



Matching the Optimum Plant Density and Adequate N-rate with High-density Tolerant Genotype for Maximizing Maize (*Zea mays* L.) Crop Yield

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Authors' contributions

This work was carried out in collaboration between all authors. Author AMMAN designed the study, wrote the protocol, and wrote the first draft of the manuscript. Authors RS and MMMA managed the literature searches and discussed the conclusion, while author THAK performed analyses of the study. All authors read and approved the final manuscript.

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ABSTRACT

Increasing plant density and improving N-fertilizer rate along with the use of high-density tolerant genotypes would lead to maximize maize grain productivity from unit land area. The objective of this investigation was to match the functions of optimum plant density and adequate nitrogen fertilizer application to produce the highest possible yields from unit area with the greatest maize genotype efficiency. A split-split plot design in randomized complete blocks arrangement with three replications was used for yield evaluation across two seasons (2012 and 2013). Main plots were assigned to three N-rates viz., 0 (LN), 120 (MN) and 240 (HN) kg/feddan; fed) (one fed = 4200 m²). Sub-plots were assigned to three plant densities viz., 20,000 (LD), 30,000 (MD) and 40,000 (HD) plant/fed and sub-sub plots to 23 maize genotypes (6 inbreds, 15 diallel F₁ crosses made among these inbreds and 2 check hybrids). Nine environments (E) had therefore been created (3 plant densities × 3 N levels). In general, the highest grain yield/plant (GYPP) was obtained from HN with LD (E1), while the highest grain yield/fed (GYPF) was obtained from HN with HD (E3). The environment LN and HD (E9) showed maximum reductions (70.9% and 67.6% in GYPP and 55.5%

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and 49.6% in GYPF for inbreds and hybrids, respectively) as compared to E1 as a result of both stresses (LN and HD). These reductions in grain yield were associated with reductions in all yield components like number of grains/plant (GPP), ears/plant (EPP), 100-grains weight (100-GW), harvest index, total dry matter, chlorophyll concentration index (CCI) and penetrated light; with maximum reduction in (GPP) and CCI. On the contrary, both stresses together caused increases in barren stalks (BS), anthesis-silking interval (ASI) and economic nitrogen use efficiency (NUE_e); with maximum increase in E9. The relationships between the nine environments and GYPF showed near linear regression function for inbreds L54, L29 and L55 and hybrids L18×L53 and L18×L55 with an optimum density of 20,000 plants/fed and N-rate of 240 kg N/fed and a curvilinear regression function for inbreds L17, L18 and L53 and the rest of hybrids with an optimum density of 40,000 plants/fed combined with N-rate of 240 kg N/fed. We could maximize GYPF in the present study to 60.4 ard/fed (one ard = 140 kg) for L17×L54 and 58.7 ard/fed for L17×L18 by using the high density and high N-rate; with a significant superiority in GYPF over the best check cultivar (SC-10) under E9 environment of 26.9% and 23.3%, respectively. The highest yielding genotypes under high-density in this study are characterized with one or more of adaptive traits to high-density and/or low-N.

Keywords: Quadratic regression; optimum plant density; appropriate N-rate; high-density tolerant maize; unit area productivity; ASI; prolificacy; NUE.

1. INTRODUCTION

Hybrid varieties currently released in Egypt by the National Maize Breeding Program (NMBP) are bred and grown at low plant density (24,000 plants/fed ca. 57,000 plants/ha), *i.e.*, almost half of the density used in developed countries. This may be one of the important reasons of getting lower grain yield from unit area of land grown by maize than that in the developed countries. One of the potential methods to maximize total production of maize in Egypt is through raising productivity per land unit area and thus upgrading our global rank in average productivity, especially with the irrigation system used in Egypt and good weather and soil conditions that suit maize crop as compared to other regions in the world. Grain yield/land unit area is the product of grain yield/plant and number of plants/unit area [1]. Maximum yield/unit area may be obtained by growing maize hybrids that can withstand high plant density up to 100,000 plants/ha (ca. 40,000 plants/fed) [2]. Average maize grain yield/unit area in the USA increased dramatically during the second half of the 20th century, due to improvement in crop management practices and greater tolerance of modern hybrids to high plant densities [3,4].

Trying to grow hybrid varieties released by NMBP at high plant densities causes a drastic reduction in grain yield/plant and consequent reduction in grain yield/unit area. The reason is probably due to the fact that these varieties are not tolerant to high plant densities, because of

their tallness, one-eared, decumbent leaf and large-size plant type. On the contrary, modern maize hybrids in developed countries are characterized with high yielding ability from unit area under high plant densities, due to their morphological and phenological adaptability traits, such as early silking, short anthesis-silking interval (ASI), less barren stalks and prolificacy. Duvick et al. [5] and Radenovic et al. [6] pointed out that maize genotypes with erect leaves are very desirable for increasing the population density due to better light interception.

To increase maize grain yield/unit area in Egypt, breeding programs should be directed towards the development of inbreds and hybrids that characterize with adaptive traits to high plant density tolerance. Although high plant density results in interplant competition (especially for light, water and nutrients), which affects vegetative and reproductive growth of maize [4,7]; the use of high-density tolerant hybrids and improving the fertilization management practices would overcome the negative impacts of such competition and lead to maximizing maize productivity from the same unit area. Maize tolerance to high plant population density was suggested as an alternative breeding strategy to improve tolerance to diverse abiotic stresses including drought and low N [8].

Nitrogen is an essential nutrient for maize crop growth [9]. It is the principal raw material required for the growth of plants and is found to be essential constituent of metabolically active compounds such as amino acids, proteins,

enzymes, coenzymes and some non-proteinous compounds [10,11]. Low N stress is one of the limiting factors under high plant density that limits maize production. Low-N availability in soils is an important yield-limiting factor frequently found in farmers' fields where fertilization is not commonly used and organic matter is rapidly mineralized [12]. Ears/plant and anthesis-silking interval are considered as the most important low-N adaptive traits [13]. Under these circumstances, since the smallholder farmers cannot afford additional inputs, it would be desirable to increase the tolerance of the crop to stresses that occur in their fields [14].

Matching the functions of optimum plant density and adequate nitrogenous fertilizer rate to produce the highest possible yields with the greatest maize hybrid efficiency has been the aim of many researchers [15-17]. Modern hybrids have shown tendencies to withstand higher levels of stresses (*i.e.*, low-N, high plant densities), which allow them to better sustainable and suitable photosynthetic rates, appropriate assimilate supplies, and maintain plant growth rates attributable to enhanced nitrogen use efficiency [18]. Along with the prevailing belief that high yields require more plants and that more plants require more N, the idea that different hybrids respond differently to both N and plant density should be considered. Moreover, different hybrids may behave differently in their tolerance to both low-N and high density stresses [19].

The objectives of the present investigation were:

- (i) To study the effect of stresses resulted from elevating plant density combined with low N-rate on studied traits of six inbreds and their diallel F_1 crosses and
- (ii) To match the function of optimum plant density and adequate nitrogenous fertilizer application with the greatest maize inbred or hybrid efficiency to produce the highest possible yields from unit land area.

2. MATERIALS AND METHODS

This study was carried out in 2012 and 2013 seasons at the Agricultural Experiment and Research Station of the Faculty of Agriculture, Cairo University, Giza, Egypt. Six maize (*Zea mays* L.) inbred lines (Table 1) in the 6th selfed generation (S_6), showing clear differences in

performance and general combining ability for grain yield/feddan (fed) under high plant density were chosen as parents of diallel crosses. In 2011 season, all possible diallel crosses (except reciprocals) were made among the six parents, so seeds of 15 direct F_1 crosses were obtained. Two field evaluation experiments were carried out in 2012 and 2013 seasons. Sowing date was on April 12 and May 2 in 2012 and 2013 seasons, respectively. Each experiment included 15 F_1 crosses, their six parents and two check cultivars, *i.e.*, SC 10 (white grains) obtained from the Agricultural Research Center (ARC) and SC 2066 (yellow grains) obtained from Hi-Tech Company-Egypt.

Evaluation in each season was carried out under 9 environments (from E1 to E9), *i.e.*, three nitrogen levels, namely, low- (LN), medium- (MN) and high-N (HN) by adding 0, 120 and 240 kg N/fed, respectively in two equal doses in the form of Urea before 1st and 2nd irrigations and three plant densities, namely, low- (LD), medium- (MD) and high-D (HD) plant density (20,000, 30,000 and 40,000 plant/fed, respectively) as follows: E1: HN-LD, E2: HN-MD, E3: HN-HD, E4: MN-LD, E5: MN-MD, E6: MN-HD, E7: LN-LD, E8: LN-MD and E9: LN-HD. Available nitrogen in 30 cm soil depth was determined immediately prior to sowing. The available nitrogen to each plant (including soil N and added N) was calculated for each environment and found to be 15.72, 10.48, 7.86, 9.72, 6.48, 4.86, 3.72, 2.48 and 1.86 g N/plant in 2012 season and 15.42, 10.28, 7.71, 9.42, 6.28, 4.71, 3.42, 2.28 and 1.71 g N/plant in 2013 season, with an average across the two seasons of 15.57, 10.38, 7.79, 9.57, 6.38, 4.79, 3.57, 2.38 and 1.79 g N/plant for the nine environments (E1 through E9), respectively. A split-split plot design in randomized complete blocks (RCB) arrangement with three replications was used. Main plots were devoted to nitrogen rates (LN, MN and HN). Sub-plots were assigned to plant density (LD, MD and HD). Sub sub-plots were devoted to 23 maize genotypes (6 parents, 15 F_1 's and 2 checks). Each sub sub-plot included one ridge of 4 m long and 0.7 m width. Seeds were sown in hills at 15, 20 and 30 cm apart, thereafter (before the 1st irrigation) excess seedling were thinned out to one plant/hill to achieve the 3 plant densities, *i.e.*, 20,000, 30,000 and 40,000 plant/fed, respectively. The soil of the experimental site was clayey loam. All other agricultural practices were followed according to the recommendations of ARC, Egypt.

Table 1. Designation, origin, and most important traits of 6 inbred lines (L) used for making diallel crosses of this study

| Entry designation | Origin | Institution (country) | Prolificacy | Productivity under high density and/or low-N |
|-------------------|----------|-----------------------|-------------|--|
| L17-Y | SC 30N11 | Pion. Int. Co. | Prolific | High |
| L18-Y | SC 30N11 | Pion. Int. Co. | Prolific | High |
| L53-W | SC 30K8 | Pion. Int. Co. | Prolific | High |
| L29-Y | Pop 59 | ARC-Thailand | One-eared | Low |
| L54-W | SC 30K8 | Pion. Int. Co. | One-eared | Low |
| L55-W | SC 30K8 | Pion. Int. Co. | One-eared | Low |

ARC = agricultural research center, Pion. Int. Co. = pioneer international company in egypt, SC = single cross, W = white grains, Y = yellow grains

Data were collected on 14 traits are not mentioned, namely, anthesis-silking interval (ASI), plant height (PH), barren stalks (BS) percentage, leaf angle (LANG) measured as the angle between stem and blade of the leaf just above ear leaf and chlorophyll concentration index (CCI) measured by Chlorophyll Concentration Meter, Model CCM 200 as the ratio of transmission at 931 nm to 653 nm through the leaf of top-most ear (<http://www.apogeeinstruments.co.uk/apogee-instruments-chlorophyll-content-meter-technical-information/>). At 80 days after sowing light intensity was measured and then penetrated light inside the canopy was calculated for each genotype, by using Lux-meter apparatus. The light intensity was measured in lux at 12 am (noon time) at the top of the plant and at the base of top-most ear. Penetrated light, at 80 days (PL-M80) after sowing, inside the canopy was measured as a percentage of light penetrated from the top of the plant to the base of top-most ear as follows:

Penetrated light =

$$100 \times \frac{\text{light intensity at the base of top – most ear}}{\text{light intensity at the top of the plant}}$$

At harvest, number of ears per plant (EPP), number of grain/plant (GPP), 100-grain weight (100-GW), grain yield/plant (GYPP), grain yield/feddan (GYPF), total above ground dry matter/plant (TDM), harvest index (HI) and economic nitrogen use efficiency (NUE_e) calculated as follows: $NUE_e = \frac{GDM}{N_s}$, where GDM = grain dry matter and N_s = available soil-N/plant according to Moll et al. [20].

Combined analysis of variance of split-split plot across the two seasons was performed if the homogeneity test was non-significant and LSD values were calculated to test the significance of

differences between means according to Snedecor and Cochran [21] by using statistical analysis system (SAS) computer software. Rank correlation coefficients were calculated between pairs of the studied (nine) environments for grain yield/feddan (GYPF). Computation was performed by using SPSS 17 computer software and the significance of the rank correlation coefficient was tested according to Steel et al. [22].

3. RESULTS AND DISCUSSION

3.1 Analysis of Variance

Combined analysis of variance across years (Y) of the split-split plot design for the studied 23 genotypes (G) of maize (6 inbreds +15 F₁s + 2 check commercial single-cross hybrids) under three plant densities (D) and three nitrogen (N) rates is presented in Table 2. Mean squares due to years were significant (P≤0.01) for all studied traits, except for anthesis-silking interval (ASI), plant height (PH), ears/plant (EPP) and 100-grain weight (100-GW), indicating significant effect of climatic conditions on most studied traits. Mean squares due to plant densities, N-rates and genotypes were significant (P≤0.01) for all studied characters, indicating that each of the three main factors in this study, *i.e.*, plant density, N-rate or genotype has an obvious effect on all studied traits. Mean squares due to the 1st order interaction, *i.e.*, N×Y, D×Y, G×Y, D×N, G×N and G×D were significant (P≤0.01) for all studied traits, except for chlorophyll concentration index (CCI) for N×Y, ASI and barren stalks (BS) for D×Y, DTS, ASI, BS, EPP, grain/plant (GPP), harvest index (HI) and economic nitrogen use efficiency (NUE_e) for G×Y and leaf angle (LANG) for G×N. Mean squares due to the 2nd order interaction, *i.e.*, D×N×Y and G×D×N were significant or highly significant for all studied traits, except, ASI, BS,

LANG, CCI and 100-grain weight (100-GW) for $D \times N \times Y$ and LANG for $G \times D \times N$.

On the contrary, mean squares due to $G \times N \times Y$ and $G \times D \times Y$ were insignificant for all studied traits, except for PH, BS and penetrated light at the top-most ear at 80 days from sowing (PL-M80) and grain yield/plant (GYPP) for $G \times N \times Y$ and PH, LANG, PL-M80, GPP and GYPP for $G \times D \times Y$ interaction, which were significant.

Mean squares due to the 3rd order interaction $G \times N \times D \times Y$ were significant ($P \leq 0.01$) for PH, PL-M80, GYPP, total dry matter (TDM), HI and NUE_e , indicating that the rank of maize genotypes differed from one nitrogen rate to another, from one density to another and from one year to another and that of selection for improved performance under a specific combination of soil nitrogen and plant density is possible as proposed by Kamara et al. [23], Al-Naggar et al. [24] and Al-Naggar et al. [25].

Combined analysis of variance of a randomized complete blocks design was performed for 14 traits in one set of diallel crosses among contrasting maize inbreds under each of the nine environments (from E1 to E9); representing combinations of 3 plant densities \times 3 N-rates, i.e., E1 = high nitrogen and low plant density (HN-LD), E2 = high nitrogen and medium plant density (HN-MD), E3 = high nitrogen and high plant density (HN-HD), E4 = medium nitrogen and low plant density (MN-LD), E5 = medium nitrogen and medium plant density (MN-MD), E6 = medium nitrogen and high plant density (MN-HD), E7 = low nitrogen and low plant density (LN-LD), E8 = low nitrogen and medium plant density (LN-MD), E9 = low nitrogen and high plant density (LN-HD) across two seasons (Data not presented). Mean squares due to genotypes, parents and crosses under all environments were highly significant for all studied traits, except ASI under E3, E5, E6 and E7, EPP under E8, HI under E7 and E9 and NUE_e under E9 for inbred parents and BS under E1 through E6 for F_1 crosses, indicating the significance of differences among studied parents and among F_1 diallel crosses in the majority of cases. Mean squares due to parents vs F_1 crosses were highly significant for all studied traits under all nine environments, except for CCI under E6 and 100-GW under E2, E3, E4 and E6, suggesting the presence of significant heterosis. Mean squares due to the interactions of parents \times years ($P \times Y$) and crosses \times years ($C \times Y$) were significant and highly significant for all studied traits under all environments, except ASI under E3, E5, E6, E7,

E8 and E9 for parents \times years and E1 and E6 for crosses \times years, PH under E1 for crosses \times years, BS under E6 and E8 for $P \times Y$ and under E1, E3, E6 and E8 for $C \times Y$, LANG under E7 for $P \times Y$, CCI under all environments for $P \times Y$ and E1 through E5 and E7 through E9 for $C \times Y$, EPP under E1, E2, E4 through E7 for $P \times Y$ and E2 and E4 for $C \times Y$, KPP under E1, E2, E4 through E6 for $P \times Y$, GYPP under E5 for $P \times Y$, GYPP under E3, E5 through E8 for $P \times Y$, TDM under E8 for $P \times Y$ and HI under E6 for $P \times Y$ and under E6 and E8 for $C \times Y$. Mean squares due to parents vs crosses \times years were significant and highly significant in 101 out of 162 cases, indicating that heterosis differ from season to season in those cases.

3.2 Effects of Combinations between Plant Densities and Nitrogen Levels

The effect of nine combinations between 3 rates of nitrogen and 3 plant densities on the studied traits is presented in Table 3. These combinations resulted in nine different environments, namely, E1 (HN-LD), E2 (HN-MD), E3 (HN-HD), E4 (MN-LD), E5 (MN-MD), E6 (MN-HD), E7 (LN-LD), E8 (LN-MD) and E9 (LN-HD). The highest GYPP was obtained from the E1 (a combination of highest N-rate and lowest plant density) which is logic, since available nitrogen for each plant was at maximum (15.57 g N/plant) across seasons and therefore we considered this environment as the best one for GYPP. Thus, the percent change, in different studied traits was calculated relevance to this environment, either in increase or decrease. Both stresses (nitrogen and plant density) were exhibited in order of $E9 > E8 > E6 > E5$ for severity, with a minimum severity by E5, while other environments exhibit only one stress (E2, E3 and E7) or no stress (E1 and E4). It can be observed that the rigidity of the stresses combinations on GYPP was at maximum (70.9% and 67.6% reduction for inbreds and hybrids, respectively) under the environment E9 (LN-HD), where both severe stresses (lowest nitrogen and highest plant density) exist. The reduction in GYPP due to the effect of both stresses in different combinations showed in order of $E9 > E8 > E6 > E5$ for severity (70.9%, 61.0%, 41.6% and 32.2%, respectively for parents and 67.6%, 59.5%, 39.6% and 29.6%, respectively for crosses). Significant reductions in GYPP of maize crosses observed in environments E8 and E9 relative to E1 (37.7% and 49.6%, respectively) were due to both N-rate and density stresses. It is observed that reduction in GYPP of

both inbreds and crosses was maximum under environment E9 (55.5% and 49.6%, respectively) due to both stresses (lowest nitrogen and highest plant density).

On the contrary, GYPF of both inbreds and hybrids under the environments E3 and E2 showed a tendency of increase over that under E1. The highest GYPF was obtained from E3 (a combination of highest density and highest N level) for inbreds and hybrids. Maximum increase (41.1% and 18.1%) in GYPF was shown by F₁ crosses under E3 (HN-HD) and E6 (MN-HD), respectively, due to high plant density. Reductions in grain yield resulted from both stresses (elevated plant densities and reduced N

levels) in both inbreds and hybrids were associated with reductions in all yield components (EPP, GPP, 100-GW), harvest index, TDM, CCI, LANG, PL-M80 and DTS. Such reductions were more pronounced in E9 environment (maximum stresses) followed in order of E8>E6>>E5 for severity. Maximum reductions were exhibited by grain/plant (81.9% and 82.0%) and CCI (76.5% and 76.8%) for inbreds and hybrids, respectively under E9 due to severe stresses of N-rate and plant density. On the other hand, the two stresses together (shown by the four environments E9, E8, E6 and E5) caused increases in BS, ASI and NUE_e.

Table 2. Analysis of variance of split-split plot design for studied 23 maize genotypes under three rates of nitrogen (N) and three plant densities (D) combined across two years

| SOV | df | Mean squares | | | | | | |
|---------------------|-----|--------------|---------------|-------------|-------------|------------|-----------|------------------------|
| | | ASI | PH | BS | LANG | CCI | PL-M80 | EPP |
| Years (Y) | 1 | ns | ns | ** | ** | ** | ** | ns |
| Nitrogen levels (N) | 2 | ** | ** | ** | ** | ** | ** | ** |
| N×Y | 2 | ** | ** | ** | ns | ns | ** | ns |
| Error | 8 | 0.01 | 308.6 | 0.04 | 6.1 | 53.1 | 27.0 | 0.02 |
| Densities (D) | 2 | ** | ** | ** | ** | ** | ** | ** |
| D×Y | 2 | ns | ** | ns | ** | ** | ** | ** |
| D×N | 4 | ** | ** | ** | ** | ** | ** | ** |
| D×N×Y | 4 | ns | ** | ns | ns | ns | ** | ** |
| Error | 24 | 0.005 | 64.1 | 0.01 | 1.6 | 11.4 | 4.6 | 0.01 |
| Genotypes (G) | 22 | ** | ** | ** | ** | ** | ** | ** |
| G×Y | 22 | ns | ** | ns | ** | ** | ** | ns |
| G×N | 44 | ** | ** | ** | ns | ** | ** | ** |
| G×N×Y | 44 | ns | * | ** | ns | ns | ** | ns |
| G×D | 44 | ** | ** | ** | ** | ** | ** | ** |
| G×D×Y | 44 | ns | ** | ns | ** | ns | ** | ns |
| G×D×N | 88 | ** | ** | ** | ns | ** | ** | ** |
| G×N×D×Y | 88 | ns | ** | ns | ns | ns | ** | ns |
| Error | 792 | 0.0006 | 54.1 | 0.002 | 0.6 | 6.5 | 2.2 | 0.007 |
| | | GPP | 100-GW | GYPF | GYPF | TDM | HI | NUE_e |
| Years (Y) | 1 | ** | ns | ** | ** | ** | ** | ** |
| Nitrogen levels (N) | 2 | ** | ** | ** | ** | ** | ** | ** |
| N×Y | 2 | ** | * | ** | * | ** | ** | ** |
| Error | 8 | 31899.8 | 10.6 | 873.4 | 26.8 | 725.4 | 24.1 | 14.9 |
| Densities (D) | 2 | ** | ** | ** | ** | ** | ** | ** |
| D×Y | 2 | ** | ** | ** | ** | ** | ** | ** |
| D×N | 4 | ** | ** | ** | ** | ** | ** | ** |
| D×N×Y | 4 | ** | ns | ** | * | ** | ** | ** |
| Error | 24 | 28090.1 | 2.8 | 252.4 | 6.9 | 175.5 | 7.5 | 8.0 |
| Genotypes (G) | 22 | ** | ** | ** | ** | ** | ** | ** |
| G×Y | 22 | ns | ** | ** | ** | ** | ns | ns |
| G×N | 44 | ** | ** | ** | ** | ** | ** | ** |
| G×N×Y | 44 | ns | ns | * | ns | ns | ns | ns |
| G×D | 44 | ** | ** | ** | ** | ** | ** | ** |
| G×D×Y | 44 | * | ns | ** | ns | ns | ns | ns |
| G×D×N | 88 | ** | ** | ** | ** | ** | ** | ** |
| G×N×D×Y | 88 | ns | ns | ** | ns | ** | ** | * |
| Error | 792 | 3228.5 | 1.5 | 27.1 | 0.8 | 23.3 | 1.4 | 1.3 |

* and ** indicate significant at 0.05 and 0.01 probability levels, respectively, ns = non-significant

Table 3. Means of studied traits and relative change (%) to E1 for nitrogen rates × plant densities interaction across nine environmental conditions combined across two seasons

| Para-meter | E1 | E2 | E3 | E4 | E5 | E6 | E7 | E8 | E9 |
|--|--|-------|-------|-------|-------|-------|-------|-------|-------|
| | HN-LD | HN-MD | HN-HD | MN-LD | MN-MD | MN-HD | LN-LD | LN-MD | LN-HD |
| Anthesis-silking interval (ASI) day | | | | | | | | | |
| Parents | 2.3 | 2.3 | 2.7 | 2.2 | 2.6 | 2.8 | 5.1 | 6.6 | 8.8 |
| Change | - | 1.2 | 21.0 | -1.2 | 16.0 | 22.2 | 125.9 | 195.1 | 292.6 |
| Crosses | 1.6 | 1.4 | 1.5 | 1.5 | 1.7 | 1.4 | 4.1 | 5.0 | 6.6 |
| Change | - | -12.1 | -4.3 | -2.1 | 5.7 | -13.5 | 162.4 | 217.7 | 320.6 |
| LSD 0.05 | N = 0.01, D = 0.01, G = 0.01, N×D = 0.01 | | | | | | | | |
| Plant height (PH) cm | | | | | | | | | |
| Parents | 195.4 | 200.8 | 212.4 | 196.9 | 204.0 | 204.7 | 177.6 | 178.8 | 189.9 |
| Change | - | 2.8 | 8.7 | 0.8 | 4.4 | 4.7 | -9.1 | -8.5 | -2.9 |
| Crosses | 219.9 | 226.6 | 242.3 | 228.9 | 233.8 | 255.7 | 200.9 | 206.2 | 218.3 |
| Change | - | 3.0 | 10.2 | 4.1 | 6.3 | 16.2 | -8.7 | -6.3 | -0.8 |
| LSD 0.05 | N = 2.65, D = 1.08, G = 2.77, N×D = 1.87 | | | | | | | | |
| Barren stalks (BS) % | | | | | | | | | |
| Parents | 4.3 | 5.8 | 9.6 | 9.3 | 13.4 | 13.0 | 30.4 | 40.5 | 43.3 |
| Change | - | 35 | 122 | 115 | 212 | 203 | 607 | 842 | 907 |
| Crosses | 0.1 | 0.0 | 0.1 | 0.3 | 0.4 | 0.6 | 16.5 | 18.9 | 22.1 |
| Change | - | -100 | -38 | 212 | 420 | 652 | 19249 | 22060 | 25719 |
| LSD 0.05 | N = 0.03, D = 0.02, G = 0.02, N×D = 0.03 | | | | | | | | |
| Leaf angle (LANG) ° | | | | | | | | | |
| Parents | 31.3 | 30.0 | 28.8 | 31.1 | 28.9 | 27.7 | 28.8 | 27.3 | 26.9 |
| Change | - | -4.2 | -8.0 | -0.7 | -7.7 | -11.7 | -8.1 | -13.0 | -14.3 |
| Crosses | 35.6 | 31.2 | 30.6 | 34.8 | 31.0 | 29.9 | 32.3 | 29.1 | 28.4 |
| Change | - | -12.3 | -14.2 | -2.3 | -12.8 | -16.0 | -9.4 | -18.2 | -20.3 |
| LSD 0.05 | N = 0.12, D = 0.33, G = 0.48, N×D = 0.57 | | | | | | | | |
| Chlorophyll concentration index (CCI) % | | | | | | | | | |
| Parents | 56.4 | 52.0 | 58.0 | 57.4 | 45.0 | 48.4 | 28.9 | 19.5 | 13.3 |
| Change | - | -7.9 | 2.7 | 1.7 | -20.3 | -14.1 | -48.8 | -65.5 | -76.5 |
| Crosses | 64.6 | 62.9 | 60.5 | 61.9 | 57.9 | 47.8 | 33.7 | 21.8 | 15.0 |
| Change | - | -2.5 | -6.2 | -4.1 | -10.4 | -26.0 | -47.8 | -66.2 | -76.8 |
| LSD 0.05 | N = 0.39, D = 0.98, G = 1.28, N×D = 1.70 | | | | | | | | |
| Penetrated light at the base of top-most ear at 80 day (PL-M80) % | | | | | | | | | |
| Parents | 11.2 | 10.3 | 8.7 | 12.5 | 11.5 | 9.9 | 14.5 | 12.4 | 11.2 |
| Change | - | -7.7 | -22.2 | 11.6 | 2.7 | -11.6 | 29.7 | 10.6 | 0.1 |
| Crosses | 9.5 | 8.1 | 6.8 | 10.9 | 9.4 | 7.8 | 13.7 | 11.0 | 9.0 |
| Change | - | -14.5 | -28.0 | 15.0 | -1.2 | -18.0 | 43.8 | 15.7 | -5.3 |
| LSD 0.05 | N = 0.32, D = 0.70, G = 0.56, N×D = 1.21 | | | | | | | | |
| Number of ears/plant (EPP) | | | | | | | | | |
| Parents | 1.2 | 1.2 | 1.0 | 1.1 | 0.9 | 0.9 | 0.9 | 0.8 | 0.6 |
| Change | - | 3.7 | -13.3 | -6.4 | -21.4 | -21.0 | -27.2 | -34.8 | -50.0 |
| Crosses | 1.4 | 1.3 | 1.1 | 1.3 | 1.1 | 1.0 | 1.0 | 0.9 | 0.7 |
| Change | - | -5.1 | -19.7 | -7.8 | -17.5 | -24.7 | -29.1 | -32.2 | -47.0 |
| LSD 0.05 | N = 0.02, D = 0.02, G = 0.03, N×D = 0.03 | | | | | | | | |
| Number of grains/plant (GPP) | | | | | | | | | |
| Parents | 924.1 | 859.9 | 680.2 | 679.5 | 505.0 | 479.0 | 326.3 | 246.8 | 167.5 |
| Change | - | -6.9 | -26.4 | -26.5 | -45.4 | -48.2 | -64.7 | -73.3 | -81.9 |
| Crosses | 1103.3 | 908.2 | 742.5 | 787.2 | 620.8 | 518.6 | 370.7 | 299.5 | 198.3 |
| Change | - | -17.7 | -32.7 | -28.7 | -43.7 | -53.0 | -66.4 | -72.9 | -82.0 |
| LSD 0.05 | N = 26.97, D = 22.57, G = 21.43, N×D = 39.09 | | | | | | | | |
| 100-grain weight (100-GW) g | | | | | | | | | |
| Parents | 40.2 | 36.1 | 33.7 | 35.8 | 32.0 | 29.7 | 27.1 | 25.5 | 21.6 |
| Change | - | -10.2 | -16.2 | -11.0 | -20.3 | -26.0 | -32.6 | -36.5 | -46.2 |
| Crosses | 39.4 | 36.9 | 33.8 | 35.5 | 32.9 | 29.8 | 27.8 | 28.7 | 25.3 |
| Change | - | -6.3 | -14.1 | -9.8 | -16.5 | -24.2 | -29.3 | -27.1 | -35.6 |
| LSD 0.05 | N = 0.49, D = 0.23, G = 0.46, N×D = 0.39 | | | | | | | | |

| Parameter | E1 HN-LD | E2 HN-MD | E3 HN-HD | E4 MN-LD | E5 MN-MD | E6 MN-HD | E7 LN-LD | E8 LN-MD | E9 LN-HD |
|---|--|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|
| Grain yield/plant (GYPP) g | | | | | | | | | |
| Parents | 163.8 | 124.3 | 110.2 | 144.6 | 111.0 | 95.6 | 87.8 | 63.9 | 47.7 |
| Change | - | -24.1 | -32.7 | -11.7 | -32.2 | -41.6 | -46.4 | -61.0 | -70.9 |
| Crosses | 224.5 | 175.4 | 158.1 | 199.4 | 150.1 | 135.6 | 119.5 | 90.0 | 72.8 |
| Change | - | -21.9 | -29.6 | -11.2 | -33.2 | -39.6 | -46.8 | -59.9 | -67.6 |
| LSD 0.05 | N = 4.46, D = 2.14, G = 1.96, N×D = 3.71 | | | | | | | | |
| Grain yield/feddan (GYPF) ard/fed | | | | | | | | | |
| Parents | 21.7 | 22.6 | 24.5 | 18.8 | 20.4 | 21.6 | 12.0 | 12.6 | 9.6 |
| Change | - | 4.5 | 12.9 | -13.2 | -5.9 | -0.5 | -44.5 | -41.8 | -55.5 |
| Crosses | 30.0 | 34.6 | 42.3 | 26.6 | 29.7 | 35.4 | 16.2 | 18.7 | 15.1 |
| Change | - | 15.5 | 41.1 | -11.2 | -1.0 | 18.1 | -45.9 | -37.7 | -49.6 |
| LSD 0.05 | N = 0.78, D = 0.35, G = 0.34, N×D = 0.61 | | | | | | | | |
| Total above ground dry matter/plant (TDM) g | | | | | | | | | |
| Parents | 322.3 | 277.5 | 256.7 | 292.5 | 255.9 | 236.1 | 211.3 | 179.5 | 151.4 |
| Change | - | -13.9 | -20.4 | -9.2 | -20.6 | -26.7 | -34.4 | -44.3 | -53.0 |
| Crosses | 391.9 | 338.3 | 312.7 | 360.9 | 307.7 | 283.9 | 256.6 | 217.7 | 189.5 |
| Change | - | -13.7 | -20.2 | -7.9 | -21.5 | -27.6 | -34.5 | -44.4 | -51.6 |
| LSD 0.05 | N = 4.07, D = 1.78, G = 1.82, N×D = 3.09 | | | | | | | | |
| Harvest index (HI) % | | | | | | | | | |
| Parents | 42.5 | 37.4 | 35.9 | 41.3 | 36.1 | 33.7 | 35.0 | 29.9 | 26.5 |
| Change | - | -12.0 | -15.5 | -2.9 | -15.1 | -20.6 | -17.6 | -29.7 | -37.6 |
| Crosses | 48.0 | 43.4 | 42.2 | 46.3 | 40.9 | 39.8 | 39.0 | 34.7 | 32.2 |
| Change | - | -9.7 | -12.2 | -3.6 | -14.9 | -17.1 | -18.7 | -27.7 | -33.0 |
| LSD 0.05 | N = 0.74, D = 0.37, G = 0.44, N×D = 0.64 | | | | | | | | |
| Economic nitrogen use efficiency (NUE_e) g/g | | | | | | | | | |
| Parents | 8.9 | 10.1 | 12.0 | 12.8 | 14.7 | 16.8 | 20.8 | 22.6 | 22.5 |
| Change | - | 13.9 | 34.6 | 43.7 | 65.4 | 89.7 | 134.1 | 155.0 | 153.5 |
| Crosses | 12.2 | 14.3 | 17.2 | 17.6 | 19.9 | 23.9 | 28.2 | 31.9 | 34.4 |
| Change | - | 17.2 | 40.8 | 44.5 | 63.1 | 96.3 | 131.9 | 162.2 | 182.8 |
| LSD 0.05 | N = 0.58, D = 0.38, G = 0.44, N×D = 0.66 | | | | | | | | |

H = high, M = medium, L = low, N = nitrogen, D = density and Change = 100*(RE – E1)/E1, RE = Respective environment

Maximum increases appeared under E9 followed by E8 environment and by BS trait (Table 3). Increases in NUE_e are favorable, while those in BS and ASI are unfavorable. It is worthy to note that plant height of both parents and crosses showed a tendency to increase under E5 and E6 environments, but showed a tendency of reduction under E8 and E9. The reason for PH increase under E5 and E6 may be attributed to elevated levels of plant density, while the reduction under E8 and E9 may be due to the severe stress of nitrogen.

Rank correlation coefficients estimated for pairs of the studied (nine) environments for GYPF are presented in Table 4. In general, the magnitude and number of significant correlation coefficients for GYPF were much higher in inbreds than those in hybrids, indicating that the interaction of inbreds with different environments (combinations of 3 N-rate × 3 plant densities) was much less than that of F1 crosses. The crosses have therefore higher ability to exhibit

the differences between environments than the inbreds, since heterozygotes are more responsive to improved environments than homozygotes, expressed in grain yield/feddan. This conclusion was previously confirmed by Rodrigues et al. [26] and Monneveux et al. [27].

In both inbreds and hybrids, the environment E7 LN and LD and environment E9 LN × HD showed no correlation with any other environment for GYPF. The environment E8 was correlated with E9 for GYPF (0.94**); these two environments are the most stressed. The maximum number of significant correlations (4) in F₁ crosses was found between E4 and each of E1, E2, E5 and E6 (Table 4).

3.3 Genotype × Nitrogen × Plant Density Interaction

Mean grain yield/fed across years under nine combinations of N-rates and plant densities for each inbred, hybrid and check is presented in Table 5. The rank of inbred parents for GYPF

was approximately similar in all nine environments, indicating less effect of interaction between inbred, nitrogen rate and plant density on GYPF. The percent reduction in GYPF due to both stresses, relative to E3 (HN-HD) which gave the highest GYPF was smaller in the low-performing lines (L29, L54 and L55) than in high-performing ones (L17, L18 and L53), which could be attributed to the lower yield potential of the first group of lines than the second one, under good environmental conditions. The first group of lines was considered tolerant to both stresses expressed in GYPF, while the second one was considered sensitive.

The best GYPF was obtained from E3 (HN-HD) for the first group of inbreds (L17, L18 and L53) and E1 (HN-LD) followed by E2 (HN-MD) for the second group (L29, L54 and L55). Regarding GYPF of the F₁ crosses, the rank varied from one environment (a combination of N rate with plant density) to another, especially when comparing environments that combine two stresses with those have only one stress or no stress, indicating the presence of cross × nitrogen × density interaction and that the GYPF of a cross differs from one combination (between N rate and plant density) to another. The best GYPF in this experiment was obtained under E3 (high N high D) and the best crosses in this environment were L17×L54 (60.4 ard), L17×L18 (58.7 ard), L53×L54 (55.1 ard), L53×L55 (53.7 ard) and L29×L55 (52.0 ard), with a significant superiority over SC 10 (the best check under this environment) by 26.9%, 23.3%, 15.8%, 12.8% and 9.2%, respectively.

The optimum combination of N-rates and plant densities (that gives the highest grain yield/unit area; GYPF) should be identified for each genotype (Table 5). It differs among inbreds, hybrids and checks.

The optimum environment in this study was E3 (HN-HD) followed by E2 (HN-MD) for three inbreds L17, L18 and L53, the crosses L18×L53, L18×L29, L18×L55, L29×L55, L54×L55 and the check cultivar SC 10. For remaining inbreds (L29, L54 and L55), the optimum combination of N-rates and plant density was E1 (HN-LD) followed by E2. Moreover, for the crosses L17×L18, L17×L53, L17×L29, L17×L54, L17×L55, L53×L29, L53×L54, L53×L55, L29×L54 and the check cultivar SC 2066, the optimum combination was E3 (HN-HD) followed by E6 (MN-HD) and for the cross L18×L54 the optimum combination was E6 (MN-HD) followed by E3 (HN-HD). Some hybrids in this experiment showed significant superiority over the best check in the respective environment (one cross under E9, 5 crosses under E6 and two crosses under E5).

These superiorities reached 36.65% over SC 2066 under E6 for the cross L17×L54 (the best cross in this experiment). It is worthy to note that five crosses (L17×L54, L17×L18, L53×L54, L53×L 55 and L29×L55) were considered the highest responsive ones, while other crosses (L18×L53, L18×L55, L18×L29, L53×L29 and L29×L54) were considered the most tolerant ones to both stresses (LN combined with HD).

Table 4. Rank correlation coefficient between pairs of nine environments for GYPF of parental inbreds (above diagonal) and F₁ crosses (below diagonal) across two seasons

| Environ- ment | E1 | E2 | E3 | E4 | E5 | E6 | E7 | E8 | E9 |
|------------------|-------|--------|--------|--------|-------|--------|-------|--------|--------|
| | HN-LD | HN-MD | HN-HD | MN-LD | MN-MD | MN-HD | LN-LD | LN-MD | LN-HD |
| E1 | | 0.94** | 0.83* | 0.77* | 0.77* | 0.71* | -0.14 | 0.94** | 1.00** |
| E2 | 0.39 | | 0.94** | 0.83* | 0.71* | 0.77* | 0.03 | 1.00** | 0.94** |
| E3 | -0.11 | 0.44* | | 0.94** | 0.60* | 0.71* | 0.14 | 0.94** | 0.83* |
| E4 | 0.56* | 0.55* | -0.08 | | 0.54* | 0.60* | 0.09 | 0.83* | 0.77* |
| E5 | 0.27 | 0.36 | -0.14 | 0.55* | | 0.94** | 0.09 | 0.71* | 0.77* |
| E6 | -0.06 | 0.06 | .45* | 0.39* | 0.23 | | 0.26 | 0.77* | 0.71* |
| E7 | -0.01 | -0.28 | -0.10 | 0.10 | -0.26 | 0.23 | | 0.03 | -0.14 |
| E8 | 0.22 | 0.19 | -0.47* | 0.38 | 0.49* | -0.20 | -0.09 | | 0.94** |
| E9 | -0.17 | 0.01 | -0.01 | 0.33 | -0.09 | 0.16 | -0.25 | 0.04 | |

H = high, M = medium, L = low, N = nitrogen, D = density and * and ** significant at 0.05 and 0.01 probability levels, respectively

Table 5. Mean grain yield/feddan (GYPF) and relative change (%) to E1 under nine environmental conditions

| Genotypes | E1 | E2 | E3 | E4 | E5 | E6 | E7 | E8 | E9 |
|----------------|------------------------|---------|-------|---------|---------|---------|---------|---------|---------|
| | HN-LD | HN-MD | HN-HD | MN-LD | MN-MD | MN-HD | LN-LD | LN-MD | LN-HD |
| Parents | | | | | | | | | |
| L17 | 26.9 | 27.7 | 32.8 | 24.4 | 27.4 | 29.6 | 11.6 | 12.1 | 10.4 |
| Change | -18.0** | -15.5** | - | -25.6** | -16.5** | -9.8** | -64.6** | -63.1** | -68.3** |
| L18 | 25.5 | 29.5 | 34.7 | 22.6 | 24.9 | 30.5 | 12.5 | 13.5 | 10.2 |
| Change | -26.5** | -15.0** | - | -34.9** | -28.2** | -12.1** | -64.0** | -61.1** | -70.6** |
| L53 | 27.0 | 32.0 | 36.2 | 23.8 | 29.4 | 31.9 | 12.9 | 17.1 | 10.6 |
| Change | -25.4** | -11.6** | - | -34.3** | -18.8** | -11.9** | -64.4** | -52.8** | -70.7** |
| L29 | 16.3 | 15.0 | 13.8 | 13.9 | 13.1 | 11.5 | 10.8 | 10.7 | 8.8 |
| Change | 18.1** | 8.7* | - | 0.7 | -5.1 | -16.7** | -21.7** | -22.5** | -36.2** |
| L54 | 18.7 | 17.3 | 16.0 | 14.4 | 14.1 | 13.0 | 12.7 | 11.8 | 9.5 |
| Change | 16.9** | 8.1* | - | -10.0* | -11.9* | -18.8** | -20.6** | -26.3** | -40.6** |
| L55 | 15.7 | 14.4 | 13.3 | 13.6 | 13.3 | 12.8 | 11.6 | 10.5 | 8.4 |
| Change | 18.0** | 8.3* | - | 2.3 | 0.0 | -3.8 | -12.8* | -21.1** | -36.8** |
| Crosses | | | | | | | | | |
| 1. L17×L18 | 34.7 | 40.5 | 58.7 | 30.7 | 35.3 | 45.7 | 16.7 | 20.8 | 11.8 |
| Change | -40.9** | -31.0** | - | -47.7** | -39.9** | -22.1** | -71.6** | -64.6** | -79.9** |
| 2. L17×L53 | 26.6 | 30.3 | 35.5 | 23.8 | 26.1 | 31.0 | 17.8 | 20.8 | 16.8 |
| Change | -25.1** | -14.6** | - | -33.0** | -26.5** | -12.7** | -49.9** | -41.4** | -52.7** |
| 3. L17×L29 | 24.0 | 28.0 | 32.0 | 22.5 | 25.3 | 29.4 | 15.7 | 21.0 | 10.9 |
| Change | -25.0** | -12.5** | - | -29.7** | -20.9** | -8.1** | -50.9** | -34.4** | -65.9** |
| 4. L17×L54 | 41.2 | 48.7 | 60.4 | 37.8 | 40.0 | 52.9 | 16.1 | 17.5 | 14.6 |
| Change | -31.8** | -19.4** | - | -37.4** | -33.8** | -12.4** | -73.3** | -71.0** | -75.8** |
| 5. L17×L55 | 24.7 | 27.5 | 30.9 | 23.9 | 25.2 | 28.4 | 14.0 | 17.4 | 14.2 |
| Change | -20.1** | -11.0** | - | -22.7** | -18.4** | -8.1** | -54.7** | -43.7** | -54.0** |
| 6. L18×L53 | 32.3 | 36.5 | 42.2 | 24.6 | 25.3 | 29.1 | 19.3 | 23.0 | 21.3 |
| Change | -23.5** | -13.5** | - | -41.7** | -40.0** | -31.0** | -54.3** | -45.5** | -49.5** |
| 7. L18×L29 | 24.5 | 31.5 | 35.9 | 23.2 | 26.1 | 29.6 | 16.4 | 20.0 | 18.3 |
| Change | -31.8** | -12.3** | - | -35.4** | -27.3** | -17.5** | -54.3** | -44.3** | -49.0** |
| 8. L18×L54 | 29.2 | 26.5 | 31.2 | 24.9 | 23.3 | 32.0 | 18.9 | 18.7 | 15.2 |
| Change | -6.4* | -15.1** | - | -20.2** | -25.3** | 2.6 | -39.4** | -40.1** | -51.3** |
| 9. L18×L55 | 32.6 | 37.4 | 45.9 | 24.4 | 28.6 | 34.5 | 16.1 | 18.9 | 19.5 |
| Change | -29.0** | -18.5** | - | -46.8** | -37.7** | -24.8** | -64.9** | -58.8** | -57.5** |
| 10. L53×L29 | 26.6 | 32.9 | 37.7 | 26.1 | 30.2 | 34.9 | 19.7 | 19.1 | 17.6 |
| Change | -29.4** | -12.7** | - | -30.8** | -19.9** | -7.4** | -47.7** | -49.3** | -53.3** |
| 11. L53×L54 | 36.1 | 41.9 | 55.1 | 31.8 | 38.0 | 44.3 | 11.5 | 9.9 | 9.6 |
| Change | -34.5** | -24.0** | - | -42.3** | -31.0** | -19.6** | -79.1** | -82.0** | -82.6** |
| 12. L53×L55 | 31.7 | 38.5 | 53.7 | 28.6 | 34.3 | 43.3 | 9.9 | 16.5 | 10.9 |
| Change | -41.0** | -28.3** | - | -46.7** | -36.1** | -19.4** | -81.6** | -69.3** | -79.7** |
| 13. L29×L54 | 24.0 | 26.1 | 30.8 | 22.2 | 24.4 | 26.1 | 18.7 | 19.0 | 17.5 |
| Change | -22.1** | -15.3** | - | -27.9** | -20.8** | -15.3** | -39.3** | -38.3** | -43.2** |
| 14. L29×L55 | 37.1 | 45.0 | 52.0 | 34.8 | 38.9 | 44.6 | 16.9 | 16.5 | 14.3 |
| Change | -28.7** | -13.5** | - | -33.1** | -25.2** | -14.2** | -67.5** | -68.3** | -72.5** |
| 15. L54×L55 | 24.6 | 28.0 | 32.9 | 20.2 | 24.6 | 25.3 | 15.4 | 21.3 | 14.4 |
| Change | -25.2** | -14.9** | - | -38.6** | -25.2** | -23.1** | -53.2** | -35.3** | -56.2** |
| Checks | | | | | | | | | |
| SC 10 | 35.5 | 40.8 | 47.6 | 27.5 | 32.9 | 38.7 | 21.1 | 23.2 | 11.5 |
| Change | -25.4** | -14.3** | - | -42.2** | -30.9** | -18.7** | -55.7** | -51.3** | -75.8** |
| SC 2066 | 35.4 | 37.3 | 40.3 | 36.0 | 37.2 | 38.8 | 17.5 | 21.5 | 20.7 |
| Change | -12.2** | -7.4** | - | -10.7** | -7.7** | -3.7* | -56.6** | -46.7** | -48.6** |
| LSD 0.05 | G = 0.34, G×D×N = 1.03 | | | | | | | | |

H = high, M = medium, L = low, N = nitrogen, D = density, * and ** significant at 0.05 and 0.01 probability levels, respectively and Change = 100*(RE - E1)/E1, RE = respective environment

3.4 Superiority of Tolerant (T) Over Sensitive (S) Genotypes

The higher absolute GYPF and lower ratio of reduction in GYPF under LN combined with HD to yield under HN combined with LD were considered as an index of tolerance to the two stresses together. Based on this index, the tolerant inbreds were L17, L18 and L53, while the sensitive inbreds were L29, L54 and L55. The F₁ crosses L18×L53, L18×L55 and L18×L29 were, therefore, considered tolerant and L53×L54, L17×L29 and L17×L18 were considered sensitive crosses. Data averaged for each of the two groups (T and S) for inbreds and hybrids differing in tolerance to both stresses together indicate that GYPF of tolerant (T) was greater than that of the sensitive (S) inbreds and crosses by 17.1 and 36.3%, respectively under LN combined with HD (no N addition %) conditions (Table 6).

Superiority of LN-HD tolerant (T) over sensitive (S) inbreds in GYPF under LN-HD was associated with superiority in most studied traits, namely GYPP (10.5%), EPP (14.3%), GPP (39.9%), 100-GW (9.0%), HI (2.7%), NUE_e (10.0%), BS (-11.2%), PH (-9.3%) and ASI (-5.8%). Superiority of T over S crosses in GYPF under LN was due to their superiority in GYPP (28.3%), EPP (23.4%), GPP (8.0%), 100-GW (14.1%), HI (11.6%), NUE_e (32.1%), BS (-62.8%) and ASI (-21.7%). The superiority of T over S under LN for crosses was greater than that for inbreds. This might be attributed to the high nitrogen use efficiency traits of the hybrids due to heterosis as compared to their inbred parents. These results are in agreement with those reported by Lafitte and Edmeades [28], Shieh et al. [29], Kling et al. [30] and Gama et al. [31].

The superiority of modern maize hybrids tolerant to high plant density was also attributed to decreased barrenness [32], more leaf erectness [6], synchronization of 50% anthesis with 50% silking [33] and increased prolificacy, *i.e.*, more ears/plant [34]. A shortened ASI was considered as an indication of higher flow of assimilates to the developing ears during the early reproductive stage under conditions of high density stress [35, 36]. High plant density-tolerant genotypes possess shorter ASI than intolerant ones [8,37,38]. Al-Naggar et al. [25] also reported that under high plant density, the tolerant testcrosses showed 314.4% more GYPP, 115.0% more GPP, 48.4% heavier 100-GW, 42.9 more EPP,

98.2% less BS and 63.3% shorter ASI than sensitive testcrosses.

CIMMYT breeders found that maize grain yield under LN was closely related to some secondary traits such as improved N-uptake, high plant nitrate content, large leaf area, high specific leaf-N content, ears/plant, ASI and leaf senescence [12,36,39]. These results are in consistency with those reported by Al-Naggar et al. [24]. Reduction in barren stalks and shortening in ASI of tolerant as compared to sensitive inbreds and hybrids in the present study are desirable and may be considered as important contributors to LN as well as to HD tolerance. Similar conclusions were reported by Buren et al. [37], Dow et al. [35], Beck et al. [38], Vasal et al. [8], Edmeades et al. [36] and Al-Naggar et al. [25] and Al-Naggar et al. [40].

3.5 Differential Response of T×T, T×S and S×S Crosses

Mean performance of traits were averaged across three groups of F₁ crosses, *i.e.*, T×T, T×S and S×S groups based on grain yield per feddan of their parental lines under stress and non-stress conditions, *i.e.*, both high-D and low-N stresses together and presented in Table (7). Number of crosses was 3, 9 and 3 for the T×T, T×S and S×S groups, respectively. In general, T×T crosses had favorable (higher) values for grain yield and its attributes and lower (favorable values for DTS, ASI, BS and LANG) than S×S and T×S crosses under each stress and both stresses. In general, low-N and high density T×T crosses were the most superior for all studied traits (Table 7), under the most severe environment (E9) where both severe stresses (low-N and density of 40,000 plants/fed) existed. The T×S crosses for both stresses came in the second rank for superiority in ASI, PH, PL-M80, EPP, GPP and 100-GW and the S×S crosses for both stresses were in the second rank for superiority in the remaining traits (BS, LANG, 100-GW, GYPP, GYPF, TDM, HI and NUE_e).

Under low-N and high-D stresses together (E9), grain yield/fed of low-N and high-D T×T crosses (16.6 ard) was greater than that of S×S (15.4 ard) and T×S (14.5 ard) by 7.79 and 14.48%, respectively. This indicates that to obtain a tolerant cross to both stresses in the same time, its two parental inbred lines should be tolerant to the same stresses. Superiority of low-N and high-D T×T over S×S and T×S crosses in GYPF

Table 6. Superiority (%) in some selected characters of the most 3 tolerant (T) over the most 3 sensitive (S) inbreds and crosses to LN-HD under LN-HD combined across two seasons

| Trait | Inbreds | | | Crosses | | |
|------------------------|---------|-------|---------------|---------|-------|---------------|
| | T | S | % Superiority | T | S | % Superiority |
| GYPF (ard) | 12.3 | 10.5 | 17.1 | 19.4 | 14.2 | 36.3 |
| GYPP (g) | 69.8 | 63.2 | 10.5 | 107.4 | 83.7 | 28.3 |
| EPP | 0.80 | 0.70 | 14.3 | 0.95 | 0.77 | 23.4 |
| GPP | 287.9 | 205.8 | 39.9 | 299.8 | 277.7 | 8.0 |
| 100-GW (g) | 25.8 | 23.7 | 9.0 | 29.7 | 26.0 | 14.1 |
| TDM (g) | 187.2 | 174.3 | 7.4 | 242.5 | 207.0 | 17.1 |
| HI (%) | 30.9 | 30.1 | 2.7 | 37.1 | 33.2 | 11.6 |
| NUE _e (g/g) | 23.0 | 20.9 | 10.0 | 36.4 | 27.6 | 32.1 |
| BS (%) | 35.8 | 40.3 | -11.2 | 10.8 | 28.9 | -62.8 |
| PH (cm) | 173.2 | 190.9 | -9.3 | 206.9 | 202.7 | 2.1 |
| ASI (day) | 6.6 | 7.1 | -5.8 | 4.4 | 5.6 | -21.7 |

$$\% \text{ Superiority} = 100 \times [(T - S)/S]$$

Table 7. Trait differences averaged across the T×T, T×S and S×S groups of F₁ crosses for both stresses under the low nitrogen-high plant density environment (E9) across two seasons

| Trait | T × T | T × S | S × S | Trait | T × T | T × S | S × S |
|------------|-------|-------|-------|------------------------|-------|-------|-------|
| ASI (days) | 5.2 | 6.7 | 7.6 | GPP | 245.6 | 190.2 | 75.5 |
| PH (cm) | 211.1 | 219.4 | 21.9 | 100-GW (g) | 26.6 | 25.0 | 25.0 |
| BS (%) | 13.1 | 25.8 | 19.8 | GYPP (g) | 81.8 | 70.1 | 72.2 |
| LANG (°) | 27.6 | 28.9 | 27.7 | GYPF (ard) | 16.6 | 14.5 | 15.4 |
| CCI (%) | 11.2 | 16.6 | 13.8 | TDM (g) | 203.3 | 184.8 | 190.0 |
| PL-M80 (%) | 9.8 | 8.9 | 8.6 | HI (%) | 33.7 | 31.7 | 32.0 |
| EPP | 0.8 | 0.7 | 0.7 | NUE _e (g/g) | 38.7 | 33.1 | 34.1 |

T = tolerant and S = sensitive

under low-N and high-D stresses was due to their superiority in GYPP by 9.5 and 11.7 g, GPP by 70.4 and 55.4, 100-GW by 1.6 and 1.6 g, EPP by 0.1 and 0.1, TDM by 13.3 and 18.5 g/plant, HI by 1.7 and 2.0%, NUE_e by 4.6 and 5.6 g/g and PL-M80 by 1.2 and 0.9%, respectively. Moreover, low-N and high-D T×T crosses were earlier in DTS by 4.7 and 1.9 days, of shorter ASI by 2.1 and 1.5 days, shorter PH by 10.8 and 8.3 cm, lower BS by 6.7 and 12.7% and narrow LANG by 0.1 and 1.3° than S×S and T×S crosses, respectively under the most severe stresses in this experiment existed in E9 environment. In general, crosses classified as low-N and high-density tolerant × low-N and high-density tolerant crosses in terms of grain yield under low-N and high-D stresses had a better nitrogen use efficiency traits and high density adaptive traits such as lower values of DTS, ASI, PH, BS and LANG as compared with low-N and high density sensitive × low-N and high density sensitive crosses.

3.6 Grouping Genotypes Based on Tolerance and Responsiveness

Mean grain yield/plant or per feddan across years of studied crosses under LN-HD together was plotted against same trait of the same genotypes under HN and LD together (Figs. 1 and 2) where numbers from 1 to 15 refer to F₁ hybrids names 1 = L17×L18, 2 = L17×L53, 3 = L17×L29, 4 = L17×L54, 5 = L17×L55, 6 = L18×L53, 7 = L18×L29, 8 = L18×L54, 9 = L18×L55, 10 = L53×L29, 11 = L53×L54, 12 = L53×L55, 13 = L29×L54, 14 = L29×L55 and 15 = L54×L55, which made it possible to distinguish between efficient and inefficient genotypes on the basis of above-average and below-average grain yield under LN and HD together and responsive and non-responsive genotypes on the basis of above-average and below-average grain yield under HN and LD together [41]. According to tolerance to both stresses, *i.e.*, LN and HD together and responsiveness to HN and LD conditions, the 15 studied crosses were classified into four groups, *i.e.*, efficient (tolerant) and responsive, efficient (tolerant) and non-

responsive, inefficient (sensitive) and responsive and inefficient (sensitive) and non-responsive. Based on grain yield/plant (Fig. 1) or grain yield/feddan (Fig. 2), the two crosses No. 6 (L18×L53) and No. 9 (L18×L55) had the highest GYPP or GYPF under HN-LD (E1) and LN-HD (E9), *i.e.*, they could be considered tolerant (efficient) to both stresses and responsive to the non-stressed environment.

The five crosses No. 4 (L17×L54), No. 14 (L29×L55), No. 1 (L17×L18), No. 12 (L53×L55) and No. 11 (L53×L54) were considered inefficient (sensitive) but responsive based on GYPP and GYPF. The group of efficient (tolerant) to both stresses but not responsive included the crosses No. 7 (L18×L29), No. 2 (L17×L53), No. 10 (L53×L29) and No. 13 (L29×L54) based on both GYPP and GYPF but included one more cross No. 8 (L18×L54) based on GYPF only.

On the contrary, the group of inefficient (sensitive to both stresses) and non-responsive to HN and LD included the crosses No. 3 (L17×L29), No. 5 (L17×L55) and No. 15 (L54×L55) based on both GYPP (Fig. 1) and GYPF, but included one more cross, *i.e.*, No. 8 (L18×L54) based on GYPF only (Fig. 2).

3.7 Identifying Optimum Density and/or Appropriate N Application

Data were reanalyzed to evaluate GYPF responses of inbreds and hybrids across varying levels of stress. For each genotype or group of genotypes, quadratic regression function was performed for N-rate × plant density interaction. The regression functions were used to distinguish which treatments provide optimum value for each genotype (or group of genotypes). The relationships between the nine environments (combinations of 3 N-rates and 3 plant densities) and grain yield/fed across seasons are illustrated in Fig. 3 for inbreds and Fig. 4 for F₁ crosses. The 9 environments were arranged in Figs. 3 and 4 based on the severity of both N and plant density stresses together, where the poorest environment (E9) represents maximum stress (LN and HD), while the best environment (E1) represents the no-stressed one (highest N and lowest plant density). The three inbred parents (L17, L18 and L53) showed a quadratic regression function, with an optimum combination of HN and LD, with available N/plant of 7.79 g. While, the inbreds L54, L29 and L55

showed a weak quadratic regression very close to linear response (Fig. 3), with an optimum environment of combination between HN (240 kg N/fed and LD (20,000 plants/fed), *i.e.*, with available N/plant of 15.57 g.

The grain yield/fed across years of all groups of F₁ crosses showed a quadratic regression function under the nine combinations of plant densities and N-rates (Fig. 4), except E-R group the two crosses, which showed near linear regression. The optimum N-rate and plant density combination was 40,000 plants/fed with a fertilization rate of 240 kg N/fed across the four groups of F₁ crosses. The most responsive group of hybrids to the improvement of environmental conditions was E-R group, while the lowest responsive group was I-NR. In this context, Shapiro and Wortmann [42] reported that the corn grain yield typically exhibits a quadratic response to plant density with a near-linear increase across a range of low densities, a gradually decreasing rate of yield increase relative to density increase and finally a yield plateau at some relatively high plant density. Clark [16] mentioned that there was little yield response to N-rates above 90 kg N/ha at the low and high densities, as there was a curvilinear increase until yield plateau at the low density (8.1 Mg/ha at 133 kg N/ha) and the high density (5.9 Mg/ha at 102 kg N/ha). He added that response to N was greatest at the middle density (83,980 plants/ha), as there was a quadratic response with maximum yield at 188 kg N/ha (8.7 Mg/ha). He found that across the low-stress environments, the lowest density (44,460 plants/ha) responded little to N-rates above 90 kg N/ha, while there was greater response to N-rates at the middle density (13.5 Mg/ha at 162 kg N/ha) and the high density (13.4 Mg/ha at 174 kg N/ha). He concluded that no support was found for the idea that increasing corn yield requires increases in both plant density and N-rate above rates typically used. A recent Indiana study [19] showed that under large ranges of plant density (54,000-104,000 plants/ha) and N-rate (0-330 kg N/ha), higher densities required more N. This seems logical, given the prevailing belief that high yields require more plants, and that more plants require more N. Their and our results advance our understanding of N rate-plant density interaction within contrasting environmental condition, but understanding the complexities of hybrid interactions with N-rate and plant density will require additional work.

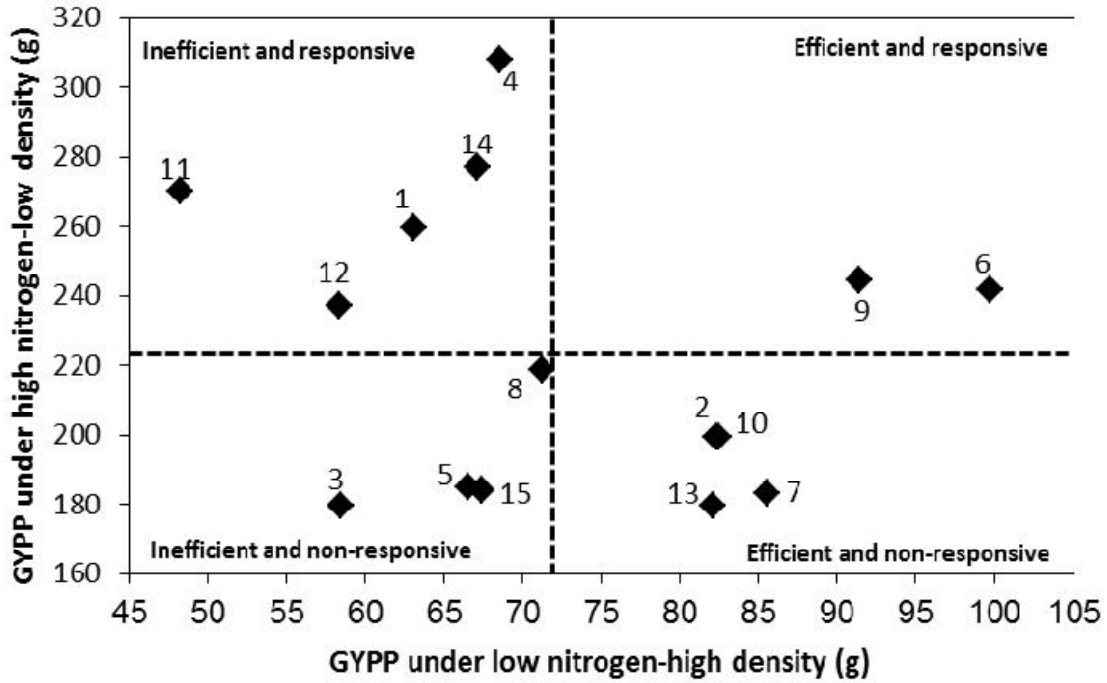


Fig. 1. Relationships between grain yield/plant (GYPP) of 15 F₁ maize hybrids under high nitrogen-low density and low nitrogen-high density combined across two seasons. broken lines represent mean of GYPP (numbers from 1 to 15 refer to F₁ hybrids in Table 5)

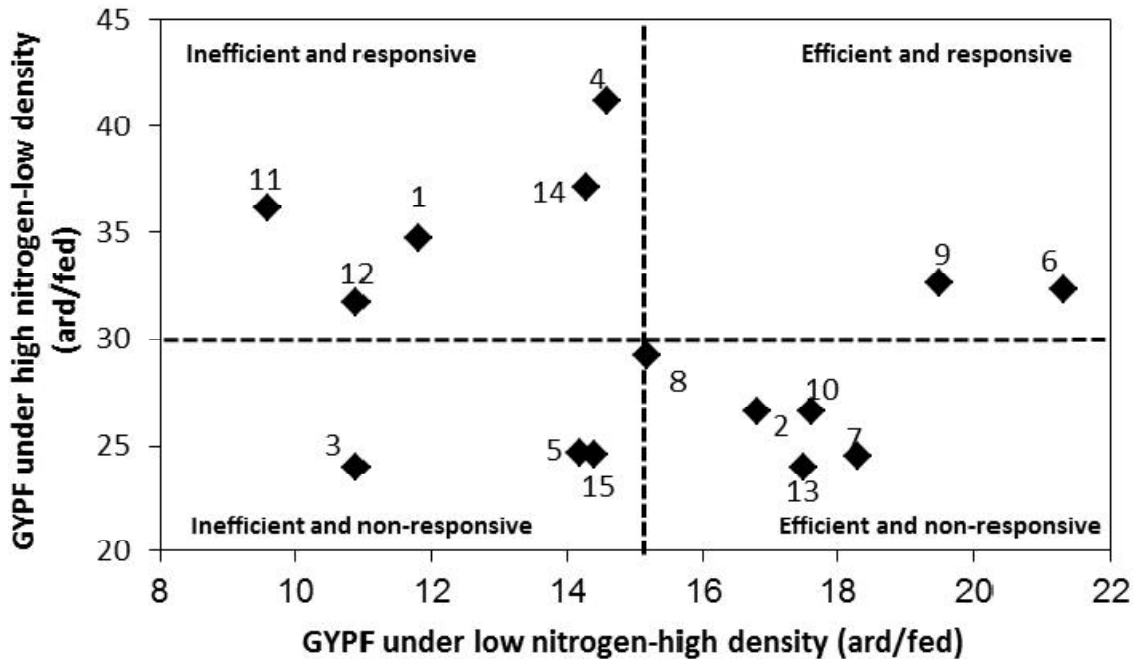


Fig. 2. Relationships between grain yield/feddan (GYPF) of 15 F₁ maize hybrids under high nitrogen-low density and low nitrogen-high density combined across two seasons. broken lines represent mean of GYPF (numbers from 1 to 15 refer to F₁ hybrids in Table 5)

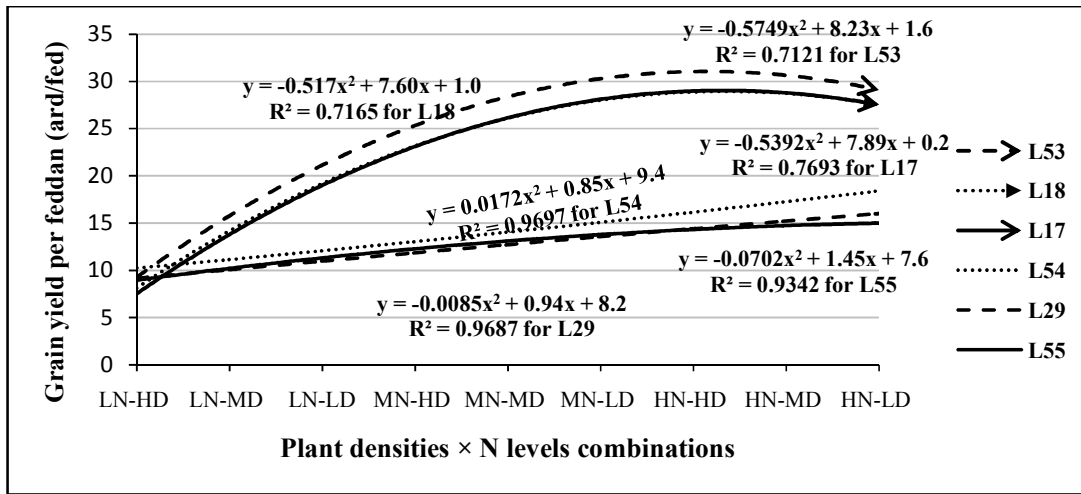


Fig. 3. Relationship between GYPF of inbreds and nine environment combinations between three plant densities and three N levels across two seasons

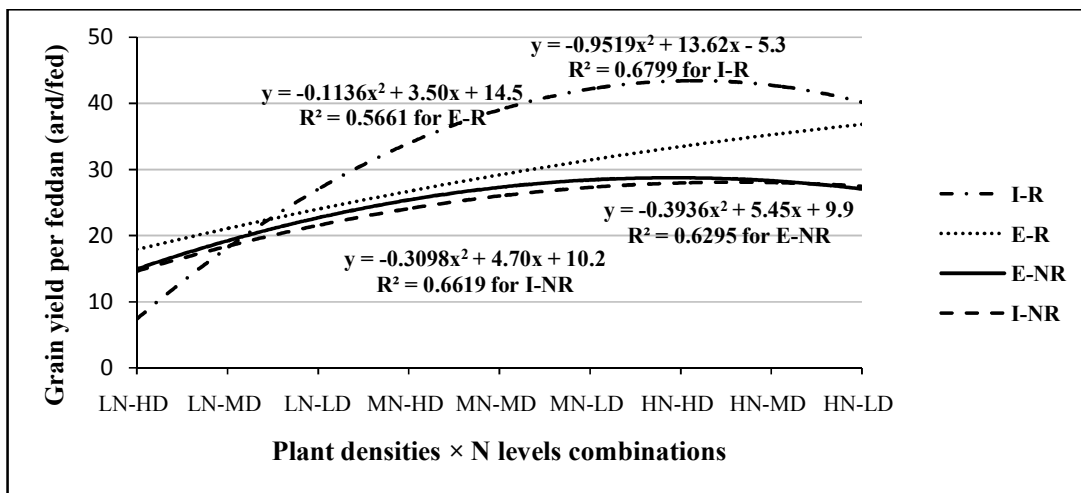


Fig. 4. Relationship between GYPF of four groups of F₁ crosses, namely, five inefficient and responsive (I-R), two efficient and responsive (E-R), four efficient and non-responsive (E-NR) and four inefficient and non-responsive (I-NR) crosses and nine combinations between three plant densities and three N-rates across two seasons

4. CONCLUSION

Some newly-developed maize genotypes in the present investigation could double maize productivity, reaching 60.6 ard/fed in the cross L17×L54 from the same unit land area, if they are grown in double plant population density used in Egypt, *i.e.* 40,000 plants/fed, but in condition they are given the double recommended N-rate (240 kg/fed). Fortunately, the same cross gave also a high grain yield/fed (52.9 ard) under medium N-rate (120 kg/fed) and high plant density (40,000 plants/fed). The optimum combination in the present study was

high-N×high-density for 3 out of 6 inbreds and 14 out of 15 F₁ crosses, while it was high-N×low-density for the remaining 3 inbreds and medium-N×high-density for the remaining cross (L18×L54). Investigations on the optimum combination between plant population density and N-rate for each new maize hybrid should be carried out for giving the highest grain yield per unit land area.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Hashemi AM, Herbert SJ, Putnam DH. Yield response of corn to crowding stress. *Agron. J.* 2005;97:839-846.
2. Huseyin G, Omer K, Mehmet K. Effect of hybrid and plant density on grain yield and yield components of maize (*Zea mays* L.). *Indian J. Agron.* 2003;48(3):203-205.
3. Duvick DN, Cassman KG. Post-green revolution trends in yield potential of temperate maize in the North-Central United States. *Crop Sci.* 1999;39:1622-1630.
4. Tollenaar M, Wu J. Yield improvement in temperate maize is attributable to greater stress tolerance. *Crop Sci.* 1999;39:1597-1604.
5. Duvick DN, Smith J, Cooper M. Long-term selection in a commercial hybrid maize breeding program. In Janick J (ed). *Plant Breeding Reviews*. John Wiley and Sons: New York USA; 2004.
6. Radenovic C, Konstantinov K, Delic N, Stankovic G. Photosynthetic and bioluminescence properties of maize inbred lines with upright leaves. *Maydica*, 2007;52(3):347-356.
7. Tetio-Kagho F, Gardner FP. Response of maize to plant population density: II. Reproductive developments, yield, and yield adjustment. *Agron. J.* 1988;80:935-940.
8. Vasal SK, Cordova H, Beck DL, Edmeades GO. Choices among breeding procedures and strategies for developing stress tolerant maize germplasm. *Proceedings of Symposium held on March 1996;25-29, El Batan, Mexico, D.F.: CIMMYT.* 1997;336-347.
9. Khaliq TA, Ahmad AH, Ali MA. Maize hybrid response to nitrogen rates at multiple locations in semiarid environment. *Pakistan Journal of Botany*, 2009;41:207-224.
10. Biswas TD, Mukherjee SK. *Text Book of Soil Science*. (5th ed.). Tata McGraw-Hill, New Delhi. 1993;170-197.
11. Brady NC, Weil RR. *The Nature and Properties of Soils*. (13th ed.). Pearson Education Ltd., USA; 2002.
12. Banziger M, Lafitte HR. Efficiency of secondary traits for improving maize for low-nitrogen target environments. *Crop Sci.* 1997;37:1110-1117.
13. Banziger M, Edmeades GO, Beck D, Bellon M. Breeding for drought and nitrogen stress tolerance in maize: From theory to practice (on line). Available: <http://www.cimmyt.mx>. Mexico, DF, CIMMYT. 2000;68.
14. Banziger M, Edmeades GO, Lafitte HR. Selection for drought tolerance increases maize yields across a range of nitrogen levels. *Crop Sci.* 1999;39:1035-1040.
15. Bhatt PS. Response of sweet corn hybrid to varying plant densities and nitrogen levels. *African Journal of Agricultural Research*. 2012;7(46):6158-6166.
16. Clark, RA. Hybrid and plant density effects on nitrogen response in corn. M. Sc. Thesis, Fac. Graduate, Illinois State Univ., USA; 2013.
17. Tajul, MI, Alam MM, Hossain SMM, Naher K, Rafii MY, Latif MA. Influence of plant population and nitrogen-fertilizer at various levels on growth and growth efficiency of maize. *The Scientific World Journal*. 2013;1:1-9.
18. O'Neill PM, Shanahan JF, Schepers JS, Caldwell B. Agronomic responses of corn hybrids from different eras to deficit and adequate levels of water and nitrogen. *Agron. J.* 2004;96(6):1660-1667.
19. Boomsma CR, Santini JB, Tollenaar M, Vyn TJ. Maize morphophysiological responses to intense crowding and low nitrogen availability: An analysis and review. *Agron. J.* 2009 101(6):1426-1448.
20. Moll RH, Kamprath EJ, Jackson WA. Analysis and interpretation of factors which contribute to efficiency of N utilization. *Agron. J.* 1982;74:562-564.
21. Snedecor GW, Cochran WG. *Statistical Methods*. 8th edition, Iowa State Univ. Press., Ames, Iowa, USA; 1989.
22. Steel RGD, Torrie GH, Dickey DA. *Principles and Procedures of Statistics: A Biometrical Approach*. 3rd ed. McGraw-Hill, New York, USA; 1997.
23. Kamara AY, Menkir A, Kureh I, Omoigui LO, Ekeleme F. Performance of old and new maize hybrids grown at high plant densities in the tropical Guinea savanna. *Communications in Biometry and Crop Sci.* 2006;1(1):41-48.
24. Al-Naggar AMM, Shabana R, Al-Khalil TH. Tolerance of 28 maize hybrids and populations to low-nitrogen. *Egypt. J. Plant Breed.* 2010;14(2):103-114.
25. Al-Naggar AMM, Shabana R, Rabie AM. Per se performance and combining ability of 55 new maize inbred lines developed for

- tolerance to high plant density. Egypt. J. Plant Breed. 2011;15(5):59-84.
26. Rodrigues LRF, Da Silva N, Mori ES. Baby corn single-cross hybrids yield in two plant densities. Crop Breeding and Applied Biotechnology. 2003;(3):177-184.
 27. Monneveux P, Zaidi PH, Sanchez C. Population density and low nitrogen affects yield-associated traits in tropical maize. Crop Sci. 2005;45:535-545.
 28. Lafitte HR, Edmeades GO. Association between traits in tropical maize inbred lines and their hybrids under high and low soil nitrogen. Maydica. 1995;40:259-267.
 29. Shieh G, Ho C, Lu H. The effect of nitrogen rate on the combining ability and heterosis in maize traits. Journal of Agricultural Research of China. 1995;4(1):15-25.
 30. Kling JG, Keh SOO, Akintoy HA, Heuberger HT, Horst WJ. Potential for developing nitrogen use efficient maize for low input agriculture system in the moist Savannas of Africa. Proceedings of a conference on Developing Drought and Low N Tolerant Maize, 25-29 March 1997, El-Battan, Mexico. 1997;490-501.
 31. Gama, EE, Marriel IE, Guimaraes PEO, Parentoni SN, Santos MX, Pacheco CAP, Meireles WF, Ribeiro PHE, Oliveira ACD. Combining ability for nitrogen use in a selected set of inbred lines from a tropical maize population. Revista Brasileira de Milho e Sorgo. 2002;1(3):68-77.
 32. William JC. Corn silage and grain yield responses to plant densities. J. of Production Agric. 1997;10(3):405-409.
 33. Edmeades GO, Bolanos J, Hernandez M, Bello, S. Causes for silk delay in a lowland tropical maize population. Crop Sci. 1993;33:1029-1035.
 34. Miller LC, Vasilas BL, Taylor RW, Evans TA, Gempesaw CM. Plant population and hybrid consideration for dryland corn production on drought-sensitive soils. Can. J. Plant Sci. 1995;75:87-91.
 35. Dow EW, Daynard TB, Muldoon JF, Major DJ, Thurtell GW. Resistance to drought and density stress in Canadian and European maize (*Zea mays* L.) hybrids. Can. J. Plant Sci. 1984;64:575-583.
 36. Edmeades GO, Bolanos J, Chapman SC, Lafitte HR, Banziger M. Selection improves drought tolerance in a tropical maize population: gains in biomass, grain yield and harvest index. Crop Sci. 1999;39:1306-1315.
 37. Buren LL, Mock JJ, Anedrson IC. Morphological and physiological traits in maize associated with tolerance to high plant density. Crop Sci. 1974;14:426-429.
 38. Beck DL, Betran J, Banziger M, Willcox M, Edmeades GO. From landrace to hybrid: Strategies for the use of source populations and lines in the development of drought tolerant cultivars. Proceedings of a Symposium, March 25-29, CIMMYT, El Batan, Mexico. 1997;369-382.
 39. Lafitte HR, Edmeades GO. Improvement for tolerance to low soil nitrogen in tropical maize. I. Selection criteria. Field Crops Res. 1994;39:1-14.
 40. Al-Naggar AMM, Shabana R, Rabie AM. Genetics of maize rapid silk extrusion and anthesis-silking synchrony under high plant density. Egypt. J. Plant Breed. 2012;16(2):173-194.
 41. Sattelmacher, B, Horst WJ, Becker HC. Factors that contribute to genetic variation for nutrient efficiency of crop plants. Z. fur Pflanzenernahrung und Bodenkunde 1994;157:215-224.
 42. Shapiro CA, Wortmann CS. Corn response to nitrogen rate, row spacing and plant density in Eastern Nebraska. Agron. J. 2006;98(3):529-535.

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