COMBINING ABILITY FOR NITROGEN USE IN A SELECTED SET OF INBRED LINES FROM A TROPICAL MAIZE POPULATION

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ABSTRACT - Tolerance rather than resistance, is the crop response mechanism sought by maize breeders for cultivation under low levels of nitrogen. Although improved nitrogen use is in intensive research by maize breeders, little is known about its inheritance on tropical maize germplasm. The objectives of this study were to evaluate the N use through yield and other traits, and to determine the importance of general (GCA) and specific (SCA) combining ability effects in the inheritance of N use, for yield, using a diallel of crosses among a selected set of tropical maize lines under two levels of N. A comparison of relative magnitudes of GCA and SCA for ear yield (EY) suggests that this trait was governed more by dominance than additive gene action, and the contribution of additive genes for nitrogen utilization efficiency decreases under N nutrient shortage. Since, for this set of lines, only non additive effects were significant for this trait, the lines with favorable SCA could be used as donors of N use efficiency in a hybrid development program. The line L2 was identified as being the most suitable combining overall parents in this study for improving EY. The crosses L 3 x L 7, L 1 x L 2 and L 7 x L 10 showed to be promising combinations based on the significant SCA effects for lines and hybrids development. The results of this study can enhance the use of promising inbred lines in a program focused on developing hybrid that efficiently can take up and uses nitrogen.

Key words: *Zea mays L.*, nitrogen stress, general and specific combining ability, yield.

CAPACIDADE DE COMBINAÇÃO PARA USO DE NITROGÊNIO DE UM GRUPO DE LINHAGENS SELECIONADAS DE UMA POPULAÇÃO DE MILHO TROPICAL.

RESUMO - Tolerância, em vez de resistência, é um mecanismo de resposta das plantas que tem sido buscado por melhoristas de milho, para cultivos com baixos níveis de nitrogênio. Mesmo com o crescimento das pesquisas por melhoristas, visando o uso mais eficiente do nitrogênio, pouco se conhece sobre sua implicação no comportamento de linhagens e a respeito de sua herdabilidade em germoplasma de milho tropical. Os objetivos deste estudo foram avaliar o uso do nitrogênio, com base na produção de espigas (PE) e de outros caracteres, bem como determinar a importância dos efeitos das capacidades geral (CGC) e específica (CEC) de combinação na herança do uso de nitrogênio, usando-se cruzamento dialelo de um grupo de linhagens selecionadas, em dois níveis de nitrogênio. Uma comparação da magnitude relativa das CGC e CEC para PE sugere que esta característica foi mais controlada por genes em dominância que devido à ação gênica aditiva e que a contribuição dos genes aditivos para o uso eficiente de nitrogênio diminuiu com o estresse de nitrogênio. Como, para esse grupo de linhagens, somente os efeitos não aditivos foram significativos nas condições de estresse de N, as linhagens deste estudo com CEC favoráveis poderiam ser usadas como doadoras da tolerância ao estresse de N, em programas de desenvolvimento de híbridos. A linhagem L2 foi identificada como sendo a mais promissora para combinação com as demais linhagens, para se aumentar a PE. Os cruzamentos L3 x L7, L1 x L2 e L7 x L10, baseados nos efeitos da CEC, mostram ser combinações promissoras para um programa de desenvolvimento de linhagens e híbridos. Os resultados deste estudo indicam o potencial a ser explorado com o uso de linhagens promissoras, em programa de melhoramento focado no desenvolvimento de híbridos eficientes na utilização do nitrogênio.

Palavras-chave: *Zea mays L*., estresse de nitrogênio, capacidade geral e específica de combinação, produção.

Brazil produced about 41 million tons of maize in the 2000/2001 harvests, making maize the second most cultivated cereal, with an area of 13.750 million ha (CONAB, 2001) and a mean consumption of 25 kg ha^{-1} nitrogen estimated from ANDA (2001)data.

Nitrogen deficiency is one of the most important stresses affecting maize production in tropical areas (Lafitte & Bänziger, 1995). The great majority of tropical soils present low levels of fertility, consequently higher levels of yields require higher inputs of chemical fertilizers, mainly Nitrogen. Thus, a maize cultivar with genetic potential to use N efficiently could produce economically in poor soils with low levels of fertilizer applications or high yields with better inputs of fertilizers due to its capacity to utilize N efficiently. Moll *et al.,* (1987) and Pollmer *et al*., (1979) pointed out that variation in the capacity of maize genotypes to take up N from the soil and to utilize plant nitrogen for grain production has been widely reported. Efficient and inefficient maize hybrids respond differently to the nutrient supply in the soil (Parentoni *et al.* 1998).Genetic variation in response to N supply of inbred line (Balko & Russel, 1980), has been observed and it appears to be possible to develop hybrids with tolerance to low N in soils. In most breeding programs one of the main objectives is to identify inbred lines with productive potential “per se” and high combining ability for hybrid production that express high heterotic levels for grain yield. Trials conducted at low yielding environments have a higher frequency of producing statistically non-significant differences, or having a larger coefficient of error variation than trials conducted under high yielding environments. This is because the error variance usually does not decrease as much as the genetic variance when going from high to low yielding environments (Bänziger *et al*., 1997). Despite the environmental and economical limitations related to N fertilizer utilization, traditionally, the greater majority of the maize breeding programs in the tropics are under optimum fertilization conditions ,where heritability and potential selection genetic gains are usually much greater. Maize cultivars present different behavior when grown in low levels of N and show different N partition and biomass inside the plant, especially in terms of N removed from the vegetative tissues (Ta & Wieland, 1992). Nitrogen affects cell and tissue growth, thereby influencing leaf area and photosynthetic capacity (Pan *et al*., 1985 and Settini & Maranville, 1998).

Some morphological and physiological responses of maize when in N deficiency conditions are shown through low plant height, low efficiency in light interception, accelerate senescence, increment in N mobilization to grain and reduction in N concentration in the plant (Muchow & Davis, 1988). With respect to genetics parameters related to N use efficiency, dominance effects had the great contribution to the observed genetic variance (Clark & Duncan, 1991). The genetic variation due to the general combining ability was greatly related to N plant structure (productivity and dry matter), indicating that differences among crosses could be attributed to additive gene effects (Rizzi *et al.*, 1993). The objectives of this work were to evaluate the N use efficiency of inbred lines and determine the relative importance of general and specific combining ability for N efficiency by using diallel crosses.

Material and Methods

A selected group of ten tropical inbred lines of maize derived from a tropical yellow dent tuxpeño synthetic population (CMS 61) was crossed in a diallel system through controlled hand pollination. The resulting 45 single crosses, the 10 parental lines and a control - N use efficient line, were grown in the experimental area of Embrapa Milho e Sorgo, in Sete Lagoas, MG, in 2000. For both trials was used a lattice design 7 x 8 with two replications, and plot size was a single 5 m row spacing 0,90 x 0,20m between and within rows, respectively. Each of the two nitrogen levels was considered a separate field trial plant adjacent to each other in a Latosoil dark-red, dystrophic and of clay texture, typical of the Brazilian savannas central areas, with low levels of N. The first area, with N stress (N0=10 kg ha^{-1} of N), was planted using 250 kg ha^{-1} of the formula 4:14:8 plus Zn and 20 kg ha^{-1} of FTE BR 12 (micronutrient source). The second area, with no N stress (N1=120 kg ha^{-1} of N) was used 250 kg ha^{-1} of the formula 4:14:8 plus Zn, 20 kg of FTE BR 12 (micronutrient source) and 30 kg ha^{-1} of N. At the latter, five weeks after sowing, 90 kg ha^{-1} of N (Urea) was side dressed applied. Data were collected for ear yield (EY), plant height (PH), ear height (EH), number of root and stalk lodge (NRSL), and prolificacy (Prol). Each experiment was analyzed separately as a lattice design. For evaluation of the genotype performance there were imposed two criteria**:** a) yield in the environment under N stress, and b) relative performance in the environments under no and with N stress. This performance was evaluated using a N stress index defined as: NSI= $(Y_{N1}-Y_{N0}) / (Y_{\overline{M0}}-Y_{\overline{M1}})$. Where: Y_{N1} and Y_{N0} are productions under no stress (N1) and with stress (N0); $Y_{\overline{M0}}$ and $Y_{\overline{M1}}$ are mean productions, considering all genotypes, under N0 and N1 environments, respectively. Low values for NSI indicate better genotype tolerance to N stress. Values around 1,0 indicate median tolerance, and high values are associated with low tolerance to N stress. Thus, a high standing genotype for N tolerance is the one that yields above the general mean under low level of N and with NSI value less than one (NSI<1,0). Also date for EY were analyzed using Griffing's Model I, Method 2 (Griffing, 1956) to calculate general and specific combining ability.

Results and Discussion

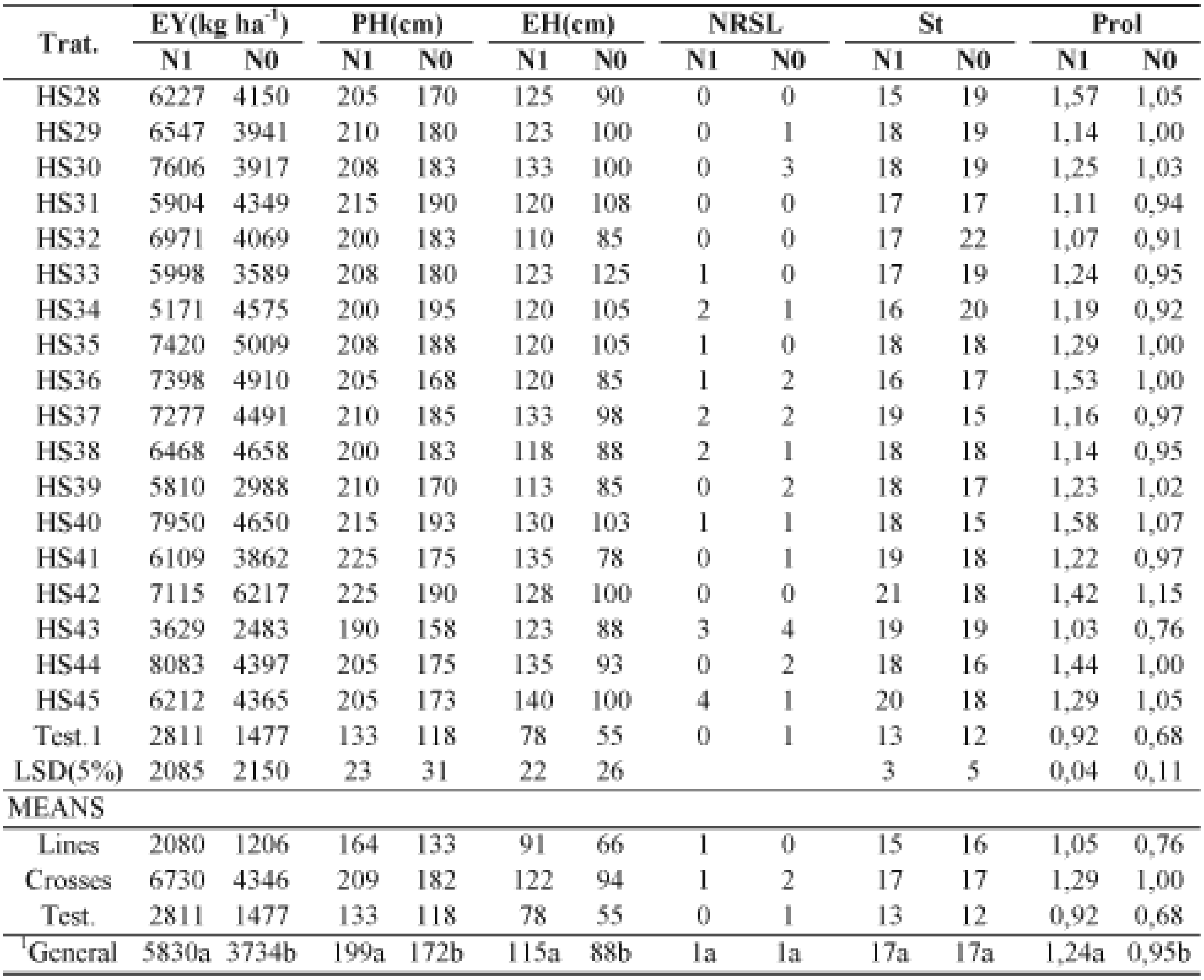
Nitrogen absorption played an important role in confirming that N stress was a major factor in creating differences in the studied traits between the environments. The use of fields that has been previously depleted of N result in a severe N stress in the low N level experiment.

Nitrogen treatment had a significant effect on ear yield (EY), plant height (PH), ear height (EH), and prolificacy (Prol) traits (Table 1).Yield reduction under low nitrogen level (N0) in comparison to higher nitrogen level (N1) treatments, in terms of average EY, ranged from 1206 kg ha^{-1} to 2080 kg ha^{-1} for lines and from 4346 kg ha^{-1} to 6730 kg ha^{-1} for the crosses. One of the main effects of N stress is a

TABLE 1. Means for ear yield (EY), plant height (PH), ear height (EH) number of root and stalk lodge (NRSL) and prolificacy (Prol) for 10 inbred lines and the diallel set of crosses among these lines grown in two levels of N.

Trat.	EY(kg ha^{-1})		PH(cm)		EH(cm)		NRSL		St		Prol	
	N1	N0	N1	N0	N1	N0	N1	N0	N1	N0	N1	N0
L1	2164	884	145	135	85	68	2	0	13	15	1,04	0,89
L2	2545	2782	163	133	88	70	0	0	15	16	1,36	1,20
L3	1614	251	155	105	78	43	0	1	16	18	1,03	0,35
L4	1173	679	158	140	85	68	2	0	17	18	0,88	0,58
L5	2121	1628	155	138	98	73	1	0	16	17	0,89	1,00
L6	2423	1460	160	138	88	65	0	0	16	17	1,02	0,80
L7	2615	834	180	125	95	60	0	1	15	16	1,07	0,75
L8	2797	1384	173	135	98	73	1	1	12	15	1,26	0,81
L9	2129	1025	183	150	98	73	0	1	14	14	1,18	0,69
L10	1220	1133	170	135	98	68	4	2	19	19	0,74	0,49
HS1	9432	6619	203	185	105	93	0	3	20	17	1,26	1,09
HS2	6213	4585	210	190	115	95	1	6	17	18	1,03	0,94
HS3	8091	4380	208	190	135	105	2	7	19	20	1,53	1,21
HS4	5623	3638	208	158	113	83	1	1	17	17	1,25	1,00
HS5	7399	4536	218	190	138	90	0	1	13	18	1,63	0,94
HS6	8386	4536	223	183	130	88	0	0	18	19	1,53	0,94
HS7	7058	3962	205	173	113	85	2	2	15	20	1,37	0,98
HS8	6902	3586	213	180	128	105	1	6	19	18	1,29	0,97
HS9	7739	4972	213	188	118	95	2	5	18	20	1,39	1,05
HS10	6456	4051	218	193	115	98	2	1	13	18	1,48	0,87
HS11	7080	4465	195	173	108	70	1	1	16	17	1,32	1,07
HS12	6196	3911	215	188	115	90	0	1	17	18	1,17	0,91
HS13	6480	4506	210	193	125	93	1	2	16	18	1,25	1,00
HS14	6840	4488	223	190	123	78	1	1	20	19	1,08	1,03
HS15	7586	4450	203	183	115	88	0	2	18	19	1,31	1,00
HS16	6504	3330	213	195	128	98	0	0	7	5	1,48	1,30
HS17	8545	4991	218	213	133	118	0	1	22	16	1,20	1,05
HS18	6105	3828	193	160	120	85	0	1	17	18	1,15	0,97
HS19	6581	4938	210	188	110	95	0	2	16	16	1,07	1,03
HS20	6761	5093	218	190	130	98	1	3	17	18	1,39	1,06
HS21	5267	6395	208	195	118	90	1	1	15	19	1,17	1,11
HS22	6413	4112	205	185	113	100	2	2	16	17	1,46	0,88
HS23	6774	4404	205	183	115	93	6	3	19	19	1,13	0,89
HS24	6909	4441	198	188	108	105	0	2	18	17	1,46	1,00
HS25	4727	3061	193	158	105	78	1	0	19	15	1,08	1,00
HS26	7486	5004	215	173	133	100	3	0	18	18	1,13	1,00
HS27	5416	2652	208	160	123	88	1	0	16	17	1,41	0,80

Continuação da Tabela 1.



Trat.	EY(kg ha^{-1})		PH(cm)		EH(cm)		NRSL		St		Prol	
	N1	N0	N1	N0	N1	N0	N1	N0	N1	N0	N1	N0
HS28	6227	4150	205	170	125	90	0	0	15	19	1,57	1,05
HS29	6547	3941	210	180	123	100	0	1	18	19	1,14	1,00
HS30	7606	3917	208	183	133	100	0	3	18	19	1,25	1,03
HS31	5904	4349	215	190	120	108	0	0	17	17	1,11	0,94
HS32	6971	4069	200	183	110	85	0	0	17	22	1,07	0,91
HS33	5998	3589	208	180	123	125	1	0	17	19	1,24	0,95
HS34	5171	4575	200	195	120	105	2	1	16	20	1,19	0,92
HS35	7420	5009	208	188	120	105	1	0	18	18	1,29	1,00
HS36	7398	4910	205	168	120	85	1	2	16	17	1,53	1,00
HS37	7277	4491	210	185	133	98	2	2	19	15	1,16	0,97
HS38	6468	4658	200	183	118	88	2	1	18	18	1,14	0,95
HS39	5810	2988	210	170	113	85	0	2	18	17	1,23	1,02
HS40	7950	4650	215	193	130	103	1	1	18	15	1,58	1,07
HS41	6109	3862	225	175	135	78	0	1	19	18	1,22	0,97
HS42	7115	6217	225	190	128	100	0	0	21	18	1,42	1,15
HS43	3629	2483	190	158	123	88	3	4	19	19	1,03	0,76
HS44	8083	4397	205	175	135	93	0	2	18	16	1,44	1,00
HS45	6212	4365	205	173	140	100	4	1	20	18	1,29	1,05
Test.1	2811	1477	133	118	78	55	0	1	13	12	0,92	0,68
LSD(5%)	2085	2150	23	31	22	26			3	5	0,04	0,11
MEANS												
Lines	2080	1206	164	133	91	66	1	0	15	16	1,05	0,76
Crosses	6730	4346	209	182	122	94	1	2	17	17	1,29	1,00
Test.	2811	1477	133	118	78	55	0	1	13	12	0,92	0,68
[1]General	5830a	3734b	199a	172b	115a	88b	1a	1a	17a	17a	1,24a	0,95b

[1] Means followed by the same letter are not statistically different (P=0.05).

reduction in the photosynthesis structures components levels, e.g. chlorophyll, resulting in a reduction in the photosynthesis capacity also in carboxilase efficiency and, therefore low yielding (Delgado *et al*., 1994; Settini & Maranville, 1997). Guang Jauh *et al*., (1995), working with maize hybrids from a 6 parent half-diallel cross pointed out that in general grain yield showed high heterosis at the higher nitrogen levels. Yield under N0 level was lowest for line L 3 and highest for line L 2, and yield under N1 level line L 4 was lowest and highest for line L 8. Differences among inbred lines and single crosses were observed for PH, EH and Prol traits. Inbred L3 presented the lowest values under low N level, and there were consisting differences for the results under high level of N, for these three traits. For the trait Prol, means of single crosses and inbred lines were greater under N1 level than for N0 level, maybe due to the reduction in PH and EH. Nitrogen treatments had an important effect on the means of inbred lines under N1 in comparison with N0 resulting in a reduction of 18,9% (PH), 27,5% (EH) and 27,6% (Prol). Similarly, the reduction for single crosses means were 12,9%, 22,9% and 22,5% for

PH, EH and Prol, respectively. Thus, as was expected reductions in terms of average performance were lower for single crosses than for the lines. Similar results were found by Laffite *et al*., 1995, Kling *et al*., 1997; and Arellano *et al*. 1997. The analysis of variance for ear yield showed significant (P<0,01) for genotypes under N1 and N0 levels (Table 2).

TABLE 2. Summaries of the analysis of variance for ear yield (kg ha^{-1}) for maize inbred lines and a diallel set of 45 crosses among these lines in experiments with two levels of N. EMS/ EMBRAPA,SL(MG), 2002.

SV	DF	MS	F
		High N (N1)	
Genotype	54	84.856.487.706	**
G.C.A.	9	34.453.093.416	**
S.C.A.	44	94.937.166.564	**
Error	40	10.656.020.055	
		Low N (N0)	
Genotype	54	42.629.329.923	**
G.C.A.	9	15.785.081.132	ns
S.C.A.	45	47.998.179.681	**
Error	40	11.333.520.108	

CV(N1) = 17,70 %; CV (N0) = 27,50 %
** significant at the 1% level of probability by F test; ns= Non significant

Significant (P<0,01) was detected for SCA under both N treatments, but GCA was only significant (P<0,001) under N1. Arellano, *et al*. 1997, found significant differences for GCA and SCA effects in a study involving a diallel of 14 inbred lines, and because SCA effects for yield were larger under low N level, they consider that these effects were more important under low than under high N availability. On the other hand, Below *et al*. (1997) working with six temperate lines found that additive effects were more important than dominance effects. Estimates of the SCA and GCA effects for both N levels, for EY of the 10 inbred lines are presented in Table 3. GCA and SCA effects differed according to the level of available nitrogen. Although the additive effects have been detected, the SCA effects predominated over the GCA. According to Baker (1978), for the non-random diallel cross model or variance, the proportion of GCA in relation to SCA can be calculated by GCA/(GCA+SCA). The value of this proportion for the trait EY, were 26,8% and 24,7% for N1 and N0 conditions, respectively. The GCA estimated effects were positive for the lines L1, L2, L3, L6, L7 and L10, under N0 level. Under N1 level, lines L1, L2, L6, L7, L8 and L10 also presented favorable GCA effects. Therefore, five out of the ten lines present high GCA effects in both N0 and N1 levels. Lines L4, L5 and L9 had negative GCA effects at both levels of N. This led to a conclusion that the involvement of additive gene action should facilitate selection efforts for better line identification under non-stress environments. Shieh (1995) and Morarium (1994) evaluating diallel crosses and levels of nitrogen, found significant difference for GCA for the genotypes at the same N level and SCA and GCA were consistent for most of the genotypes over different N levels. Normally, breeders are interested in hybrid combinations, with more favorable SCA and where there is at least one of the lines with more favorable GCA effect. Therefore, crosses like L1 x L2 should be highly desirable.

Therefore, our results agree with earlier results found (Bellow *et al*., 1997; Laffite & Edmeades, 1995; Rizzi *et al*, 1995) that show yield of inbred lines related to N use are under genetic control. As shown by the results of the others traits, the identification of traits related to EY at high and low N levels could allow the development or identification of hybrids with high performance to stress environments.

A further part of this study was the examination of the most promising lines to N efficiency. As seen in Figure 1, quadrant 1, characterized by low

TABLE 3. Estimates of specific (above diagonal - N1 and below diagonal - N0) combining abilities (SCA) and general combining abilities (GCA N1 and GCA N0) for ear yield, for a diallel set of crosses among to inbred lines, in experiments with two levels of Nitrogen. EMS/EMBRPA, SL,(MG),2000

	L1	L2	L3	L4	L5	L6	L7	L8	L9	L10	GCA N1	GCA N0
L1	-	2554,301	127,002	1928,117	-306,119	886,708	1802,193	546,318	993,923	1055,549	536,412	88,238
L2	2351,437	▨	450,116	997,091	346,269	47,202	336,836	1155,072	676,392	1941,605	456,274	404,223
L3	652,778	-197,331	▩	813,711	1523,609	1120,633	-445,050	773,259	1738,403	1097,365	-335,476	68,829
L4	914,417	638,376	381,588	▩	-407,789	1768,601	-372,923	510,718	1433,840	1717,435	-258,490	-397,229
L5	-131,146	-174,184	1188,874	-222,703	▩	419,096	1415,743	514,492	290,616	1764,533	-491,653	-94,007
L6	512,152	165,498	1088,997	1465,201	507,496	▩	1259,740	1211,127	1005,436	428,725	91,370	161,017
L7	512,486	148,410	2390,709	-885,954	228,197	814,094	▩	1812,230	574,891	805,806	162,504	160,335
L8	304,818	476,618	474,420	978,399	113,925	761,342	920,656	▩	-1833,006	1846,097	90,108	-205,793
L9	78,600	-493,651	915,934	919,537	1249,541	1077,890	282,703	-730,465	▩	578,392	-513,208	-355,658
L10	938,639	642,384	427,381	362,144	1158,083	-1117,870	2112,135	658,302	776,210	▩	262,154	170,044

DP (Gi) = 199,991 e DP (Gi - Gj) = 297,997 (N1)   N1 = 120 kg/N ha^{-1} and, N0 = 10 kg/N ha^{-1}
DP (Gi) = 206,160 e DP (Gi - Gj) = 307,322 (N0)
DP (Sij) = 672,363 e DP (Sij - Six) = 988,332 (N1)
DP (Sij) = 693,415 e DP (Sij - Six) = 1019,24 (N0)

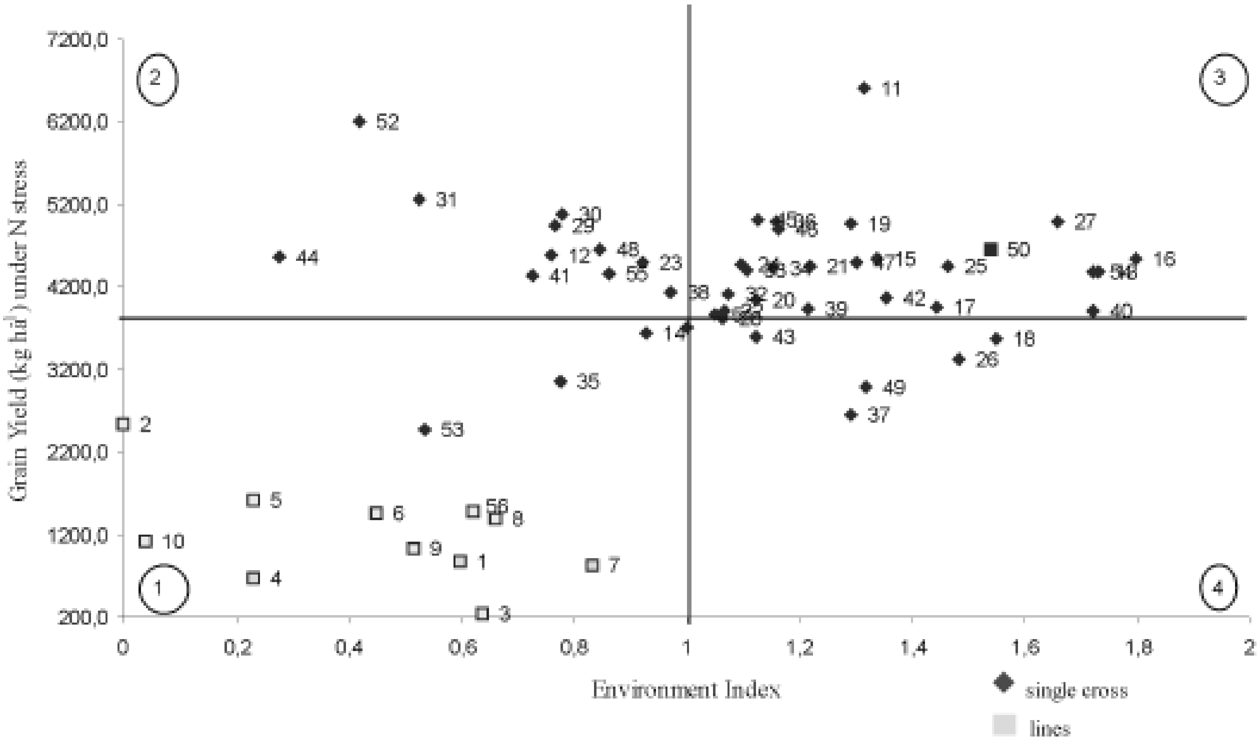


FIGURE 1. Ear yield as or function of environment index (EI) for10 lines, 45 single crosse, and 1 line tester trials under two N levels. SL (MG), 2000.

NSI (N Stress Index) and low yield, was composed of all the lines and the inbred tester. Quadrant 3, characterized by high NSI and yield, was composed of 17 single crosses being classified as high yielding and responsive to N stress.

These findings are of importance to breeders who want to select more efficient genotypes to abiotic stresses such as N stress ,which is one of the most widely stresses occurring in maize farmers' fields in the tropics.

Conclusions

The productivity of the inbred lines and the single crosses were different and were dependent on the N level.

Under low N level was observed a reduction in the traits EY, PH, EH, and Prol.

For the trait EY, non-additive gene action was predominant under both environments and additive only in the non stress environment.

The highest SCA estimates were observed in crosses between parents contrasting in magnitude and sign of GCA.

Some lines and hybrids identified in this study showed to be promising for growing under conditions of low soil nitrogen availability.

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