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Improvement for Tolerance to Low Soil Nitrogen in A Quality Protein Maize (QPM) Variety: Variability and Selection Gains

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Abstract

A set of 250 S_1 lines of a quality protein maize (QPM), ART/98/ILE 1-OB, were evaluated along with six checks under low soil N in Mokwa and Zaria in 2016 to determine the level of genetic variability in the QPM population with the aim of improving it for tolerance to low soil N. Data were collected on days to flowering, Anthesis-Silking Interval (ASI), plant height, stay green ability, plant aspect, number of ears per plant and grain yield under low and high N. Analysis of variance was conducted from which genetic variances were estimated. Heritability and selection gains were also estimated. Mean square of entry was significant for all the traits studied under low N except for days to silking and ASI. Genetic variance estimates ranged from low to moderate but significantly different from zero for most of the traits studied. Estimates of environmental variance were higher than genetic variance for most of the traits except plant height and grain yield. Heritability estimate was moderate for most of the traits studied. It was however highest for grain yield under low N (49.7%). Gain/cycle was also moderate to high for most of the traits. Stay green ability would improve with selection by 6.07% of the mean. ASI would reduce by -3%, while grain yield would increase by 71% of the mean yield. Appreciable genetic variability exists in the QPM population for tolerance to low soil N and hence, selection will be effective.

Keywords: Low soil nitrogen, Quality Protein Maize (QPM), Selection gains, Variability.

Introduction

Maize has always been preferred to any other crop, including cassava because most of the world's civilizations developed around grains rather than tuber crops (Fakorede, 2001). Normal maize has a major nutritional constraint as human food because the protein content (about 10%) is deficient in two essential amino acids-lysine and tryptophan. Nigeria and some sub-Saharan Africa countries are faced with problem of malnutrition (protein deficiency) most importantly in children due to poverty, causing a disease known as Kwashiorkor. In livestock industry, especially monogastrics production, the use of low quality protein maize as feed ingredient introduces an extra cost in the form of protein supplements necessary to meet dietary protein requirements.

The discovery of quality protein maize (QPM) with recessive *Opaque-2 mutant* gene has now alleviated the problem of this deficiency in protein quality of maize. QPM contains twice the levels of tryptophan and lysine in most normal endosperm maize. It was developed by combining the genetic systems of the mutant opaque-2 (O2) gene and O2-endosperm genetic modifiers

(Ngaboyisonga et al., 2012). The protein of QPM has 90% of the relative value (RV) of milk compared to 40% for normal maize (Badu-Apraku & Fontem-Lum, 2010). Quality protein maize which is cheaper, more affordable and easier to produce compared to animal protein, deserves further attention for the benefit of mankind.

Soils of the major maize producing ecologies in Sub-Saharan Africa are inherently low in nitrogen (N) which limit maize yield. The effect of this low N stress on QPM characteristics is not well known, nitrogen being an essential element in protein. Increasing soil nitrogen has been reported to increase protein quality of maize (Kniep &Mason 1989; Oikeh et al., 1997, Naik, 2011). However, the increase in price of chemical fertilizer leads to the sub-optimal N-fertilizer application on farmers' field. The estimated annual loss of grain yield due to low N stress alone was reported to be between 10-50% (Wolfe et al., 1988; Logrono &Lothrop, 1997) which amount to loss of millions of naira. Therefore, development of maize genotypes with tolerance to low N is crucial to increase productivity of maize in Sub-Sahara Africa. Various low soil nitrogen tolerant maize varieties have been developed through the effort of International Institute of Tropical Agriculture (IITA) and International Maize and Wheat Improvement Centre (CIMMYT) but most of these maize varieties are non-QPM.

To be commercially successful, QPM cultivars must be agronomically competitive with normal endosperm cultivars for low soil N tolerance while maintaining the expected protein quality level. To improve QPM for tolerance to low soil nitrogen, understanding of the gene action controlling the trait and the level of variability in the maize population for tolerance to low soil N is essential. This study therefore aims at determining (i) the level of variability in the QPM population for tolerance to low soil nitrogen and (ii) the level of gains that is attainable in selection towards improving the population.

Materials and Methods

The quality protein maize (QPM) variety, ART/98/ILE 1-OB was planted and about 400 plants were self-pollinated. Out of these, 250 S_1 were randomly selected and evaluated along with six checks under low (LN) and high soil nitrogen (HN) in Zaria and Mokwa, Nigeria in 2016. The study was conducted at the low N screening site of both locations. Soil samples were taken for physico-chemical analysis before the experiment. The result of the soil analysis showed that soil in Mokwa is luvisol with 0.06% N, 35.66ppm P and 0.22cmol/kg K on low N plots, and 0.07%N, 13.94ppm P and 0.25cmol/kg K on the high N field. Soil in Zaria is a fine-loamy, isohyperthermic plinthusstalf; USDA taxonomy with 0.03%N, 2.79ppm P and 0.40cmol/kg K on the low N field, and 0.034% N, 4.72ppm P and 0.22cmol/kg K on the high N field.

The experiment was laid out in a 16 x 16 alpha lattice design in two replications. The high N and low N fields were separated 5m apart to avoid any seepage of nitrogen. In each field, each S_1 was planted in single row plot of 5 m long with 0.75 m space between rows and 0.25 m apart within rows. Thinning was done three weeks after planting (WAP) to one plant per hill to have a plant population of 53,333 plants/ha. In the LN trial, urea fertilizer was applied at rate of 30kgN/ha in a split dose, whereas the HN trial received 90kgN/ha urea, insplit dose at 2 and 4 WAP. Fields were kept weed-free throughout the trials. Both high and low N fields received 60kgP_2O_5/ha P as single superphosphate and K as muriate of potash at rate of 60kgK_2O/ha

Data collection

Days to anthesis and silking were recorded in each of the plot as the number of days from sowing to when half of the plants shed pollen grains and emerged silks, respectively. Anthesis-silking interval (ASI) was computed as the interval in days between silking and anthesis. Plant height was measured in centimeters as the distance from the base of the plant to the height of the first tassel branch. Plant aspect was rated on a scale of 1 to 9, where 1 = excellent overall phenotypic appeal and 9 = poor overall phenotypic appeal. Stay green ability was scored only under LN at 10 WAP on a scale of 1 to 9, where 1 = less than 10% senesced leaf and 9 = more than 80% senesced leaf area below the ear. The number of ears per plant was estimated as proportion of total number of ears divided by the number of harvested plants. Few ears were shelled from each plot to determine percentage moisture. Grain yield adjusted to 14% moisture was estimated from field weight at 80% shelling percentage.

Statistical analyses

Mean, CV and ranges were estimated. Combined analyses of variance (ANOVA) was performed separately for traits recorded under LN and HN using a random model in SAS version 9.2 (SAS Institute, 2009). From ANOVA, genetic variances and heritability were estimated **as** described by Hallauer and Miranda (1988). The expected response from selection (Gs) was also computed according to Hallauer and Miranda (1988) as:

$$Gs = k\,\sigma^2 g / \sigma p$$

Where σp is the square root of phenotypic variance, and k is selection intensity (10% selection intensity was used).

Gains were expressed as percentage of means for ease of comparison. Standard error of genetic variance and heritability estimates were also calculated using the method of Hallauer & Miranda (1988). A random mating population in linkage equilibrium without epistasis was assumed in this study.

Results and Discussion

Mean, CV and ranges are as shown in Table 1. Ranges were high and means were significant for most of the traits studied. CV was moderate for most of the traits except in the derived traits. Days to flowering and ASI were longer under LN compared with HN condition. Plants under HN were also taller with mean of 133cm than under LN (121cm). Plant aspect and stay green ability were fairly good under LN. Even though number of ears were more under LN than under HN, grain yield was significantly higher under HN condition (2.10tons/ha) than under LN (0.76tons/ha) (Table 1).This result revealed that ASI could be longer under LN as earlier reported by Banziger et al.(2000). Higher grain yield under HN is expected. This is because grain yield is a function of cob number and weight. Usually under LN, there is poor grain filling, giving rise to many small and poorly-filled cobs. The aggregate of these small poorly-filled cobs are usually not able to compete with aggregate of well-filled cobs under optimum condition.

Combined analysis of variance under low soil nitrogen is shown in Table 2. Mean square of entry was significant for all the traits except days to silking and ASI.

Table 1. Mean± SE, CV and ranges for traits under low and high soil N in the QPM population at Zaria and Mokwa in 2016

Traits	N level	Mean ± SE	CV (%)	Range
Days to silking	LN	63.48 ± 8.55	13.47	0 - 75
	HN	61.61± 0.36	15.0	0 -77
Days to pollen shed	LN	63.12 ± 3.86	6.11	0 -74
	HN	60.79 ± 0.28	10.1	0 -73
Plant height (cm)	LN	120.65 ± 12.52	10.38	0 -205
	HN	133.03±1.17	12.79	0 - 210
Plant aspect (1 – 9)	LN	4.80 ± 0.65	13.43	2 -7
	HN	6.10 ± 0.03	10.20	0 - 9
Stay green ability (1 – 9)	LN	4.68 ± 0.63	13.38	2- 9
	HN	-	-	-
Anthesis-Silking Interval	LN	4.99 ± 0.73	14.67	-4 -6
	HN	1.59 ± 0.05	65.89	-3 - 5
Ears per plant	LN	1.36± 0.83	60.9	0 – 2.0
	HN	0.76 ± 0.01	30.6	0 -2.0
Grain yield (tons/ha)	LN	0.76± 0.23	30.28	0.09 - 9.9
	HN	2.10 ± 0.04	30.0	0-9.6

SE: standard error; (1 -9): 1 for excellent, 9 for poor; CV: coefficient of variation
LN: Low nitrogen; HN: High nitrogen

Significant mean square of entry under low N was also reported by Meseka et al. (2006). Mean square of environment was significant for all the traits except stay green ability. Mean square of entry by environment interaction was also significant for all the traits except days to flowering. On the other hand, combined analysis of variance under HN did not revealed significant mean squares for entry for days to pollen shed, plant aspect, ASI and number of ears per plant (Table 3). However mean square of environment was significant for all traits under HN, while mean squares of entry by environment interaction was significant for all the traits studied except plant aspect and ASI. Significant mean squares of entry by environment interaction under low and high N for most of the traits suggested that the entries responded differently to the two environments. Mafouasson (2014) gave similar report for grain yield under low N.

Estimates of genetic variance, heritability and genetic gains under low soil nitrogen are presented in Table 4. Genetic variance estimates ranged from low to moderate but significant for most of the traits studied. Genetic by environment and environmental variance components were higher than genetic variance estimates for days to silking, plant aspect, stay green ability, ASI and number of ears per plant resulting in high phenotypic variance. Estimate of environmental variance was

Table 2. Mean squares from combined ANOVA for traits under low soil N in the QPM population in Zaria and Mokwa in 2016

Source	df	Days to silking	Days to pollen shed	Plant height (cm)	Plant aspect (1 – 9)	Stay green ability (1 – 9)	Anthesis-Silking Interval	Ears per plant	Grain yield (tons/ha)
Env	1	9735.01**	22600.52**	47110.18**	1.57*	0.34	413.93**	9.15**	358.49**
Rep(Env)	2	208.10	27.52	4132.46**	1.76*	2.41**	0.5	0.1	4.90**
Block(Env*Rep)	60	108.11*	29.90**	1622.39**	3.56**	3.25**	0.74	0.12**	2.92**
Entry	249	95.34	24.41**	679.94**	1.30*	1.11**	0.69	0.10*	1.53**
Env*Entry	249	85.16	17.67	395.24**	1.00**	0.77**	0.86*	0.08**	0.77**
Error	436	73.13	14.89	156.71	0.42	0.39	0.53	0.68	0.05

df: degree of freedom; *, **: significant at P = 0.05 and 0.01 respectively

Table 3. Combined ANOVA for traits under high soil N in the QPM population in Zaria and Mokwa in 2016

Source	df	Days to silking	Days to pollen shed	Plant height (cm)	Plant aspect (1-9)	Anthesis-Silking Interval (1-9)	Ears per plant	Grain yield (tons/ha)
Env	1	1260.40**	7255.60**	330457.10**	26.93	393.21*	0.29**	52.26**
Rep(Env)	2	61.25**	57.12**	20995.20**	6.05	0.07	0.02	14.47**
Block(Env*Rep)	60	417.94*	94.61*	3172.71**	3.80	1.81*	0.17*	9.53**
Entry	249	93.76**	50.88	755.43**	0.74	1.13	0.12	1.69*
Env*Entry	249	175.40*	82.02*	583.87*	0.94	1.12	0.15**	1.96**
Error	436	83.20	11.32	160.1	1.00	1.09	0.05	0.04

df: degree of freedom; *, **: significant at P = 0.05 and 0.01 respectively

Table 4. Genetic variances, heritability estimates and genetic gains under low soil N in the QPM population in Zaria and Mokwa in 2016.

Trait	σ^2g ± SE	σ^2ge	σ^2e	σ^2ph	H^2% ± SE	Expected gain	% gains/cycle
Days to silking	2.55± 2.85	6.02	18.28	23.84	10.68± 2.74	0.92	1.45
Days to pollen shed	1.69± 0.67	1.39	3.72	6.10	27.61± 0.61	1.20	1.90
Plant height	71.18±17.55	119.27	39.18	169.99	41.87± 14.50	9.61	7.96
Plant aspect (1-9)	0.08± 0.04	0.29	0.11	0.33	23.08±0.42	0.23	4.81
Stay green ability (1 – 9)	0.09±0.03	0.19	0.10	0.28	30.63±0.48	0.28	6.07
Anthesis-Silking Interval (1-9)	-0.04± 0.02	0.17	0.13	0.17	-24.64±0.75	-0.18	-3.61
Ears per plant	0.01± 0.003	-0.30	0.17	0.03	20.00±5.00	0.06	4.09
Grain yield (tons/ha)	0.19± 0.06	0.36	0.01	0.38	49.67±0.36	0.54	71.14

σ^2g: Component of variance due to genotypes; σ^2ge: Component of variance due to genotypes by environment interaction; σ^2e: environmental variance; σ^2ph: Phenotypic variance; H^2: Broad-sense heritability; SE: Standard error.

Scale 1-9: 1 for excellent; 9 for poor

higher than genetic variance for most of the traits except grain yield and plant height. The higher environmental variance for days to flowering, plant aspect, ASI and number of ears per plant is an indication that environmental force had strong influence on expression of these traits. This suggests that there may be need for testing in wide arrays of sites and across years. Banziger et al. (2000) however stated that low N stress usually increase over time, thus relatively severe low N stress regime should be sufficient to assess low N stress tolerance.

Negative genetic variance and heritability were recorded for ASI. Heritability estimate ranges from 10.7% for days to silking to 49.7% for grain yield. Days to pollen shed, stay green ability and plant height also had reasonable heritability estimates of 27.61%, 30.6% and 41.87% respectively. The high heritability for plant height, stay green ability, days to anthesis and grain yield suggests that the traits are under genetic control and improvement would be made through selection in advance generations for these traits. The high heritability and low phenotypic variance reported in the present study for grain yield under low N is contrary to the report of Ajala et al. (2018) who recorded low heritability for grain yield under low N. High heritability alone is not enough for sufficient improvement in selection (Hallauer & Miranda 1988). Heritability should be accompanied with substantial genetic advances for adequate progress from selection. This reflected in the traits studied which are suggested to be traits to select for under low soil N trials according to Banziger et al. (2000).

The wide ranges, moderate to high heritability estimates and significant mean squares of entries revealed that high genetic variability exists in the QPM variety for good progress to be made from selection for tolerance to low soil nitrogen and grain yield. S_1 recurrent selection has been reported to be an efficient method for increasing the frequency of favourable alleles in a population (Sprague & Eberhart 1977; Hallauer & Miranda 1988). Many reports have established that additive gene action, and both additive and non-additive gene actions control tolerance to low soil N in both QPM and non-QPM varieties which further supports the use of S_1 recurrent selection in improving this maize population (Wegary et al., 2011; Annor & Badu 2016; Njeri 2017; Kling et al., 1997; Badu et al., 2015; Tapera 2017; Mafouasson 2014; Meseka et al., 2006).

Reasonable genetic gains are expected from selection under LN for all the traits studied. There would be a slight increase in days to silkng and pollen shed with selection by 1.45 and 1.9% of the means respectively. ASI will reduce by 3% per cycle. Stay green ability will improve with selection by 6.07% of the mean. Plant height will also increase with selection by about 8%. Number of ears per plant will increase by 4.09% per cycle, while gain of 71% is attainable with selection for grain yield. This amount to about 0.53tons/ha increase per cycle. The reduction in ASI per cycle is desirable under low soil nitrogen which suggests rapid progress because ASI tends to be high under low soil nitrogen stress (Banziger et al., 2000). They also reported increase in plant height with selection but this is not desirable since tall plants are prone to lodging. Therefore, breeders should stick to a standard plant height during selection. The reduction in ASI and increased gain in plant height and grain yield with selection under low N in the present study is similar to the report of Ajala et al. (2018). The percentage increase reported also falls within the percentage increase in the present study. Ajala *et al.* (2012) also reported desirable changes in gene frequency after 3 cycles of selection for grain yield under low soil N without adverse effect on other agronomic traits. Badu *et al.* (2017) testing three eras of maize under low N reported genetic gains of 13.29% and 16.84% in grain yield per cycle per era under low N and high N, respectively.

Interaction between application of N fertilizer and protein quality of QPM varieties has been reported. Jaliya *et al.* (2015) reported that there was a significant effect of interaction between nitrogen and sulfur on up-take of both nitrogen and sulfur by quality protein maize (QPM). Naik (2011) recorded higher protein content (11.72%) and higher grain yield in QPM when 10 tons of farmyard manure and 100% recommended fertilizer rate was used, but a lower protein content (10.09%) when the recommended fertilizer rate was reduced to half. Buah et al. (2009) reported that grain yield had a linear and quadratic response to N application. Grain yield of QPM increases as a result of 90 kg N/ha application. However, Zaidi et al. (2008) studied the stability performance of CIMMYT tropical and subtropical elite QPM hybrids across stressed (drought at ?owering stage and low soil N) and non-stressed environments and found that the variation in protein quality across environments was statistically signi?cant, but was largely due to genotypic variability.

Conclusion

It can be concluded that sufficient genetic variability exists in ART/98/ILE 1-OB to be exploited for improvement for tolerance to low soil N. S_1 family selection will be effective in improving the maize population. The influence of N application on the protein quality however requires further study.

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