate that at higher rates of N availability, N does not need to be translocated from vegetative plant parts to fill grains as efficiently as in the case where N could not be taken up from soil. However, wheat NRE correlated strongly and negatively with tiller traits. They suggested that N translocation would

Table 3. N harvest index (NHI), N remobilization efficiency (NRE), N harvest efficiency (NHE), N fertilizer utilization efficiency (NFUE) for competition types at different N rates

Treatments	NHI	NRE	NHE	NFUE
	%		Kg Kg ⁻¹	
Nitrogen (N)				
N ₀	77.04 a+	77.21 a	0.65 a	-
N ₅₀	71.31 ab	71.44 ab	0.58 ab	120.09 a
N ₁₀₀	69.46 b	70.70 ab	0.54 ab	56.99 b
N ₁₅₀	68.87 b	68.61 b	0.51 b	41.00 c
Plant × Weed density				
D ₁ W ₀	73.68 a++	73.55 a	0.71 a	58.50 a
D ₁ W ₁	68.47 a	67.10 a	0.51 b	48.99 b
D ₂ W ₀	73.37 a	75.35 a	0.59 b	59.07 a
D ₂ W ₁	71.16 a	71.96 a	0.48 b	51.51 ab
Third order interactions				
N ₀ D ₁ W ₀	77.00 a-c	73.63 a	0.81 ab	-
N ₀ D ₁ W ₁	73.96 a-d	73.47 a	0.48 de	-
N ₀ D ₂ W ₀	76.56 a-c	80.82 a	0.77 a-c	-
N ₀ D ₂ W ₁	80.64 a	80.93 a	0.56 c-e	-
N ₅₀ D ₁ W ₀	74.31 a-d	70.81 ab	0.82 a	130.54 a
N ₅₀ D ₁ W ₁	59.92 e	69.50 ab	0.59 b-e	102.63 b
N ₅₀ D ₂ W ₀	79.41 ab	72.81 a	0.45 de	129.32 a
N ₅₀ D ₂ W ₁	71.58 a-e	72.64 a	0.47 de	117.87 ab
N ₁₀₀ D ₁ W ₀	72.85 a-d	71.69 a	0.66 a-d	54.67 cd
N ₁₀₀ D ₁ W ₁	70.53 a-e	71.93 a	0.49 de	50.99 d-f
N ₁₀₀ D ₂ W ₀	65.87 c-e	70.75 ab	0.50 de	69.03 c
N ₁₀₀ D ₂ W ₁	68.60 b-e	68.41 ab	0.52 de	53.26 c-e
N ₁₅₀ D ₁ W ₀	69.36 a-e	78.08 a	0.55 c-e	48.81 d-f
N ₁₅₀ D ₁ W ₁	69.48 a-e	53.51 b	0.49 de	42.35 d-f
N ₁₅₀ D ₂ W ₀	72.83 a-d	77.00 a	0.63 a-d	37.94 ef
N ₁₅₀ D ₂ W ₁	63.83 de	65.85 ab	0.38 e	34.90 f

+ In each section, means followed by the same letter within columns are not significantly different ($p \leq 0.05$) according to LSD test.

++ D₁: ~ 450 plants m⁻² (optimum wheat density), D₂: ~750 plants m⁻² (high wheat density), W₀: control (weed absence), W₁: weed presence (30 weed.m⁻²).

have been from tillers during a very late grain filling phase, whereas there was no strong N translocation from vegetative parts of the main shoots, which could still have been competing with filling grains for N. They also, reported that higher levels of nitrogen fertilization before flowering lead to a decrease in N remobilization efficiency as the resulting higher level of nitrogen availability just after flowering renders N remobilization unnecessary. However, high NUE may alternatively be associated with large capacities for uptake and assimilation of N and with high post-anthesis N remobilization efficiency. For maize it has been reported that early remobilization of N from the stem before the leaf lamina is consistent with the use of stem N as a buffer for the flow of N from the leaf lamina to the grain. In this case, a high stem N absorption capacity coupled with high stem N remobilization efficiency (proportion of N in the plant organ at anthesis which is not recovered in the organ at harvest; NRE) would potentially favor high NUTE through delayed senescence of the leaf lamina. A high stem N absorption coupled with a low stem NRE, however, would be associated with an inefficient use of absorbed and a low NUTE (Fageria 2009).

Nitrogen harvest efficiency (NHE): NHE or grain N accumulation efficiency evaluates crop physiological processes that contribute to the overall efficiency at which N accumulates in the grain. NHE was significantly ($r= 0.41$, $p= 0.03$) influenced by both N rates and competition ($r= 0.41$, $p= 0.002$) treatments (Table 3). NHE declined as N fertilizer rates increased. NHE value at a minimum N fertilizer rate of 0 Kg N ha⁻¹ was

0.65 to 0.51 kg kg⁻¹ which was obtained at 150 Kg N ha⁻¹. This variation of NHE was 21.54%. The competition × N rates interaction showed that only the rate of 50 Kg N ha⁻¹ (control) prompted no significant ($p \geq 0.05$) differences among competition types. Huggins and Pan (2003) reported that, when this efficiency is less than one, it is a warning signal of the potential for N surpluses to pollute surface and ground waters, if they are not recycled and efficiently used by subsequent crops. Also, they reported that NHE in wheat decreased with increasing levels of applied N. Therefore, they suggested that management strategies to increase NHE include improving predictive capabilities for quantifying available N supplies from N mineralization-immobilization turnover or improving available N uptake efficiencies (Huggins and Pan 2003).

Nitrogen fertilizer utilization efficiency (NFUE): Evaluation of N fertilizer utilization efficiency based on ratio of yield and applied N enables simple comparisons among treatments on efficiencies of applied N use. This index was significantly affected by competition ($r= 0.95$, $p < 0.0001$) and N fertilizer rate ($r= 0.95$, $p= 0.047$) (Table 3). The interspecific competition ($D_1W_1= 48.99$ kg kg⁻¹) and the intraspecific competition ($D_2W_0= 59.07$ kg kg⁻¹) had the lowest and the highest NFUE, respectively. The competition × N fertilizer rate interaction showed that only rates of 150 Kg N ha⁻¹ prompted no significant differences between competition treatments with regard to the NFUE index. The competition × N fertilizer rate interaction was significant ($r= 0.97$, $p < 0.0001$) between competition treatments at the lowest rates of N application (N₅₀ and N₁₀₀). Nitrogen fertilizer

utilization efficiency diminished as N fertilizer rates increased, with significant ($p < 0.0001$) differences among all N rates (Table 3). Also, NRE value at a lower N fertilizer rate of 50 Kg N ha⁻¹ was 120.09 to 41 kg kg⁻¹ which that obtained at 150 Kg N ha⁻¹. This variation of NFUE was 65.86% between N rates. The competition \times N fertilizer rate interaction showed that the both intra- and inter-specific competition treatment (D₂W₁) at the highest N rate (N₁₅₀) had lowest NFUE (34.9 kg kg⁻¹). The decrease in N fertilizer utilization efficiency that occurs as levels of applied N increase is due, in part, to the yield response to indigenous levels of available soil N when no N fertilizer is used. Consequently, as contributions of indigenous soil N decrease, the level of applied N required to obtain yield goals increases and overall utilization efficiency decreases. Increased dependence on N fertilizer and consequent decreases in utilization efficiency can increase the economic sensitivity of the cropping system to changes in grain and fertilizer N prices (Huggins and Pan 2003). NFUE varied with year and N rate. When averaged over all years, NFUE decreased as the amount of N

fertilizer applied increased. At the 84 kg N ha⁻¹ fertilizer rate, which had the highest average grain yield, NFUE averaged 56%. At the 56 kg N ha⁻¹ fertilizer rate, grain yields were 97% of the average maximum yield and had a NFUE of 69% (Halvorson *et al.* 2004).

Relationships between grain yield and nitrogen indices

Grain yield was positively correlated with NUE, NUPE, NHE, NRI, NBI and NFUE. Therefore, NUE could be used to predict high grain yield. NUE correlated positively with grain yield and negatively with protein concentration (Săulescu *et al.* 2005). Also, NUE showed higher correlation with NUTE than NUPE. In addition, grain yield had the highest correlation with NRI, indicating that high grain yield was associated with more N fertilizer application. Positive correlation between NUE and grain yield has been previously reported, indicating that grain yield could determine by NUE. No correlation of grain yield with NUTE, NRE and NHI occurred (Table 4). On the other hand, relationships between N indices for each competition type and gain yield were evaluated by linear regression (Figure 1).

Table 4. Linear correlation coefficients of grain yield with nitrogen indices

	NUE	NUPE	NUTE	NRI	NBI	NHI	NRE	NHE	NFUE	Grain yield
NUE	1									
NUPE	0.28 *	1								
NUTE	0.72 **	-0.43 *	1							
NRI	-0.08NS	0.14NS	-0.18NS	1						
NBI	0.43 **	0.65 **	-0.12NS	0.34 *	1					
NHI	0.30 *	0.21NS	0.12 NS	-0.36 *	0.01 ^{ns}	1				
NRE	0.21NS	-0.06NS	0.23NS	-0.16NS	-0.11NS	0.8NS	1			
NHE	0.36 *	0.94 **	-0.32 *	0.01NS	0.58 **	0.52 **	0.23 NS	1		
NFUE	0.55 **	0.42 **	0.15NS	0.31 *	0.92 **	-0.11NS	-0.18NS	0.34 *	1	
Grain yield	0.52 **	0.39 **	0.20NS	0.66 **	0.61 **	-0.08NS	-0.16NS	0.32 *	0.64 **	1

NUE= N use efficiency, NUPE= N uptake efficiency, NUTE= N utilization efficiency, NRI= N reliance index, NBI= N balance index, NHI= N harvest index, NRE= N remobilization efficiency, NHE= N harvest efficiency, NFUE = N fertilizer utilization efficiency.
* ($P \leq 0.05$), ** ($P \leq 0.01$), NS: non-significant ($P > 0.05$).

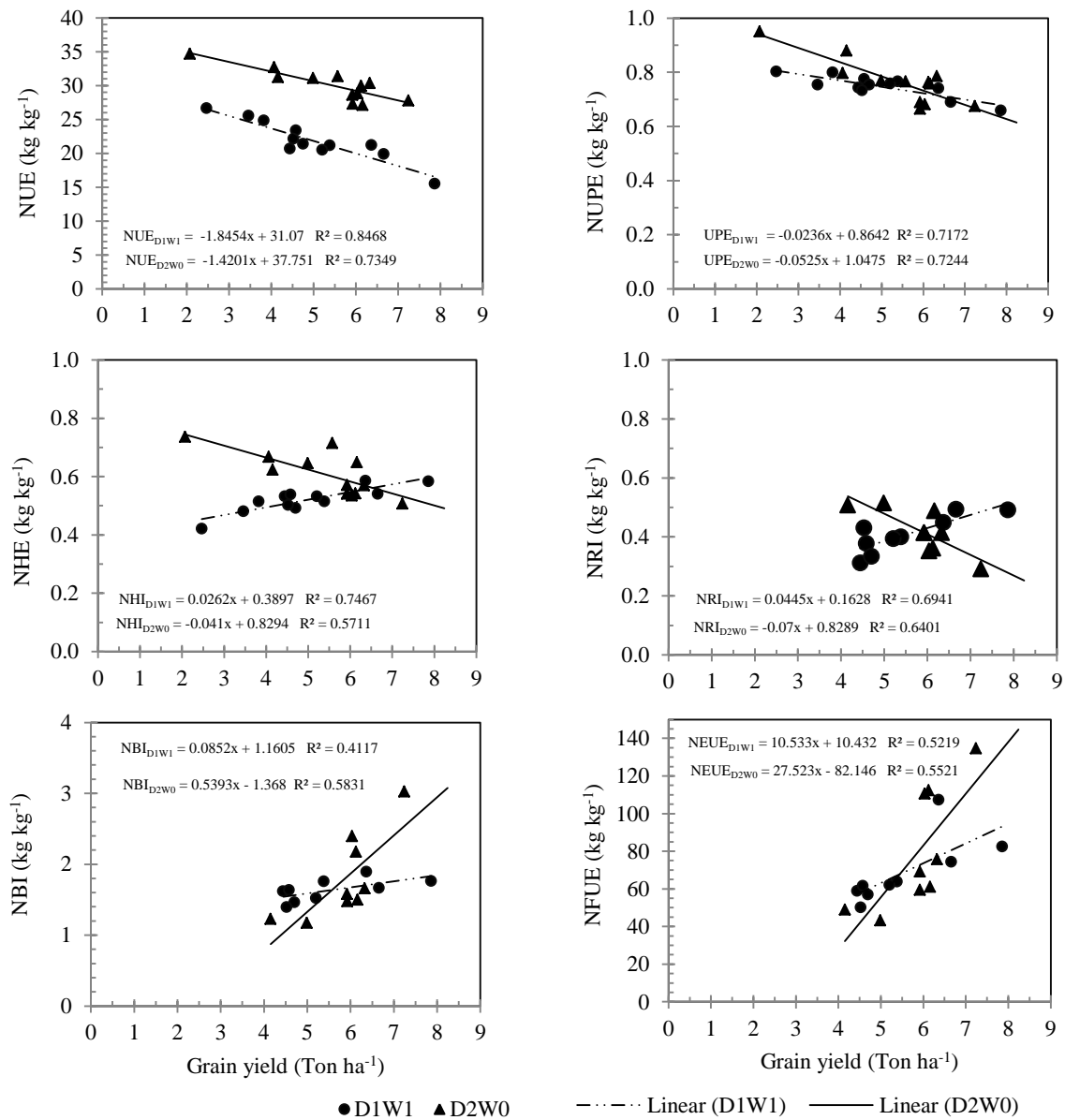


Figure1. Changes in nitrogen use efficiency with grain yield for competition types

There were significant linear relationships between six indices and grain yield of wheat. It is noteworthy that the trend of the regressions for competition types differed markedly among all N indices. The NUE of interspecific competition was much lower than the NUE of intraspecific competition. In contrast, there was a positive trend between grain yield and NBI, and also NFUE. Ortiz-Monasterio *et al.* (1997) demonstrated that

under moderate fertilization rate, the NUE values of the modern wheat cultivars (higher competitor with weeds) were higher than the old cultivars (lower competitor with weeds).

Conclusions

Wheat grain yield showed no significant increase when N application rates exceeded 50 kg ha⁻¹ (in spite of improvement with increasing N rates).

NUE decreased with increasing N level, especially under higher N rates, therefore, reduced benefits of fertilizer application. In fact, all nitrogen use efficiencies diminished as N fertilizer rates increased (except N reliance index), with significant difference among competition types. Reduction in NUE with increasing N supply could result from reduction in any (or all) of the components, including NUPE and NUTE. In addition, NUPE, NHI, NRE, NHE and NFUE were lowest in the intra- and inter-specific competition treatments than others. The differences between each intra and inter-specific competition treatments were not significant for NUPE, NBI, NHI, NRE and NHE. We suggested that greatly increased suppression of weeds through increased crop density for reducing the use of herbicide can play an important role in a change in both crop yield and crop N use efficiencies. This strategy increased NUE, NUTE

and NRI indices. In contrast, higher intraspecific competition reduced NUPE, NBI and NHE indices but, had no significant effect on NHI, NRI and NFUE. In addition, the inclusion of weed in competition with wheat had negative impacts on many N efficiency indices; in other word, the NUPE, NBI, NHI and NFUE were significantly reduced. Therefore, a combination of an appropriate N rate with a suitable choice of plant density increased NUE which may suppress weed competition adequately. Finally, there were no significant differences among applied N rates (from 50 to 150 kg N ha⁻¹); therefore, it seems that paying attention only to the traditional production criterion (i.e., grain yield) will not be a comprehensive method and in terms of sustainable agriculture point of view we should seek other indicators such as efficiency indices (especially for N).

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