The inheritance of nitrogen use efficiency traits of wheat (*Triticum aestivum L.*) under low soil-N conditions

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Abstract In order to study the inheritance of nitrogen use efficiency (NUE) traits as indicators of low-N tolerance in bread wheat, plants from a six-parental diallel cross, excluding reciprocals, were grown in the field for two seasons using a randomized complete block design with three replications in two separate experiments; the first under no N fertilization, *i.e.* 0 kg N/fed (low-N) and the second under fertilization rate of 75 kg N/fed (high-N). Results across seasons showed that variances due to both general (GCA) and specific (SCA) combining ability were significant for all studied nitrogen use efficiency traits, except nitrogen harvest index (NHI) under low-N. The best general combiners for improvement of NUE and nitrogen uptake efficiency (NUPE) under low and high N were L26, L27 and L25 inbreds. Under low–N conditions, the best specific combinations for improvement of low-N tolerance were the F_1's L25 x Gz168 for NUE and NUPE, L2 x Gem9 and L27 x Gem9 for NUPE and L25x L 26, L25 x L27 and L27 x Gem9 for nitrogen utilization efficiency(NUTE) trait. Hayman's analysis indicated that NUE, NUPE, NHI and NUTE are controlled by additive and non-additive types of gene action. The magnitude of dominance was much greater than additive (D) component for all studied traits in F_1's under both high-N and low-N, except for NUE trait under low-N, where the opposite was true. High narrow-sense heritability estimate was observed for NUE (66.90%) and NUPE (46.36%) under low-N. Expected genetic advance from selection under low-N was much higher than under high-N for all NUE traits, especially for NUPE. Under low-N, the inbreds L25, L26 and L27 had more than 75% dominant genes of NUPE.

Keywords *Triticum aestivum*, Gene action, NUE, Low-N, Diallel analysis

Introduction

Wheat (*Triticum aestivum* L.) is one of the oldest and most important cereal crops in Egypt. Although wheat productivity in Egypt has increased during the past years, wheat production supplies only 45% of its annual domestic demand. Egypt is one of the largest countries that import wheat. Wheat imports in 2011 were about 9.8 million tons, with a cost of about 3.2 billion US$ [1]. Therefore, Egypt needs to make a great effort to increase wheat production. Extending wheat growing outside the Nile Valley is the first effort toward overcoming wheat problems. However, most of the area outside the Nile Valley suffers from some abiotic stresses, the most important are nutrient deficiency and low water holding capacity; therefore increasing tolerance of wheat genotypes to such stresses, is one of the cheapest methods to spread growing wheat in these areas.

Nitrogen (N) is one of the major inputs in wheat production systems. During the green revolution, plant breeding programs have released many Mexican type semi dwarf varieties with greater responses to high nitrogen input. Cultivation of these cultivars drastically increased wheat average yield in the world [2]. Thus the consumption of nitrogen fertilizers was increased tremendously in the world. Today, elevated nitrogen level in

water, as result of leaching, is an important component of agricultural pollution causing major problems in marine ecosystems and eutrophication of freshwater [3]. Moreover, N fertilization increases emissions of the greenhouse gas nitrous oxide (N_2O) from agricultural soils [4]. Volatile ammonia emissions from fertilizer contribute to deposition of N in unmanaged ecosystems [5].

Low-N availability in soils in developing countries in an important yield- limiting factor frequently found in farmers' fields, since the smallholder farmers cannot afford additional inputs. Based on these essential economic and ecological grounds, an increased interest is being shown worldwide in cereal cultivars that are more efficient in utilizing soil resources and better fitted to water and nutrient limitations [6-13].

Among cereals, hexaploid wheat (*Triticum aestivum* L.) is commonly identified as a species with higher requirements for nutrients, especially nitrogen. Thus, breeding wheat cultivars with improved adaptation to less favorable, but more optimized N fertilization regimes has gained importance. In Egypt, such breeding strategies are also justified by problems of nitrogen that is a major constraint limiting grain production.

The efficiency of nitrogen use (NUE; defined as the grain yield per unit of the soil N) and plant adaptation to less favorable nutrition regimes is complex with various endo- and exogenous mechanisms involved [14]. Different morpho-physiological characteristics, associated with both the uptake capacity (NUPE; defined as a proportion of total N uptake to N availability in the soil) and efficiency of nitrogen utilization in grain mass formation (NUTE; defined as the grain mass formed per unit of N absorbed), appear to be critical components of NUE [15-16].

Although numerous reports on genotypic variation in components of N efficiency already suggest potential applications of this genetic knowledge for wheat improvements [2, 6, 17-23], relatively fewer attempts have been made to breed wheat for these traits [24-27].

Progress in breeding wheat better adapted to less favorable fertilization regimes is still restricted for several reasons. Wheat breeders are frequently skeptical not only because of the morpho-physiological complexity of the matter, but mainly due to limited data on both the variation among available wheat collections and the genetics of key characters involved. Hence, several important queries remain to be resolved, especially in regard to the most effective selection schemes, desirable plant ideotypes for low input ecosystems, appropriate selection criteria and features of the selection environment or cropping systems necessary for such breeding programs [27-29].

Furthermore, modern Egyptian wheat cultivars are phenotypically different but, in essence, represent a limited gene pool. The majority of them were developed under favorable or even luxurious fertilization regimes used at most breeding stations without or with scarce selection pressure for components of nutrient use efficiency. On the contrary, beneficial plant characteristics for low-input ecosystems may be different from those present in modern, high-yielding wheat cultivars [6, 30]. This raises concerns for breeders as to whether the range and spectrum of genetic variation in nutrient efficiency among modern wheat cultivars is sufficiently wide under sub-optimal habitats to guarantee progress in breeding more efficient wheat cultivars better adapted to less favorable fertilization practices.

The manner in which target traits are inherited has, of course, major consequences for the whole breeding strategy. However, our understanding of the inheritance of the morpho-physiological components of N efficiency in wheat, as in other cereals, is still extremely limited. There is some information regarding various characteristics decisive for the uptake efficiency in juvenile wheat plants [19-20, 31-32], but the limited data may lead to uncertain conclusions on N efficiency over the whole growing season. However, the genetic control of whole-season N efficiency has rarely been examined in wheat. Recent extensive molecular studies [33-35] identified numerous genome regions (QTLs) responsible for grain yield structure and nitrogen yield under N limitations, grain protein content and N metabolism in the uppermost foliage as well as for the activity of glutamine synthetase and glutamate dehydrogenase, the key enzymes involved in N assimilation. In earlier investigations [36], both additive and non-additive genetic effects were crucial for agronomic NUE components in F1 hybrids between modern cultivars. In contrast, additive gene action was only important for NUE components among F2 progenies of wheat cultivars [37].

To the best of our knowledge, however, no information exists in wheat for the major physiological measures of N efficiency, *i.e.* the uptake and utilization efficiencies (NUPE and NUTE) when whole-season indices and

conventional genetic/ breeding approaches are considered, and this may have an impact on current breeding methods, aspirations and goals. Thus, the main objectives of this study were: (i) to investigate the relative importance of GCA and SCA in a set of wheat cultivars and promising lines and their F_1 diallel crosses for four NUE traits, (ii) to assess the modes by which NUPE, NUTE, NUTE and nitrogen harvest index (NHI) traits are inherited, and (iii) to evaluate effects of varied nutrition on gene action. For these purposes, hybrids between six wheat lines and cultivars of divergent tolerance to low-nitrogen were evaluated under diverse N fertilization regimes.

Materials and Methods
This study was carried out at Giza Research Station of the Agricultural Research Center(ARC), Giza Egypt (30° 02'N latitude and 31° 13'E longitude with an altitude of 22.50 meters above sea level), in 2005/2006 season and at Noubarya Research Station of the ARC, Noubarya, Egypt (30° 66'N latitude and 30° 06' E longitude with an altitude of 15.00 meters above sea level), in 2006/2007, 2007/2008 and 2008/2009 seasons.

Breeding materials
Six bread wheat genotypes (*Triticum aestivum* L.) were chosen for their divergence in tolerance to low nitrogen, based on previous field screening carried out by Wheat Res. Dept., Field Crops Res. Inst., ARC, Egypt (Table 1).

Table 1: Designation, pedigree and tolerance to low N of the six promosing lines and Egyptian cultivars of wheat used for making diallel crosses of this study.

Designation	Pedigree	Tolerance to low nitrogen
Line 25 (L25)	MYNA/VUL//TURACO/3/TURACO/4/Gem7.	Tolerant
Line 26 (L26)	MUNIA/CHTO//AMSEL.	Tolerant
Line27 (L27)	Compact-2/Sakha//Sakha61.	Tolerant
Gemeiza(Gem7)	CMH74A.630/SX//Seri82/3/Agent.	Sensitive
Gemeiza(Gem9)	Ald "s"/HUC "s;;//CMH74A.630/SX.	Sensitive
Giza168(Gz168)	MRL/BUC//Seri.	Sensitive

Source: Wheat Res. Dept., Field Crops Res. Inst., ARC. Egypt.

Making the F_1 diallel crosses
In season 2005/2006, a half diallel of crosses involving the six parents (without reciprocals) was done at Giza Agric. Res. Stat., Agric. Res. Center, to obtain the F_1 seeds of 15 crosses. In season 2007/2008, the half diallel of crosses was again done to increase quantity of F_1 seeds.

Field evaluation of 6 parents and 15 F_1's
In the seasons 2007/2008, 2008/2009, parents (6) and F_1's (15) were sown on 17th of November each season in the field of Noubarya Res. Stat., under two rates of nitrogen fertilizer; each rate in a separate experiment; the low rate was no-fertilization , *i.e.* 0 kg Nitrogen/feddan (LN) and the high rate was fertilization at a rate of 75 kg Nitrogen/feddan (HN). This rate of nitrogen fertilizer (equals 168 kg Urea/fed) was added in two equal doses; the first dose was added just before the sowing irrigation and the second dose just before the first irrigation (21 days after sowing). In each experiment, a randomized complete block design was used with three replications. Each parent or F_1 was sown in two rows; each row was three meter long; spaces between rows were 30 cm and 10 cm between plants; the plot size was 1.8 m^2. All other agricultural practices were done according to the recommendation of Ministry of Agriculture for growing wheat in Noubarya region.

Available soil nitrogen in 30 cm depth was analyzed immediately prior to sowing and N application at the laboratories of Water and Environment Unit, ARC, Egypt in the two seasons. Soil nitrogen was found to be 55 and 57 kg N/ fed in the seasons 2007/2008, 2008/2009, respectively. Available soil nitrogen after adding nitrogen fertilizer was therefore 55 and 130 kg N/fed in the first season and 57 and 132 kg N/fed in the second season for the two treatments, *i.e.* LN and HN, respectively. The available nitrogen to each plant (including soil

and added N) was calculated for each environment to be 0.79 and 1.85 g/plant in 2007/2008 season and 1.89 kg/fed in 2008/2009 season, with an average, across the two seasons, of 0.80 and 1.87 g/plant for the two environments LN and HN, respectively. The soil analysis of the experimental soil at Noubarya Research Station, as an average of the two growing seasons, indicated that the soil is sandy loam (67.86% sand, 7.00% silt and 25.14% clay), the pH is 8.93, the EC is 0.55 dSm^{-1}, the soluble cations in meq l^{-1} are Ca^{2+} (5.30), K^+ (0.70), Na^+ (0.31), Mg^{2+} (2.60) and the soluble anions in meq l^{-1} are CO_3^{2-} (0.00), HCO_3^- (2.10), Cl^- (5.30) and SO_3^{2-} (1.51).

Data collection

Grain yield/ plant (GYPP) was measured as weight of the grains of each individual plant using an average of 10 plants each entry. At physiological maturity stage, five random guarded plants were removed from each plot by cutting at the soil surface. The plants were bulked as one sample per plot. They were separated into straws (including leaves, stems and spike residues) and grains. Samples were oven dried at 70°C to a constant weight and each part was weighed separately. Samples were ground in powder and nitrogen of straws (N straw) and grains (N_g) was determined using Kjeldahl procedure according to A.O.A.C., 1990 [38]. Total plant nitrogen (N_t) was calculated as follows: $N_t = N_g + N_{straw}$. The following traits were recorded: **1. Nitrogen use efficiency (NUE) g/g**= (GYPP/N_s). **2. Nitrogen uptake efficiency (NUPE)%** =100 (N_t/N_s). **3. Nitrogen utilization efficiency (NUTE)** (g/g)= (GYPP/N_t). **4. Nitrogen harvest index (NHI%)**= 100(N_g/N_t). Where, N_t is total nitrogen in the whole plant (grains and straw), N_s is available nitrogen in the soil for each plant, and N_g is grain nitrogen content. Nitrogen efficiency parameters No. 1, 2, and 3 were calculated according to Moll *et al.* 1982 [15].

Statistical analysis

Each environment (HN and LN) was analyzed separately across seasons as RCBD using GENSTAT 10th addition windows software. Least significant differences (LSD) values were calculated to test the significance of differences between means according to Steel *et al.*, 1997 [39].

Genetic analyses of F_1 diallel crosses
a. Griffing approach

Diallel crosses in F_1 generation were analyzed to obtain general (GCA) and specific (SCA) combining ability variances and effects for studied traits, according to Griffing [40] model I, *i.e.* the fixed model, method II as shown in Singh and Chaudhary [41].

b. Hayman's numerical approach

The genetic parameters and ratios were calculated according to reported methods [42-46]. The variance and covariance statistics across replications were used to obtain estimates of the components of variation and their respective standard errors. The validity of the assumptions of diallel analysis was tested by the following formula (Sharma, 2003): $t^2=\{(n-2)/4[(MSS(Vr)- (MSS(Wr)]^2/\{MSS(Vr)x[MSS(Wr)-MSP(Wr.Vr)^2]\}$. Where: Wr = covariance between parents and their off-spring and Vr = variance of each array in which a particular parent is involved. Significance of calculated ''t'' value was tested against the tabulated ''F'' value with 4 and (n-2) degrees of freedom. Significant value indicates failure of the assumptions [44-45]. Another test was done by estimating the regression coefficient ''bWr.Vr'' of Wr on Vr as follows: $b_{Wr.Vr}$=[cov(Wr.Vr)/var Vr]=[MSP(Wr.Vr)/MSS(Vr)]. The standard error (SE) for the regression coefficient (b) value was estimated as follows: SE_b=[MSS(Wr)-bMSP(Wr.Vr)(n-2)]^{1/2} Where: n = number of parents. The significance of (b) different from zero (t1) and from unity(=1) (t2) can be tested by t-test as under: t_1 = (b-0)/SEb and t_2 = (1-b)/SEb. The foregoing values were tested against the ''t'' tabulated value for (n-2) degrees of freedom according to [43]. If all the assumptions were valid, the regression coefficient would be significantly different from zero but not from unity. Hayman (1954 a and b) derived the expectations for the statistics calculated from the F_1 diallel table and the expected values of the component variations using least squares [44-45]. The notations of Mather and Jinks (1971) are used and described as follows: V_{0L0} (V_p) (variance of the parents) = D + Ê, V_{1L1}(V_r)(mean of all the V_r values) = ¼ D - ¼ F + ¼ H_1 + ¼ H_2 + [Ê + Ê (n-2)/2n2], Vr (variance of all the progenies in each parental

array) = ¼ D + ¼ H_1 - ¼ H_2 - ¼ F + $(n+1)/2n_2$ Ê, W_{0L01}(Wr) (mean of all the Wr. values)= ½ D - ¼ F + Ê/n, $(M_{L1} - M_{L0})^2$ = dominance relationship = ¼ h^2 + $[(n-1) Ê/n^2)]$ [47]. The components of Ê, D, H_1, H_2, h^2 and F were estimated in F_1 as follows: Ê = [(Errors S.S. + Reps S.S.)/r]/[(r-1) + (c-1) (r-1)]. D = V_{0L0} − Ê. F = 2 V_{0L0} - $4W_{0L01}$ - [2Ê (n-2)/n]. H_1 = V_{0L0} + 4 V_{0L1} - $4W_{0L01}$ - [Ê (3n-2)/n]. H_2 = 4 V_{1L1} - 4 $V0_{L1}$ - 2Ê. h^2 = $4(M_{L1} - M_{L0})^2$ - [4Ê $(n-1)/n^2$]. Where n = number of parents. Ê = expected environmental component of variance. D = variance due to additive effects of the genes. F = mean of the covariance of additive and dominance effects across all arrays. H_1= variance component due to dominance deviation. H_1=[1-(u-v)^2], where, u and v are the proportions of positive and negative genes, respectively in the parents. h^2= algebraic sum of dominance effects across all loci in heterozygous phase in all crosses. The following genetic parameters were also calculated: Average degree of dominance is estimated as $(H_1/D)^{1/2}$. 1. If the of this ratio is zero, there is no dominance. 2. If it is greater than zero, but less than one, there is partial dominance. 3. If it is equal to 1, there is complete dominance. 4. If it is greater than 1, it indicates over dominance. Ratio of dominant and recessive genes in the parents (K_D/K_R) is estimated as follows: K_D/K_R = $[(4DH_1)^{1/2}+ F]/[(4 DH_1)^{1/2} - F]$ If K_D/K_R ≈1.0, it means nearly equal proportion of dominance and recessive alleles in parents, *i.e.* symmetrical distribution; p = q = 0.5. Any deviation from 1.0 indicates asymmetry of distribution (p # q). Thus: Ratio > 1 refers to excess of dominant alleles and minority of recessive alleles (p > q). Ratio < 1 means minority of dominant alleles and excess of recessive alleles (p < q). The ratio of dominant genes with positive or negative effects in parents ($H_2/4H_1$) was determined. The maximum theoretical value of 0.25 for this ratio arises when, p = q = 0.5 at all loci. A deviation from 0.25 would stem when p ≠ q. Thus: if this ratio ≈ 0.25, it means symmetrical distribution of positive and negative dominant genes in parents, while if this ratio ≠ 0.25, it means asymmetry of distribution. Narrow-sense heritability (h^2n) was estimated using the following equation: h^2_n = [1/4D / (1/4D + 1/4H_1– 1/4F + Ê]. The expected genetic advance (GA) from direct selection as a percentage of the mean (x) was calculated according to Singh and Narayanan (2000) based on 1% selection intensity as follows: GA = $100[(k.h^2_n \delta_{ph})/x]$ Where: k = 2.64 (selection differential for 1% selection intensity), and δ_{ph}= square root of the dominator of the narrow sense heritability [48].

c. V_r-W_r graphs

Based on parental variance (V_r) and parent-offspring co-variance (W_r) relationships diallel cross progenies, a two-way representation of parental arrays along a regression line of W_r on V_r was first suggested by Jinks and Hayman (1953) and later refined by Hayman (1954 a) [43-44]. This two directional depiction is widely known as the W_r - V_r graph. For drawing the regression line, the expected W_{rei} values were calculated as follows: W_{rei} = W_r − b$\overline{V}_r$ + b $\overline{V}_{ri}$, where: W_r is array mean of variances, V_r= array mean of covariance and b= regression coefficient. The regression line was drawn by plotting W_{rei} against V_r values. The point of interception of the regression line with W_r ordinate, *i.e.*, (a) was obtained by the following equation: a = $\overline{W}_r$ - b $\overline{V}$r**.**

Results and Discussion
1. Combining ability variances

Variances estimates for general (GCA) and specific (SCA) combining ability of the F_1 diallel crosses of wheat for combined data across two years under high and low levels of nitrogen are presented in Tables (2 and 3). Mean squares due to genotypes were highly significant for all studied traits under the two levels of N. Mean squares due to GCA and SCA were also highly significant for all studied traits, except NHI for SCA under low-N, indicating that both additive and non-additive gene effects play an important role in the inheritance of most studied traits under different N application rates.

In the present study, the magnitude of GCA mean squares was higher than that of SCA, since the ratio of GCA/ SCA mean squares was higher than unity for all studied traits under the two levels of N, except GPC under low-N, where the ratio was below unity. Higher GCA/SCA ratio than unity, suggested the existence of a greater portion of additive and additive x additive than that of non–additive genetic variance in controlling the inheritance of these traits under the two levels of nitrogen.

Table 2: Mean squares due to general (GCA) and specific (SCA) combining ability and their interactions with years (Y) for studied traits in F_1' s under high N conditions across two years.

SV	df	MS			
		NUE	NUPE	NUTE	NHI
Genotypes (G)	20	14.06**	22.94**	0.11**	10.87**
GCA	5	33.28**	64.84**	0.15**	16.95**
SCA	15	7.65**	8.98**	0.10**	8.84**
GCA xY	5	1.81*	0.68	0.01**	13.24**
SCA xY	15	1.24*	2.81**	2.28**	13.97**
GCA/SCA		4.35	7.22	1.52	1.92
GCA xY /SCAxY		1.46	0.24	0.01	0.95
error	80	0.51	0.36	0.003	1.70

* &** indicate significant at 0.05 and 0.01 probability levels, respectively

Table 3: Mean squares due to general (GCA) and specific (SCA) combining ability and their interactions with years (Y) for studied traits in F_1 diallel crosses under low N conditions across two years.

SV	df	MS			
		NUE	NUPE	NUTE	NHI
Genotypes (G)	20	99.87**	291.41**	3.87**	41.32**
GCA	5	352.82**	674.51**	5.97**	120.84**
SCA	15	15.56**	163.71**	3.17**	14.81
GCA xY	5	7.39**	9.40**	4.71**	27.0*
SCA xY	15	7.73**	21.45**	2.29**	38.74**
GCA/SCA		22.67	4.12	1.88	8.15
GCA xY /SCAxY		0.95	0.43	2.05	0.70
error	80	2.72	3.832	0.012	9.73

* &** indicate significant at 0.05 and 0.01 probability levels, respectively

The greater importance of GCA relative to SCA variance as observed in this study was also reported [49-56]. for GYPP and its components. Le Gouis *et al*. (2002) reported that in N-limited diallel F_1 hybrids between modern French cultivars found markedly higher GCA/SCA ratios for grain yield, grain N yield and total above ground N than in those grown under high N nutrition [38]. A similar preponderance of GCA effects for N uptake and NUTE was identified in F_2 and F_3 progenies of factorial hybrids between modern and exotic cultivars of barley grown under reduced N fertilization (Gorny and Ratajezak, 2008) [57].

Results in Tables (2 and 3) indicate that mean squares due to SCA x year interaction were significant ($P \leq 0.01$) for the all studied traits under the two levels of N, except GPC and NHI under low N, indicating that non-additive variance was affected by years. Mean squares due to the GCA x year interaction were also significant ($P \leq 0.05$ or 0.01) for all studied traits under high and low N, except for NUPE under high-N and GPC under low-N, which were not significant, indicating that additive variance for most cases differs from one year to another. The mean squares due to SCA x year was higher than those due to GCA x year for all studied traits under both high and low- N, except for NHI and NUE under high–N, suggesting that SCA (non–additive) variance (in most cases) is more affected by year than GCA (additive) variance.

2. GCA effects

Estimates of general combining ability (GCA) effects of parents for studied traits under the two levels of nitrogen across two years are presented in Table 4 (high-N) and Table 5 (low-N). Favorable significant GCA effects were expressed by positive estimates for all studied traits.

In general, the best general combiners in F_1's for NUE and NUPE were L26 followed by L27 and L25 parents under both high-N and low-N. For NUTE, the best combiners were L27, Gem9 and Gz168 under high-N and Gem9 under low-N. However, for NHI, the best combiners were Gem9 under high-N and Gem9 and Gz 168

under low-N. On the contrary, the worst general combiners in F_1's were Gem 9, Gem 7 and Giza 168 for NUE and NUPE traits under both high–N and low–N environments.

Table 4: Estimates of general combining ability effects (g_i) of studied traits in F_1's diallel crosses under high-N conditions across two years.

Parents	NUE	NUPE	NUTE	NHI
L25	-0.74**	0.71**	-0.10**	-0.22
L26	1.27**	1.79**	-0.03*	-0.31
L27	0.77**	0.21	0.04*	-0.76*
Gem 7	-0.39*	-0.29	-0.01	-0.12
Gem 9	-0.73**	-1.27**	0.04*	0.75*
Giza 168	-0.19	-1.15**	0.05**	0.66
SEgi	0.22	0.19	0.017	0.41
SEgi-gj	0.35	0.29	0.029	0. 64

* &** indicate significant at 0.05 and 0.01 probability levels, respectively

Table 5: Estimates of general combining ability effects (g_i) of NUE traits in F_1's diallel crosses under low-N conditions across two years

Parents	NUE	NUPE	NUTE	NHI
L25	2.35**	2.21*	-0.14**	-1.82*
L26	2.35**	5.17**	-0.23**	-0.89
L27	2.46**	2.03**	-0.11*	-0.55
Gem 7	-2.20**	-2.47*	-0.13*	-0.55
Gem 9	-3.60**	-4.94**	0.71**	1.38*
Giza 168	-1.36**	-2.00*	-0.10*	2.43**
SE_{gi}	0.49	0.62	0.04	0.98
SE_{gi-gj}	0.81	0.95	0.05	1.51

* &** indicate significant at 0.05 and 0.01 probability levels, respectively

It is worthy to note that the best general combiners in this study (L25, L26 and L27) showed also high *per se* performance for the most studied NUE traits under both high and low–N environments.

3. SCA effects

Estimates of specific combining ability (SCA) effects of the F_1 crosses for the studied traits under the two levels of N are presented in Tables (6 and 7). The best crosses in SCA effects were considered those exhibited significant positive SCA effects for all studied traits. The rank of F_1 crosses for SCA effects was changed from under high–N to under low–N conditions. Under high–N, the best cross for SCA effects of was the F_1 cross L26 x Gz 168 followed by the F_1 L27 x Gem7 in two traits (NUE and NUTE), the F_1 L25 x L26 for NUTE and the F_1 L25 x Gz 168, L26 x L27, and L26 x Gem7 for NUPE and L27 x Gem9 for NUTE. These F_1's include at least one parent of high GCA effects under high N.

Table 6: Estimates of specific combining ability effects ($\hat{s}_{ij}$) of F_1's under high- N conditions across two seasons

Crosses	NUE	NUPE	NUTE	NHI
L25 X L26	0.63	-0.79	0.09*	1.52
L25 X L27	-1.58*	-0.16	-0.09*	0.55
L25 X Gem 7	-0.49	0.004	-0.03	-0.30
L25 X Gem 9	0.46	0.85	-0.03	-0.45
L25 X Gz 168	0.39	1.19*	-0.05	0.41
L26 X L27	-0.18	1.27*	-0.09*	0.91
L26 X Gem 7	-0.44	1.87**	-0.12*	0.12
L26 X Gem 9	0.60	0.16	0.01	1.29
L26 X Gz 168	1.53**	-0.92	0.14**	0.97
L27 X Gem 7	2.64**	-2.18**	0.33**	-0.36

L27 X Gem 9	0.52	-2.13**	0.19**	-0.32
L27 X Gz168	0.44	1.00*	-0.05	-0.70
Gem 7 X Gem9	-0.90	0.87	-0.13*	-0.24
Gem 7X Gz 168	0.52	-0.06	0.03	-1.89
Gem 9X Gz 168	-0.46	-0.37	-0.01	-0.10
SE_{Sij}	0.62	0.78	0.06	1.91
$SE_{Sij}-_{Sik}$	0.92	0. 89	0.09	1.27
$SE_{Sij}-_{Skl}$	0.85	0.72	0.07	1.68

* &** indicate significant at 0.05 and 0.01 probability levels, respectively

Under low–N conditions, the best SCA effects were shown the by F_1's L25 x Gz168 for NUE and NUPE, L2 x Gem9 and L27 x Gem9 for NUPE and L25 x L 26, L25 x L27 and L27 x Gem9 for NUTE trait. Again these F_1's include at least one parent of high GCA effects under low-N.

Table 7: Estimates of specific combining ability effects ($\hat{s}_{ij}$) of F_1's under low-N conditions across two years.

Crosses	NUE	NUPE	NUTE	NHI
L25 X L26	0.59	-5.22**	0.30*	1.75
L25 X L27	-0.40	-4.31*	0.24*	0.39
L25X Gem 7	2.09	2.43	0.10	1.36
L25 XGgem 9	-2.08	4.81*	-0.92**	2.11
L25 X Gz 168	2.46	6.15**	-0.01	-2.11
L26 X L27	1.19	3.80*	0.04	-0.66
L26 X Gem 7	-0.19	0.36	0.09	-0.81
L26 X Gem 9	-0.88	5.42**	-0.88**	1.90
L26 X Gz 168	-1.79	-0.8	0.06	-2.15
L27 X Gem 7	1.54	-2.88	0.28*	-1.48
L27 X Gem 9	-1.57	10.63**	-1.07**	-1.42
L27 X Gz168	0.16	-4.22*	0.27*	0.67
Gem 7 X Gem9	-0.37	0.04	-0.68**	-0.35
Gem 7 X Gz 168	-1.09	2.12	-0.02	0.43
Gem 9 X Gz 168	-1.09	-1.41	-0.42**	0.29
SE_{Sij}	2.43	2.88	0.16	4.58
$SE_{Sij}-_{Sik}$	2.13	1.91	0.14	3.03
$SE_{Sij}-_{Skl}$	1.97	2.34	0.13	4.02

* &** indicate significant at 0.05 and 0.01 probability levels, respectively

Results of Gorny *et al*. 2011 [58] on wheat crosses appear to be in accord with similar N-Shortage – induced increases in the importance of non – additive effects for grain yield and components of NUE previously reported in maize [54-55 59-60] and those for NUE in grain sorghum [61]. Gorny *et al*. (2011) reported that under high N-fertilization, the efficiency components were incanted in a manner favorable for wheat selection (preponderance of additive effects) however the enhanced contribution of non-additive gene effects and increased dominance under N-limited conditions could impede wheat selection to improve the N efficiency and adaptation to less luxurious fertilization regimes [57]. They concluded that selection methods that eliminate masking non-additive influences and take advantage of the additive variance should be employed to improve those traits.

4. Gene action, heritability and selection gain

Estimates of genetic components and ratios for studied nitrogen efficiency traits in F_1's under high- and low-N environments across two years are presented in Table (8). The dominance genetic component of variation (H_1) was highly significant for all studied traits under both high and low-N environments, indicating that the dominance gene effects in F_1's of this experiment are important for the inheritance of all studied NUE traits under low-N and high-N.

The additive component of variation (D) was also significant (P≤ 0.01or 0.05) for all studied traits in F_1's under both high- and low- N, indicating that selection may be efficient for improving all studied NUE traits. However, the magnitude of dominance (H_1) was much greater than that of additive (D) component for all studied traits in F_1's under both high N and low N, except for NUE trait under low-N, where the opposite was true, i.e. the additive was greater than dominance variance. This indicates that the dominance gene effects are more important than additive in F_1's and plays the major role in the inheritance of most studied NUE traits under both N environments. The NUE trait of F_1's under low-N showed more importance for additive than dominance variance. Selection for high values of this trait in the segregating generations of the studied diallel crosses under low-N would be efficient for obtaining improved high N- efficient wheat genotypes.

Table 8: Estimates of genetic parameters and ratios for NUE traits under high-and low-N in F_1 populations of 15 diallel crosses across two seasons

Variance components	NUE	NUPE	NUTE	NHI
High-N				
E	0.17	0.12	0.001	0.67*
D	1.58**	4.64**	0.01*	0.14*
H_1	4.33**	5.89**	0.07*	5.33**
H_2	3.86**	5.14**	-1.92	3.52**
F	-1.13*	-0.38*	-0.77*	0.89**
h^2	1.92**	-0.02	0.00	-0.40*
$(H_1/D)^{1/2}$	1.66	1.13	3.59	6.22
$H_2/4H_1$	0.22	0.22	0.23	0.15
K_D/K_R	0.64	0.93	0.78	2.73
h^2/H_2	0.50	0.01	0.07	-0.12
$h^2_b\%$	91.20	96.00	96.60	63.85
$h^2_n\%$	39.50	40.70	8.50	1.90
GA%	9.44	11.25	4.00	0.12
Low-N				
E	0.93	0.94	0.00	3.24**
D	26.63**	51.30**	0.01*	1.97**
H_1	7.17**	71.28**	0.12*	3.20**
H_2	0.08	17.61*	0.01	10.60**
F	6.15**	66.70**	0.10	1.72**
h^2	-2.34**	15.68**	0.02	-3.85**
$(H_1/D)^{1/2}$	0.52	1.18	2.85	1.27
$H_2/4H_1$	0.21	0.23	0.22	0.13
K_D/K_R	0.84	1.30	1.70	0.13
h^2/H_2	0.01	0.26	0.12	6.17
$h^2_b\%$	90.75	96.60	87.30	41.00
$h^2_n\%$	66.90	46.36	11.11	9.00
GA%	19.62	22.43	5.63	1.02

The overall dominance effects of heterozygous loci in Hayman's model (h^2) controlling all studied traits of F_1's under both high- and low-N environments, except NUPE and NUTE under high-N and NUTE under low-N were significant (P< 0.01 or 0.05); that could be due to the presence of a considerable amount of dominant genes for most studied NUE traits in the parental genotypes. The average degree of dominance $(H_1/D)^{1/2}$ in F_1's was in the range of over-dominance (greater than unity) for all studied traits under the two levels of nitrogen, except for NUE trait under low-N, which showed partial dominance (less than unity). Lower ratio of ($H_2/4H_1$) than 0.25 obtained in the present study (Table 8) indicated symmetrical distribution of positive and dominant genes in parents for all studied NUE traits of F_1's under both high- and low-N environments.

Under low-N, the ratio (K_D/K_R) in F_1's was more than unity for 3 out of 8 cases (NHI under high-N and NUPE and NUTE under low-N), indicating excess of dominant alleles and minority of recessive alleles (p>q). The

remaining cases in F_1's, where the ratio (K_D/K_R) was less than unity, indicated minority of dominant alleles and the excess of recessive alleles ($p<q$). Number of genes or groups of genes controlling the inheritance of a given trait (h^2/H_2) was one group of genes for all studied NUE traits of F_1's under both high-N and low-N environments (Table 8), except NHI under low-N which was controlled 7 genes or groups of genes.

Broad-sense heritability (h^2_b) in F_1's for all studied traits, except NHI under low-N (41.0%) and under high-N (63.85%) in this experiment was of high magnitude and ranged from 87.3% (NUTE under low-N) to 96.6% (NUTE under high-N and NUPE under low-N) (Table 8), indicating that environment had a small effect on the phenotype of F_1's for most studied NUE traits. Nitrogen harvest index (NHI) trait showed the smallest h^2_b value under both high and low N environments (63.85 and 41.00%, respectively), indicating a large effect of environment on this trait.

Narrow-sense heritability (h^2_n) in F_1's (Table 8) was of medium magnitude for NUE (39.50 and 66.90%) and NUPE (40.70 and 46.36%) under high-N and low-N, respectively. It is observed that NUTE trait recorded low h^2_n (8.50 and 11.11%), while NHI trait showed the lowest h^2_n value (1.90 and 9.00%) under high and low N, respectively. The big difference between broad- and narrow- sense heritability estimated from F_1's in this experiment, especially for NUTE trait could be attributed to the high estimates of dominance as compared to additive component. It is observed that narrow-sense heritability (h^2_n) in F_1's of the present study was generally higher in magnitude under low-N than under high-N for all studied NUE traits. This increase in h^2_n under low-N compared to high-N was more pronounced in NUE and NHI traits. Our results are in agreement with some researchers [54-56, 62-69] who support the idea that heritability is higher under stressed than non-stressed environment. On the contrary, other investigators reported that heritability is higher under good (non-stressed) environment [70-73].

Expected genetic advance (GA) from selection (based on 1% selection intensity) across two years ranged from 0.12% for NHI under high-N to 22.43% for NUPE under low-N-N. In general, the values of GA were higher under low-N than under high-N for all studied NUE traits. The traits NUE and NUPE showed much higher estimates of GA (19.62 and 22.43%, respectively) under low-N than those under high-N (9.44 and 11.25%, respectively). These results indicated that to improve nitrogen use efficiency traits in the present germplasm, it is better to practice selection for these traits under low-N conditions to obtain higher values of selection gain.

5. Graphical approach of diallel analysis

The graphical analysis of diallel crosses proposed by Hayman (1954 a and b) will be illustrated on the following bases: (1) The parabola marks the limits within which the variance-covariance points (V_r, W_r) should lie, (2) If the regression coefficient (b) of (V_r, W_r) is not different from unity, the genetic control system may be deduced to be additive without the complications of gene interactions, (3) Complementary gene effects (epistasis) generally reduces the covariance (W_r) disproportionally more than the variance (V_r) causing the slope of the regression line (b) to be less than unity, (4) When dominance is complete, the regression line with b = 1 would pass through the origin, (5) Over dominance causes the regression line to intercept the (W_r) axis below the origin, while partial dominance causes the regression line to intercept the (W_r) axis above the origin point, (6) The closeness of the regression line or (V_r, W_r) points to the limiting parabola indicates little dominance and (7) The order of the array points on the regression line is a good indicator of the dominance order of parents. The parents with more dominant genes are located nearer to the origin, while those with more recessive genes fall farther from the origin. The parents with equal frequencies of dominant and recessive genes occupy an intermediate position.

Based on the above information, in the F_1 diallel Hayman's approach**,** it is clear, from Figures (1 and 2) for 4 studied traits, that the regression line intercepted the Wr-axis below the origin, *i.e.* cutting the Wr-axis in the negative region (intercept= a < 0 (negative)) or D (additive variance) < H_1 (dominance variance), indicating the presence of over dominance for 2 out of 8 cases, namely NHI (Fig. 2) trait under high-N and low-N. The regression line passed through the origin (D = H_1), indicating complete dominance for 3 out of 8 cases in F_1's, namely NUPE (Fig. 1) and NUTE (Fig. 2) under low-N and NUTE (Fig. 2) under high-N. For 3 out of 8 cases in

F_1's, namely NUE under low-N (Fig.1) and NUE (Fig. 1) and NUPE (Fig. 1) under high-N, the regression line intercepted the Wr-axis above the origin (D > H_1), indicating partial dominance and the predominance of additive variance in these cases.
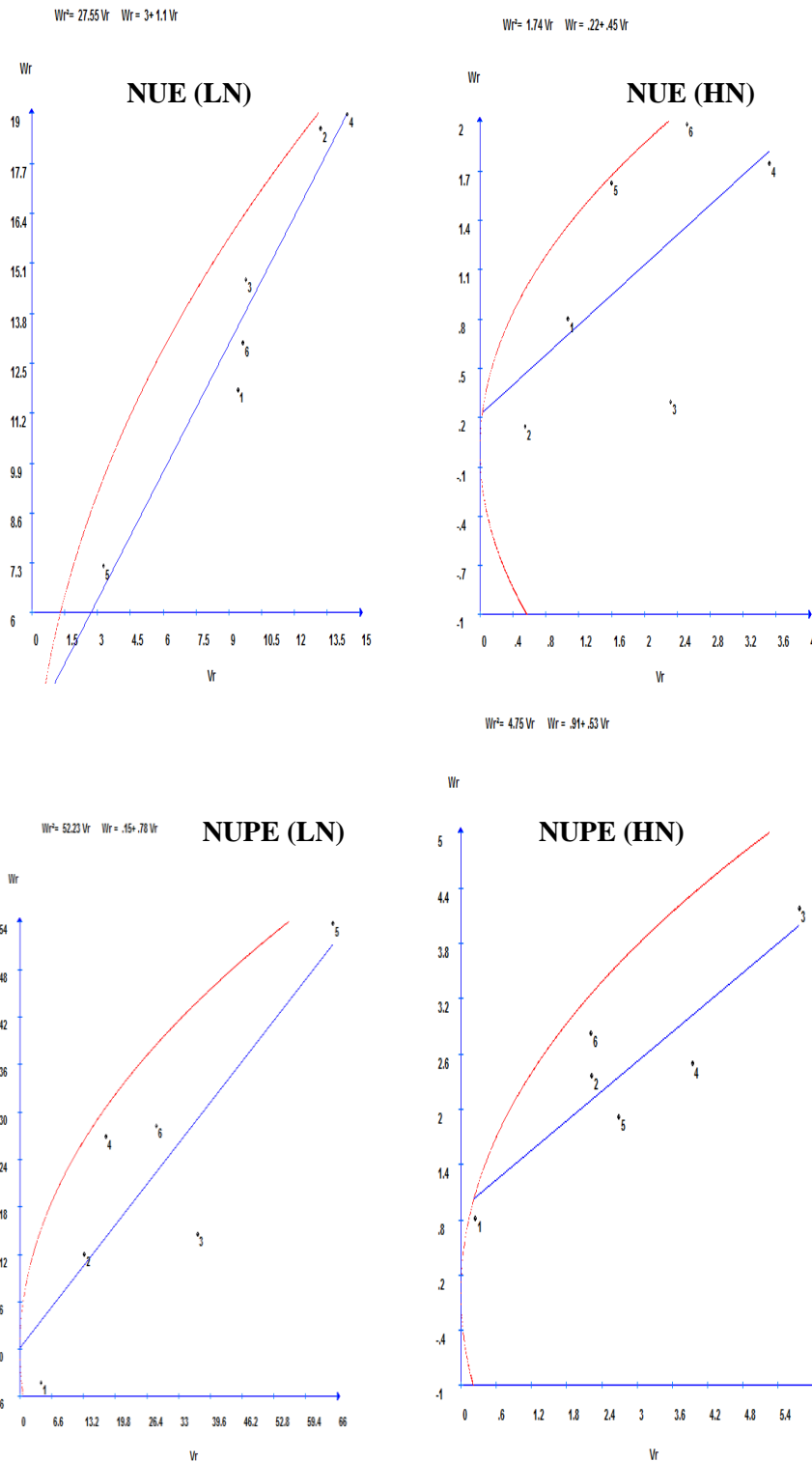

Figure 1: Wr-Vr graph of of nitrogen use efficiency (NUE) and uptake efficiency (NUPE) of F_1's for combined data across two seasons under low –(LN) and high-(HN) nitrogen.
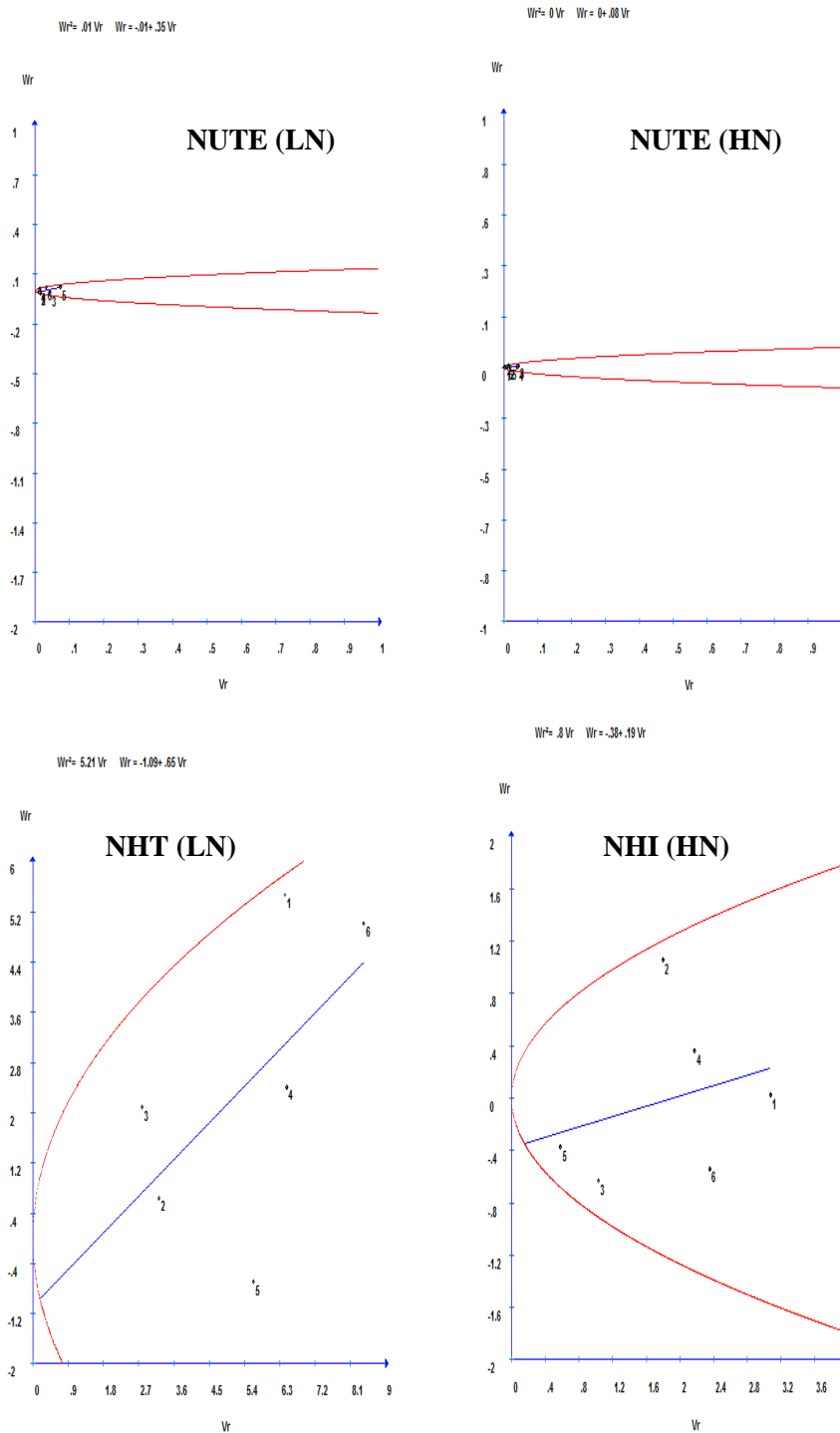
Figure 2: Wr-Vr graph of of nitrogen utilization efficiency (NUTE) and of nitrogen harvest index (NHI) of F₁'s for combined data across two seasons under low –(LN) and high-(HN) nitrogen.

The dispersion of parents (1 = L25, 2 = L26, 3 = L27, 4 = Gem7, 5 = Gem9 and 6 = Gz168) around the regression line for NUE (Fig. 1) showed that under low-N, the parent 5 (Gem9) is close to the origin of the coordinate, and accordingly has more than 75% dominant genes, parents 1, 3 and 6 (L25, L27 and Gz168) have50-75% of dominant genes, while Parents 2 (L26) and 4 (Gem7) have mostly recessive genes. Under high-N, dispersion of parents around the regression line, for NUE trait (Fig.1) showed that parent 2 is very close to the origin, indicating that it contains more than 75% dominant genes, the parents 1 (L25) and 3 (L27) contain

50-75% of dominant genes, while parents 4 (Gem7), 5 (Gem9) and 6 (Gz168) are very far from the origin, indicating that they mostly contain recessive genes.

Regarding NUPE trait (Fig. 1), under low-N, the dispersion of parents around the regression line reveals that parents 1, 2 and 3 are close to the origin of the coordinate, and accordingly have > 75% of dominant genes while parents 4 and 6 have 50-75% of dominant genes, while parent 5 is far from the origin and therefore has < 25% of dominant genes. Under high-N, for NUPE, the dispersion of parents around the regression line showed that parent 1 is close to the origin of the coordinate, and accordingly has > 75% of dominant genes, parents 2, 4, 5 and 6 have 50-75% of dominant genes, while parent 3 is far from the origin, therefore it has < 25% of dominant genes.

With respect of NUTE trait (Fig. 2), all parents under both low-N and high-N environments located very close to the origin of coordinate, and accordingly have > 75% of dominant genes. For NHI trait (Fig.2), under low-N, the dispersion of parents around the regression line reveals that parents 2 and 5 are close to the origin of the coordinate, and accordingly have > 75% of dominant genes while parents 3 and 4 have 50-75% of dominant genes, while parents 1 and 6 are far from the origin and therefore have < 25% of dominant genes. Under high-N, for NHI, the dispersion of parents around the regression line showed that parents 3 and 5 are close to the origin of the coordinate, and accordingly have > 75% of dominant genes, parent 6 has 50-75% of dominant genes, while parents 1, 2 and 4 are far from the origin, therefore they have < 25% of dominant genes.

Conclusions

This study concluded that the best general combiners with positive effects for improvement of nitrogen use efficiency (NUE) and nitrogen uptake efficiency (NUPE) traits were L26, L27 and L25 inbred parents under both high-N and low-N. Under low-N, the best combiner for NUTE was Gem9 and for NHI was Gem9 and Gz 168. The best SCA effects under low–N conditions were shown by the F_1 L25 x Gz168 for NUE and NUPE, L2 x Gem9 and L27 x Gem9 for NUPE and L25 x L 26, L25 x L27 and L27 x Gem9 for NUTE trait. These genotypes could be of value in future wheat breeding programs for improving NUE traits. Genetic analysis indicated the involvement of additive and dominant types of gene action in the inheritance of NUE, NUPE, NUTE and NHI traits, suggesting that a simultaneous exploitation of both additive and dominance variance to improve these parameters could be achieved by reciprocal recurrent selection. Selection for high values of NUE in the segregating generations of the studied crosses under low-N would be efficient for obtaining improved high N-efficient wheat genotypes. Highest narrow-sense heritability was observed for NUE and NUTE traits under low-N, hence the role of additive variance was higher than dominance variance that is likely to involve a few major genes in the genetic control of these traits. The results indicated that, to improve nitrogen use efficiency using the present material, it is better to practice selection for these traits in segregating generations under low-N conditions to obtain higher values of selection gain.

References

[1]. FAOSTAT (2011). Food and Organization of the United Nations, Statistics Division. Available online at: http:// www.faostat.fao.org. (assessed 01 April 2015).

[2]. Le Gouis, J, Beghin, D., Heumez, E. and Pluchard, P. (2000). Genetic differences for nitrogen uptake and nitrogen utilization efficiencies in winter wheat. Eur. J. Agron. 12:163–173.

[3]. London, I. G. (2005). Nitrogen study fertilizes fears of pollution. Nature: 433: 791.

[4]. Bouwman, A. F., Boumans, L. J. M. and Batjes, N. H. (2002). Emissions of N_2O and NO from fertilized fields: Summary of available measurement data. Global Biogeochemical Cycles 16(4), 1058.

[5]. Vitousek, P. M., Aber, J. D., Howarth,, R. W., Likens, G. E., Matson, P. A., Schindler, D. W., Schlesinger, W. H. and Tilman, D. G. (1997). Human alteration of the global nitrogen cycle: sources and consequences. Ecological Applications 7(3), 737–750.

[6]. El Bassam, N. (1998) A concept of selection for 'low-input' wheat varieties. Euphytica 100:95–100.

[7]. Good, A.G., Shrawat, A.K., Muench, D.G. (2004). Can less yield more? Is reducing nutrient input into the environment compatible with maintaining crop production? Trends Plant Sci 9:597–605.

[8].   Fageria, N.K. and Baligar, V.C. (2005). Enhancing nitrogen use efficiency in crop plants. Adv. Agron. 88:97-185.

[9].   Phillips, S.L. and Wolfe M.S. (2005) Evolutionary plant breeding for low input systems. J. Agric. Sci. 143:245–254.

[10]. Muurinen, S., Slafer, G.A., and Peltonen-Sainio, P. (2006). Breeding effects on nitrogen use efficiency of spring cereals under northern conditions. Crop Sci. 46:561-568.

[11]. Hirel, B., Le Gouis, J., Ney, B. and Gallais, A. (2007). The challenge of improving nitrogenuse efficiency in crops plants: towards a more central role of genetic vari-ability and quantitative genetics within integrated approaches. J. Exp. Bot. 58,2369–2387.

[12]. Lammerts van Bueren, E.T., Østergard, H., Goldringer,, I. and Scholten O. (2008). Plant breeding for organic and sustainable, low input agriculture: Dealing with genotype-environment interactions. Proceedings of the EUCARPIA symposium of working group organic plant breeding, 7–9 Nov 2007, Wageningen. Euphytica 2008, doi:10.1007/s10681-008-9731-4, pp 1–72.

[13]. Sylvester-Bradley, R. and Kindred, D. R. (2009). Analysing nitrogen responses of cereals to prioritize routes to the improvement of nitrogen use efficiency. Journal of Experimental Botany 60(7), 1939–1951.

[14]. Sattelmacher, B., Horst, W.J. and Becker, H.C. (1994). Factors that contribute to genetic variation for nutrient efficiency of crop plants. Z Pflanzenerna¨hr Bodenk 157:215–224

[15]. Moll, R.H., Kamprath, E.J. and Jackson, W.A. (1982). Analysis and interpretation of factors which contribute to efficiency to nitrogen utilization. Agron. J. 75: 562–564.

[16]. Huggins, D.R. and Pan, W.L. (2003). Key indicators for assessing nitrogen use efficiency in cereal-based agroecosystems. J. Crop Prod. 8:157–185.

[17]. Dhugga, K.S. and Waines J.G. (1989). Analysis of nitrogen accumulation and use in bread and durum wheat. Crop Sci. 29:1232–1239.

[18]. Ortiz-Monasterio, J.I., Sayre, K.D., Rajaram, S. and McMahon, M. (1997). Genetic progress in wheat yield and nitrogen use efficiency under four N rates. Crop Sci. 37(3): 898-904.

[19]. Gorny, A.G., Garczynski, S., Banaszak, Z. and Ługowska, B. (2006). Genetic variation in the efficiency of nitrogen utilization and photosynthetic activity of flag leaves among the old and modern germplasm of winter wheat. J. Appl. Genet. 47:231–237.

[20]. Laperche, A., Brancourt-Hulmel, M., Heumez, E., Gardet, O. and Le Gouis, J. (2006). Estimation of genetic parameters of a DH wheat population grown at different N stress levels characterized by probe genotypes. Theor. Appl. Genet. 112:797–807.

[21]. Kichey, T., Hirel, B., Heumez, E., Dubois, F. and Le Gouis, J. (2007). Wheat genetic variability for post-anthesis nitrogen absorption and remobilisation revealed by 15N labeling and correlations with agronomic traits and nitrogen physiological markers. Field Crop Res 102:22–32.

[22]. Baresel, J.P., Zimmermann, G. and Reents, H.J. (2008). Effects of genotype and environment on N uptake and N partition in organically grown winter wheat (*Triticum aestivum* L.) in Germany. Euphytica 163:347–354.

[23]. Barraclough, P.B., Howarth, J.R., Jones, J., Lopez-Bellido, R., Parmar, S., Shepherd, C.E. and Hawkesford, M.J. (2010). Nitrogen efficiency of wheat: genotypic and environmental variation and prospects for improvement. Eur. J. Agron. 33:1–11.

[24]. Van Ginkel, M., Ortiz-Monasterio J.I., Trethowan R. and Hernandez E. (2001). Methodology for selecting segregating populations for improved N-use efficiency in bread wheat. Euphytica 119:223–230.

[25]. Brancourt-Hulmel, M., Heumez, E., Pluchard, P., Beghin, D., Depatureaux, C., Giraud, A. and Le Gouis, J. (2005). Indirect versus direct selection of winter wheat for low-input or high-input levels. Crop Sci. 45:1427–1431.

[26]. Loschenberger, F., Fleck, A., Grausgruber, G., Hetzendorfer, H., Hof, G., Lafferty, J., Marn, M., Neumayer, A., Pfaffinger, G. and Birschitzky,J. (2008). Breeding for organic agriculture—the example of winter wheat in Austria. Euphytica 163:469–480

[27]. Wolfe, M.S., Baresel J.P., Desclaux D., Goldringer I., Hoad S., Kovacs G., Lo¨schenberger F., Miedaner T., Østergard H.and Lammerts van Bueren E.T. (2008). Developments in breeding cereals for organic agriculture. Euphytica 163:323–346.

[28]. Ceccarelli, S. (1996). Adaptation to low/high input cultivation. Euphytica. 92: 203-214.

[29]. Dawson, J.C., Huggins D.R. and Jones S.S. (2008). Characterizing nitrogen use efficiency in natural and agricultural ecosystems to improve the performance of cereal crops in low-input and organic agricultural systems. Field Crop Res 107:89–101.

[30]. Murphy, K.M., Campbell, K.G., Lyon, S.R., Jones, S.S. (2007). Evidence of varietal adaptation to organic farming systems. Field Crop Res. 102:172–177.

[31]. Gamzikova, O.I. (1992). Genetic aspects in edaphical adaptation of wheat (in Russian). Fizjologia Biohimija Kulturnyh Rastienij 24:419–428.

[32]. An, D.G., Su, J.Y., Liu, Q.Y., Zhu, J.G., Tong, Y.P., Li, J.M., Jing, R.L., Li, B. and Li, Z.S. (2006). Mapping QTLs for nitrogen uptake in relation to the early growth of wheat (*Triticum aestivum* L.). Plant Soil 284:73–84.

[33]. Habash, D.Z., Bernard, S., Schondelmaier, J., Weyen, J., Quarrie, S.A., (2007). The genetics of nitrogen use in hexaploid wheat: N utilisation, development and yield. Theor. Appl. Genet. 114, 403–419.

[34]. Laperche, A., Le Gouis, J., Hanocq, E. and Brancourt-Hulmel, M. (2008). Modelling nitrogen stress with probe genotypes to assess genetic parameters and genetic determinism of winter wheat tolerance to nitrogen constraint. Euphytica 161:259–271

[35]. Fontaine, J.X., Ravel, C., Pageau, K., Heumez, E., Dubois, F., Hirel, B. and Le Gouis, J. (2009). A quantitative genetic study for elucidating the contribution of glutamine synthetase, glutamate dehydrogenase and other nitrogen-related physiological traits to the agronomic performance of common wheat. Theor. Appl. Genet., 119:645–662.

[36]. Le Gouis, J, Beghin, D., Heumez, E. and Pluchard, P. (2002). Diallel analysis of winter wheat at two nitrogen levels. Crop Sci. 42:1129–1134.

[37]. Yildirim, M., Bahar B., Genc I., Korkmaz K. and Karnez E. (2007). Diallel analysis of wheat parents and their F2 progenies under medium and low level of available N in soil. J. Plant Nutr. 30:937–945.

[38]. A.O.A.C. (1990). Official Methods of Association of Analytical Chemists. 15th ed. Washington D.C.,USA. 290p.

[39]. Steel, R.G.D., Torrie, J.H. and Dickey, D. (1997). Principles and Procedure of Statistics. A Biometrical Approach 3rd Ed. McGraw Hill BookCo. Inc., New York. pp. 352-358.

[40]. Griffing, B. (1956). Concept of general and specific combining ability in relation to diallel crossing system. Aust. J. Biol. Sci. 9: 463-493.

[41]. Singh R.K. and Chaudhary, B.D. (1985). Biometrical Methods in Quantitative Genetics Analysis. 2nd ed. Kalyani Publishers,New Delhi, India.

[42]. Jinks, J. L. (1954). The analyses of continuous variation in diallel cross of Nicotiana rustica varieties. Genetics, 39: 767-88.

[43]. Jinks, J. L. and Hayman, I. (1953). The analysis of diallel crosses. Maize Genetics Cooperation Newsletter, 27: 48-54.

[44]. Hayman, B. L. (1954 a). The theory and analysis of diallel crosses. Genetics, 39: 789-809.

[45]. Hayman, B. L. (1954 b). The analysis of variance of diallel tables. Biometrics, 10:235-244.

[46]. Sharma, R. J. (2003). Statistical and Biometrical Techniques in Plant Breeding. New Delhi, Second Edition, 432 p.

[47]. Mather, K. and Jinks, J. L. (1971). Biometrical Genetics. Chapman and Hall, London, 382 p.

[48]. Singh, P. and Narayanan, S. S. (2000). Biometrical Techniques in Plant Breeding. Kalayani Publishers, New Delhi, India.

[49]. Larik, A.S.; Mahar, A.R. and Hafiz, H.M.I. (1995). Heterosis and combining ability estimates in diallel cross of six cultivars of spring wheat. Wheat Inform. Services, No. 80: 12-19.

[50]. Darwish, I.H.I. (2003). Diallel cross analysis of wheat (*Triticum aestivum* L.) under stress and normal irrigation treatments. Egypt. J. Plant Breed., 7: 253-269.

[51]. Riaz, R. and Chowdhry, M. A. I. (2003a). Estimation of variation and heritability of some physio-morphic traits of wheat under drought conditions. Asian J. Plant Sci., 2: 748-755.

[52]. Riaz, R. and Chowdhry, M.A. I. (2003b). Genetic analysis of some economic traits of wheat under drought condition. Asian J. Pl. Sci. 2: 790-796.

[53]. Al-Naggar, A.M.M., El-Kadi, D. A. and Abo–Zaid, Zeinab S. A. (2007). Inheritance of nitrogen use efficiency traits in grain sorghum under low-and high-N. Egypt. J. Plant Breed. 11 (3): 181-206.

[54]. Al-Naggar, A. M. M., Shabana, R., Atta, M.M.M. and Al-Khalil, T.H. (2015a). Regression of Grain Yield of Maize Inbred Lines and Their Diallel Crosses on Elevated Levels of Soil-Nitrogen. International Journal of Plant & Soil Science, Vol.4 (6): 499-512.

[55]. Al-Naggar, A. M. M., Shabana, R., Atta, M.M.M. and Al-Khalil, T.H. (2015b). Maize response to elevated plant density combined with lowered N-fertilizer rate is genotype-dependent. The Crop Journal, Vol. (3):96-109.

[56]. Al-Naggar, A. M. M., Shabana, R., Atta, M.M.M. and Al-Khalil, T.H. (2015c). Response of genetic parameters of low-N tolerance adaptive traits to decreasing soil-N rate in maize (*Zea mays* L.). Applied Science Reports, 9 (2): 110-122.

[57]. Gorny, A.G. and Ratajczak, D. (2008). Efficiency of nitrogen and phosphorus utilization in progenies of factorial crosses between European and exotic cultivars of spring barley. J. Appl. Genet. 49:349–355.

[58]. Gorny, A.G., Banaszak, Z., Lugowska, B. and Ratajczak, D. (2011). Inheritance of the efficiency of nitrogen uptake and utilization in winter wheat (*Triticum aestivum* L.) under diverse nutrition levels. Euphytica. 77: 191–206.

[59]. Di Fonzo, N., Motto, M., Maggiore, T., Sabatino, R. and Salamini, F.(1982). N-uptake, translocation and relationships among N-related traits in maize as affected by genotype. Agronomie 2:789–796.

[60]. Medici, L.O., Pereira, M.B., Lea, P.J. and Azevedo R.A. (2004) Diallel analysis of maize lines with contrasting responses to applied nitrogen. J. Agric. Sci. 142:535–541.

[61]. Al-Naggar, A.M.M., Shabana, R., Mahmoud, A. A. and Shaboon. S. A. M. (2008). Genetic improvement of maize for low-soil nitrogen tolerance *via* S$_1$ recurrent selection. Egypt. J. Plant Breed. 12 (2): 255-277.

[62]. Blum, A. (1988a). Breeding crop varieties for stress environments. Crit. Rev. Plant Sci. 2: 199-238.

[63]. Blum, A. (1988b). Plant breeding for stress environments. CRC press Inc., Florida, USA, 78-84.

[64]. Hefny, M. M. (2007). Estimation of quantitative genetic parameters for nitrogen use efficiency in maize under two nitrogen rates. Int. J. Pl. Breed. Genet., 1:54-66.

[65]. Al-Naggar, A.M.M., El-Kadi, D. A. and Abo–Zaid, Zeinab S. A. (2006). Genetic parameters of grain sorghum traits contributing to low – N tolerance. Egypt. J. Plant Breed. 10 (2) :79-102.

[66]. Al-Naggar, A.M.M., Atta, M. M. M. and Amein, M. M. (2009). Maize genotypic differences in nitrogen use efficiency under low soil-N conditions. Egypt. J. of Appl. Sci., 24(3B): 528-546.

[67]. Al-Naggar, A. M. M., Shabana, R. and Al-Khalil, T. H. (2011). Differential nitrogen use efficiency in maize genotypes of narrow- *vs* broad – base genetic background. Egypt. J. Plant Breed. 15(1): 41-56.

[68]. Al-Naggar, A. M. M. and Shehab- El- Deen, M.T. (2012a). Predicted and actual gain from selection for early maturing and high yielding wheat genotypes under water stress conditions. Egypt. J. Plant Breed. 16 (3) : 73 -92.

[69]. Al- Naggar, A. M. M., Abdel- Raouf, M. S., El- Borhamy, H. S. and Shehab- El- Deen M.T. (2012b). Gene effects controlling inheritance of earliness and yield traits of bread wheat under drought stress conditions. Egypt. J. Plant Breed. , 16(3): 41- 59.

[70]. Atlin, G.N . and Frey, K. J. (1990). Selecting oat lines for yield in low productivity Environments. Crop Science 30: 556-561.

[71]. Bänziger, M., Betrán, F.J. and Lafitte, H.R. (1997). Efficiency of high nitrogen selection environments for improving maize for low-nitrogen target environments. Crop Sci. 37: 1103-1109.

[72]. Rosielle, A.A. and Hamblin, J. (1981). Theoretical aspescts of selection for yield in stress and non-stress environments. Crop Sci., 21:43-46. doi:10.2135/cropsci1981.0011183X002100010013x.

[73]. Worku, M. (2005). Genetic and Crop-Physiological Basis of Nitrogen Efficiency in Tropical Maize. Ph.D. Thesis. Fac. Agric. Hannover Univ. Germany, 122 p.