Evaluation of biochemical and yield attributes of quality protein maize (*Zea mays* L.) in Nigeria

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Twenty two genotypes of quality protein maize (QPM) and two local checks were assessed for their lysine and tryptophan levels, as well as grain yield characteristics at the Lower Niger River Basin Development Authority station, Oke-Oyi, Ilorin, Nigeria for three years (2009-2011). The results showed that the QPM genotypes and the standard checks varied from one another, with respect to crude protein, zein dry matter, zein crude, lysine and tryptophan. The best QPM hybrids for grain yield (Dada-ba, ART98-SW5-OB, ART98-SW4-OB and TZPB-OB had percentage lysine and tryptophan advantage of 34% compared with the local checks. These hybrids also out-yielded other genotypes with yield advantage of 10, 24 and 26% over the best inbred, open pollinated variety and the standard check respectively. However, grain yield showed positive association with all the characteristics except crude protein content. Kernel number per cob had maximum correlation with grain yield followed by kernel rows per cob, cob diameter and cob weight. The direct effect for crude protein was positive but the correlation was negative. Conclusively, the QPM hybrids that combined high yield with the essential amino acids could be tested in different savanna agro-ecologies to identify those that could be released to farmers, while the superior inbreds could be introgressed for further breeding programs.

Keywords**:** Lysine, tryptophan, inbred lines, hybrids, open pollinated varieties

Maize (*Zea mays L.*) is the third most important cereal crop and a major source of energy, protein and other nutrients for human and livestock in the world (Jompuk et al. 2011; 666-674). Maize grain accounts for about 15 to 56% of the total daily calories in diets of people in about 25 developing countries, particularly in Africa and Latin America, where animal protein is scarce and expensive (Prasana et al. 2001, 1308-1319). In Nigeria, maize is the most widely available staple foods, 18.8 % in the dry savanna, 21.9% in the moist savanna and 19.8 % in the humid forest (Ibikunle et al. 2009, 9-15). Available daily intake is generally low in tropical Africa (5g in Nigeria) compared to 46-56g for an average person, and 96g for pregnant and lactating mothers as recommended by Food and Nutrition Board of the Academy of Science. A national survey revealed that an average of ₦127.20 ($0.8) is spent per week in each household on maize consumption (Ibikunle et al. 2009, 9-15). Millions of African children and nursing mothers suffered from protein deficiency inducing diseases such as stunted growth, weakened immune system and impaired intellectual development, because of poverty (Prasanna et al. 2001; 1308-1319).

An adult human cannot synthesize the eight essential amino acids (isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine) which need to be supplied through foods. Absence or deficiency of any of these amino acids limits the ability of the body to make proteins, despite the presence of all other amino acids (Matta et al. 2009; 439-446). Cereal proteins contain on an average about 2% lysine, less than one-half of the recommended concentration for human nutrition by Food and Agriculture Organization (FAO, 2005).

From the human nutrition view point, lysine is the most important limiting amino acid in the maize endosperm protein, followed by tryptophan.

Quality protein maize (QPM) was developed by combining the genetic systems of the gene mutant *opaque-2* (δ^2) (Mertz et al. 1964, 279-280) and genetic endosperm modifiers (Vasal 2000, 445-450 Prasanna et al. 2001, 1308-1319 Vasal 2001, 80-121 Gupta et al. 2009, 230-237). The genetic system of the δ^2 gene is qualitative. However, because it is recessive, the effects are expressed in the endosperm when three alleles, two from female parent and one from male parent are present. It increases lysine and tryptophan in endosperm by acting on the four types of storage proteins in maize endosperm: albumins, globulins, zeins, and glutelins. Zeins contain low lysine with 0.1g/100g while glutelins are considerably rich in lysine with 2g/100g or more (Lin et al. 1997, 355-362). The δ^2 mutant increases the level of lysine and tryptophan by suppressing or reducing the synthesis of zeins and increasing that of glutelins (Habben et al. 1993, 825-838). The δ^2 gene adversely affects several important agronomic traits including kernel characteristics. It adversely affects the accumulation of dry matter resulting in lower yields due to increased endosperm size. The kernel phenotype is changed in a soft, and dull appearance. Kernels dry slowly following physiological maturity of the grain and have a higher incidence of ear rots. Other changes include larger germ size and low kernel density (Moro et al. 1995, 94-99).

The δ^2-endosperm genetic modifiers however are genes capable of altering the expression of other genes at different loci in the genome, and alter the undesirable correlated effects of δ^2 gene (Thain et al. 2003). The parties of the endosperm modified are vitreous and hard instead of being opaque and soft (Villegas et al. 1992, 27-48). The δ^2-modified endosperm varieties have agronomic characteristics comparable with those of normal maize. Endosperm modification of QPM is also accompanied by slight increase in total proteins and fast decrease in lysine and tryptophan (Bjarnason et al. 1992; 181-216; Vasal 2000, 445-450). Several conventional breeding programs that improved those agronomic shortcomings and amino acid contents through backcrossing and recurrent selection, have developed varieties with high protein value and favorable texture of QPM (Prasanna et al. 2001, 1308-1319).

Previous studies reported lysine content of 1.80-4.5% in QPM genotypes. These values were less than those reported by some researchers in wheat, rice, barley, oats, sorghum and normal maize (Vasal 2005). Tryptophan contents of 0.94-1.06% recorded in QPM were two-fold greater than those reported for normal maize (Olakojo et al. 2007, 97-104). Earlier workers (Gupta et al. 2009, 230-237; Upadhyay et al. 2009, 9-14; Mbuya et al. 2010, 325-332) suggested that QPM could assist in reducing protein deficiencies, especially in children where maize consumption dominated in their diets. The QPM is also cheaper, more affordable and easier to produce compared to animal protein (Olakojo et al. 2007, 97-104). Hence, breeding and production of QPM stands out as an alternative protein source for poor-resource farming communities. Against these backgrounds, evaluation of QPM yield potentials and amino acid contents in the target environment is prerequisite for adoption by the farmers. Keeping in view with the aforementioned, the present study was therefore conducted to (i) evaluate some QPM, inbred lines, hybrids and open pollinated genotypes developed for acceptable level of tryptophan and lysine (ii) Assess them for grain yield characteristics in the southern Guinea savanna agro-ecology for three cropping years.

Materials and Methods

Description of genetic materials

Twenty two QPM genotypes including five open pollinated maize varieties (OPVs), nine inbred lines, eight hybrids and two local checks used as control were evaluated for nutritional qualities and grain yield characteristics during three years (2009-2011) in late cropping seasons

at the Lower Niger River Basin Development Authority station, Oke-Oyi, Ilorin (Latitude 8^0 30'N, 8^0 36'E and Longitude 4^0 31'N, 4^0 33'E), located in the southern Guinea savanna of Nigeria. The seeds of nine inbred lines and four OPVs were obtained from International Institute of Tropical Agriculture (IITA), while five hybrids from Institute of Agricultural Research and Training, (IAR & T), Ibadan, Nigeria. One of the OPVs (Obatanpa) and three hybrids were obtained from the Crops Research Institute (CRI), Kumasi, Ghana (Table 1).

Experimentation

The trials were established on 1st August, 2009, 29th July, 2010 and 27th July, 2011. The trials were laid out each cropping year in a randomized complete block design (RCBD) with four replications, and at a planting distance of 0.75m between the rows and 0.5m between plants within the rows. Each plot was six rows and the outer rows were used for destructive sampling, while observations were taken from the four middle rows. Three seeds were planted/hill and later thinned to two/hill, two weeks after planting (WAP) to provide a uniform plant population of about 53,333 plants ha^{-1}. Agronomic practices included pre-emergence application of herbicide ((a.i. 3kg l^{-1} Metolachlor, 170g l^{-1} Atrazine and 3kg l^{-1} Paraquat per hectare) to control weeds and supplemented by hoe weeding when necessary. Compound fertilizer (NPK 20:10:10) was applied as split-dosage at the rate of 80kg ha^{-1} of N, 40 ha^{-1} of K_2O and 40 ha^{-1} of P_2O_5 (3 WAP) and at anthesis (7 WAP), as side dressing.

Table 1: Source of collection and characteristics of the QPM varieties evaluated at Oke Oyi, Ilorin, Nigeria

Varieties	Source of collection	Seed colour	Kernel texture	Type
Obatanpa	CRI Ghana	White	Flint	OPV
EV8766-SR	IITA/CIMMYT	Yellow	Dent	OPV
EV8363-SR	IITA/CIMMYT	White	Dent	OPV
Pool 18-SR	IITA/CIMMYT	Yellow	Flint	OPV
Pool 19-SR	IITA/CIMMYT	White	Flint	OPV
Mama-ba	CRI Ghana	White	Flint	Hybrid
CIDA-ba	CRI Ghana	White	Dent	Hybrid
Dada-ba	CRI Ghana	White	Flint/ Dent	Hybrid
ART98-SW6-OB	IAR & T, IBADAN	White	Flint/ Dent	Hybrid
ART98-SW5-OB	IAR & T, IBADAN	White	Flint	Hybrid
ART98-SW4-OB	IAR & T, IBADAN	White	Flint	Hybrid
TZPB-OB	IAR & T, IBADAN	White	Flint	Hybrid
ILEI-OB	IAR & T, IBADAN	White	Flint	Hybrid
CML176	IITA/CIMMYT	White	Flint/ Dent	Inbred line
CML177	IITA/CIMMYT	White	Flint/ Dent	Inbred line
CML178	IITA/CIMMYT	White	Flint/ Dent	Inbred line
CML181	IITA/CIMMYT	White	Flint/ Dent	Inbred line
CML437	IITA/CIMMYT	White	Flint/ Dent	Inbred line
CML490	IITA/CIMMYT	White	Flint/ Dent	Inbred line
CML491	IITA/CIMMYT	White	Flint/ Dent	Inbred line
CML492	IITA/CIMMYT	White	Flint/ Dent	Inbred line
CML493	IITA/CIMMYT	White	Flint/ Dent	Inbred line
Oba-Super 1	LOCAL CHECK	White	Flint	Local variety
SUWAN-1-SR (DMR)	LOCAL CHECK	White	Flint	Local variety

Data collection

Data related to grain yield and yield characteristics were obtained at harvest. All ears harvested from each plot were weighed to determine grain yield per plot (assuming 80% shelling percentage), and was later converted to tonnes per hectare (t ha^{-1}) after adjusting to 15% moisture content. The sample of one thousand grain was collected from each plot at harvest for the determination of harvest moisture. The samples were first weighed to

obtain initial weight followed by drying to a constant weight in the oven at 80°C and the difference between the two weights recorded as moisture at harvest. Additional data collected on yield components were cob length (cm), cob diameter (cm), cob weight (g), kernel number per cob, kernel rows per cob, kernel row number, kernel depth (cm) and 1000 kernel weight (g). Kernel-rows per cob were counted from five cobs from each entry. The weights of sun dried 1000-grain samples drawn at random in each plot were recorded in grams at 15 per cent moisture content, with an electronic balance. Length of the ear was measured in centimeters (from the base to the tip of the cob) and recorded as cob length. Number of kernel rows per cob were counted and recorded. Number of kernels per row were counted and average values were recorded.

Laboratory analysis

The laboratory studies involved the determination of the approximate composition of QPM and the local maize genotypes. Five ears were randomly selected in each plot at harvest, followed by careful removal of the grains by hand. From each genotype, equal number of grains were selected from each plot, mixed together to form a balanced bulk. The grains obtained were ground to form a fine powder and each sample was oven dried to a constant weight at 80°C to obtain grain moisture content. Three replicate determinations were analyzed for each genotype and the mean recorded for each sample. Crude protein determination was estimated using standard micro-Kjeldahl procedure (AOAC 2006, 215-275).

The amino acid was determined using the procedure as described by Sentayehu (2008, 9-15). Maize flour samples of 0.5 gm were weighed in tarred scoop and transferred to boiling tubes. A selenium catalyst tablet was dropped into each tube and about 25ml of concentrated sulfuric acid was added. The tubes were then placed in an automatically controlled heater and set at 200°C. The mixtures were heated until the color changed to light blue. For samples which developed digest color of light brown or yellow, digestions were repeated twice or more times. Thirty mls of distilled water were added into the digestion tube. During this procedure, the organic matter of QPM flour oxidized, and the nitrogen in the protein was converted to ammonium by sulfuric acid as described by Aykroyd et al. (1964, 150) and Purseglove et al. 1968.

Ammonium in the digestion mixture was determined by distillation and titration (Aykroyd et al. 1964, 150). The digestion tube was placed onto the Tecator steam distillation apparatus. The distiller was set, the digestion tube inserted in the system and 150 mls ammonia were collected in the receiver flask containing 50 ml of boric acid solution at 4%. Then ammonia was titrated against a standard acid (0.1 N 10%HCl). Since 1 ml of 0.1 N HCl is equivalent to 1.401 mg N, the nitrogen content of the sample was calculated as follows.

$$\%N = \frac{(\text{ml HCl-ml black}) \times \text{normality} \times 14.007 \times 100}{\text{mg sample}} \quad (1)$$

$$\% \text{ Protein} = \%N \times 6.25$$

Due to the simplicity of the estimation of tryptophan, its content has been used as a criterion for screening materials with superior protein quality. For estimation of tryptophan in opaque-2 maize materials, the papain hydrolysis method was used (Hornandez et al. 1969).

A single step papain hydrolysis was utilized for protein solubilization. Iron ions oxidized acetic acid to glyoxylic acid in the presence of sulphuric acid. The indole ring of free tryptophan as well as that bound in soluble proteins reacted with glyoxylic acid and a violet-purple compound is produced. The intensity of the violet-purple color was measured at 545 nanometer a with spectrophotometer. By use of a standard curve of optical density vs. tryptophan concentration, percent tryptophan in sample was recorded as follows:

$$\% \text{ tryptophan in protein} = \frac{\% \text{ tryptophan in sample}}{\% \text{ protein in sample}} \quad (2)$$

The relationship observed by various researchers (Mertz et al. 1964, 145-279 Hornandez et al. 1969, Doll and Koie 1975, 55-59) between tryptophan and lysine in the maize endosperm protein, thus the tryptophan was used as a parameter for protein quality evaluation, and the value was increased 4 times to obtain the value of lysine (Sentayehu 2008, 9-15). The zein dry matter percentage was estimated by the formula suggested by Drochioiu et al. (2002, 47-61) given as:

% Zein in dry matter =
% crude protein x 0.386-2.22. (3)

Statistical analyses

Data collected from the field experiments and laboratory samples were statistically analyzed using PROC GLM model of SAS (SAS Institute 2007) to compute mean square for each parameter. Variety effect was considered statistically significant at $p<0.01$. Analysis of variance (ANOVA) on individual year basis was first computed before a combined ANOVA across years. Genotypic correlation coefficients were estimated from the mean squares and mean across products as suggested by Mode and Robinson 1959, 518-537. The correlation coefficients were partitioned into direct and indirect effects using the path coefficient analysis according to Dewey and Lu 1959, 515-518. Pertinent means were separated by the use of least significance difference (Steel et al. 1980). The degree of variation was estimated using percentage coefficient variation, all at $p<0.05$.

Results and Discussion

Biochemical analyses

Biochemical analyses of the QPM genotypes showed that the varietal effects of crude protein, zein dry matter, zein in crude form, lysine and tryptophan were significantly different among the QPM genotypes at $p<0.01$ (Table 2). Zein dry matter had the highest mean square value (1872.6) followed by crude protein (4.35), while lowest value of 0.072 was recorded for lysine. Obi (1982, 15-20) and Olakojo et al.(2007, 97-104) made similar observations that lysine content of some QPM genotypes varied significantly at $p< 0.05$. However, the QPM genotypes and the standard checks varied significantly from one another with respect to crude protein and other amino acids (Table 3). This probably suggests that high variability exists in maize genotypes with respect to these biochemical parameters. Therefore plant breeders could find this attribute useful in genetic manipulation and cultivar development for enhanced protein biochemical parameters. Crude protein and other amino acids were very high among the hybrids followed by inbreds, open pollinated varieties and the controls, in that order (Table 3). Mbuya et al. (2011, 317-327) reported 37% lysine content advantage over the normal maize varieties. Hybrid ART98-SW5-OB generally yielded the highest values of crude protein and amino acids compared with other genotypes. Hussain et al. (2006, 385-389) reported that while crude protein content in QPM hybrids is slightly improved, their lysine contents improved by 82 to 98% of the normal hybrids. The authors further observed that on the relative grain yield basis, the QPM hybrids have almost doubled their lysine content. Across the inbreds, crude protein ranged from 6.79% (CML493) to 8.23% (CML178), while in open pollinated varieties (OPVs), it ranged between 6.89% (Pool 19-SR) and 7.34% (EV8363-SR). The trend was comparable to that of zein with a range of 0.63% (ILEI-OB) to 0.82% (ART98-SW5-OB), 0.46% (CML491) to 0.66% (CML492) and 0.30% (EV8766-SR) to 0.63% (EV8363-SR) in the hybrids and inbreds and OPVs respectively. Percentage zein content of the two local checks, Oba-Super 1 (0.29) and SUWAN-1-SR (0.26) were the lowest compared to the other genotypes. Hybrid ART98-SW5-OB had a yield advantage of 60% for zein content over SUWAN-1-SR. Similarly, ART98-SW5-OB had the highest amount of crude zein ($mL\mu g^{-1}$) with yield advantages of 19, 24 and 32% over the best inbreds, OPVs and checks respectively.

Table 2: Mean square of crude protein and essential amino acids of 22 quality protein maize (*Zea mays* L.) genotypes and two local checks cultivated in Oke Oyi, Ilorin, Nigeria.

Source of variation	Crude protein %	Zein dry matter %	Zein crude ml/µg	Lysine %	Tryptophan %
Replicate	0.31	0.00045	3.99	0.0067	0.0018
Variety	4.35**	1872.6**	0.75**	0.072**	0.0007**
Error	0.09	0.68	0.005	0.005	0.00006

** Significant at $p<0.01$

Table 3: Means of crude protein and essential amino acids of 22 quality protein maize (*Zea mays* L.) genotypes and two local checks cultivated in Oke Oyi, Ilorin, Nigeria

Variety	Crude protein (%)	Zein dry matter %	Zein crude ml/µg	Lysine %	Tryptophan %
OPVs					
Obatanpa	7.21	0.56	140.13	3.53	0.67
EV8766-SR	7.10	0.30.	143.87	3.49	0.66
EV8363-SR	7.34	0.63	147.38	3.47	0.75
Pool 18-SR	7.23	0.45	141.42	3.56	0.70
Pool 19-SR	6.89	0.45	134.72	3.48	0.62
Hybrids					
Mama-ba	8.14	0.76	163.85	3.66	0.70
CIDA-ba	7.57	0.83.	159.67	3.59	0.71
Dada-ba	8.23	0.77	157.45	3.59	0.71
ART98-SW6-OB	6.34	0.68	194.78	3.68	0.78
ART98-SW5-OB	8.34	0.82	158.34	3.83	0.82
ART98-SW4-OB	8.33	0.81	159.,67	3.59	0.72
TZPB-OB	8.12	0.75	129. 78	3.67	0.67
ILEI-OB	6.02	0.63	126.,83	3.36	0.64
Inbreds					
CML176	7.34	0.65	156.93	3.18	0.60
CML177	7.57	0.55	148.83	3.16	0.79
CML178	8.23	0.56	151.78	3.27	0.63
CML181	7.34	0.60.	157.73	3.01	0.71
CML437	7.34	0.53	157.21	3.08	0.63
CML490	8.03	0.65	146.98	3.29	0.52
CML491	7.14	0.48	153.58	3.29	0.56
CML492	8.13	0.66	152.39	3.15	0.67
CML493	6.79	0.60.	132.65	3.26	0.46
Local checks					
Oba-Super 1	4.35	0.29	131.48	2.78	0.44
SUWAN-1-SR (DMR)	3.23	0.26	124.64	1.55	0.31
Mean	7.74	0.55	148.84	3.35	0.72
CV (%)	3.05	9.04	25.85	2.56	12.05
LSD (0.05)	0.89*	0.04*	1.97*	1.34*	0.04*

*, Significant F test at 0.05 levels of probability

Correlation coefficients analysis of biochemical parameters

Pearson correlation (r) among various biochemical parameters of the QPM genotypes in this trial revealed that crude protein was highly significantly positively correlated with tryptophan (r = 0.78**), but correlated negatively with zein dry matter, zein crude and lysine with coefficients of r = -0.58, -0.91** and 0.93** respectively (Table 4). Pixley and Bjarnason 1993, 1229-1234 earlier reported the need for monitoring protein content, tryptophan and lysine while breeding or selecting for QPM genotypes. Lysine was positive and highly significantly associated with tryptophan (r= 0.90**), zein dry matter (r= 0.71**) and zein crude (r= 0.79**). Similarly, zein dry matter was positive but not significantly correlated with zein crude. Tryptophan on the other hand was negatively correlated with zein dry matter and zein crude.

Table 4: Correlation coefficient (r) of amino acids and crude proteins of 22 quality protein maize (*Zea mays* L.) genotypes and two local checks cultivated in Oke Oyi, Ilorin, Nigeria

	Crude protein	**Lysine %**	**Tryptophan %**	**Zein dry matter %**	**Zein crude ml/µg**
Crude protein	-	-	-	-	-
Lysine %	-0.93**	-	-	-	-
Tryptophan %	0.78**	0.90**	-	-	-
Zein dry matter %	-0.58	0.71*	-0.72*	-	-
Zein crude ml/µg	-0.91**	0.79**	-0.31	0.32	-

*, ** Significant at < 0.05 < 0.01 level of probability, respectively

Table 5: Performance of 22 quality protein maize (*Zea mays* L.) genotypes and two local checks for grain yield (t ha^{-1}) evaluated in 2009-2011 cropping seasons in Oke Oyi, Ilorin, Nigeria

Genotype	**2009**	**2010**	**2011**
OPVs	(t ha^{-1})	(t ha^{-1})	(t ha^{-1})
Obatanpa	3.5	3.3	3.7
EV8766-SR	3.6	3.4	3.7
EV8363-SR	3.6	3.4	3.8
Pool 18-SR	3.7	3.7	3.9
Pool 19-SR	3.9	3.3	4.0
Hybrids			
Mama-ba	4.1	4.0	4.3
CIDA-ba	4.2	3.8	4.0
Dada-ba	4.8	4.4	4.9
ART98-SW6-OB	3.8	3.6	4.5
ART98-SW5-OB	5.0	4.5	5.1
ART98-SW4-OB	4.8	4.5	5.0
TZPB-OB	4.5	4.4	5.2
ILEI-OB	4.2	4.0	4.4
Inbred lines			
CML176	4.2	3.8	4.4
CML177	4.1	3.9	4.2
CML178	4.5	4.3	4.7
CML181	4.6	4.2	4.6
CML437	4.3	3.9	4.5
CML490	4.1	4.1	4.3
CML491	4.2	3.8	4.4
CML492	4.2	3.8	4.4
CML493	3.6	3.4	3.8
Local checks			
Oba-Super 1	3.8	3.4	3.9
SUWAN-1-SR (DMR)	3.6	3.0	3.8
Mean	4.0		
LSD (0.05)	0.45*		

Grain yield and related attributes

Grain yield in maize is a complex character, and is the result of correlation between yield and yield components and between yield components themselves. Therefore, it is imperative to examine the contribution of each of the various components in order to determine which one has the greatest influence on grain yield. This is a prerequisite to planning and evaluating meaningful breeding programs. Means for the interactive effect of genotype by year (G x Y) was significant for grain yield among the QPM genotypes and the standard checks (Table 5). The QPM genotypes and checks had the lowest grain yield in the year 2010 compared with 2009 and 2011. Rainfall distribution that was higher and consistent throughout the flowering/grain filling periods of July to September in 2009 and 2011, and a significant drop in rainfall in August, 2010 could contribute to significant differences among the genotypes for grain yield (Figure. 1). This relatively better rainfall pattern could favour accumulation and translocation of photo-assimilates in the genotypes with corresponding larger ear size and, subsequently higher grain yield (Bello et al. 2012, 52-61). This favorable condition is often encouraged by high moisture during the growing period (Akande and Lamidi 2006, 1744-1748 and Olakojo et al. 2007, 97-104). In the present study, the hybrids were superior for grain yield followed by inbreds, OPVs and local varieties irrespective of the cropping years. Hybrids ART98-SW5-OB, ART98-SW4-OB, TZPB-OB and Dada-ba had the highest grain yield of 5.0 t ha^{-1} with yield advantage of 10, 24 and 26% over the best inbreds, OPVs and checks respectively. The two local checks (Oba-Super 1 and SUWAN-1-SR) demonstrated instability of performance for grain yield with a difference of 14.7% and 21% yield loss in 2010 compared to 2009 and 2011 respectively. All the QPM genotypes had stable grain yield except CML493 (inbred) and Pool 19-SR (OPV). Hussain et al., (2006; 385-389) earlier reported that the hybrids had better yield potential than the local checks. The authors therefore suggested that QPM hybrids should be released to farmers for adoption not only for higher QPM grain yield but also superior protein quality and amino acids contents.

Grain yield was significantly higher in 2009 and 2011 by 0.8 t ha^{-1} compared to 2010, representing 22.2% yield increase (Table 6). The differences in performance among the genotypes for this character indicate variability which could be heritable and can be exploited in the overall process of selection in breeding programs. Similar results were reported by Ahmad et al. 2000, 2098-2100, and Hussein et al. (2011, 626-628) who evaluated and identified high yielding maize varieties among different genotypes tested. McCutcheon et al. 2001; 54-56 and Akbar et al. (2009; 1817-1829) also reported significant differences among maize cultivars for grain yield. The hybrids ranked best for grain yield in this study followed by inbred lines, OPVs and standard checks. The range was from 4.0-4.7, 4.0-4.4 and 3. 4-3.7 t ha^{-1} of the hybrids, inbreds and OPVS values respectively. It is worthy to note that most of the hybrids and some inbred lines out-yielded the OPVs and local checks with yield advantage of about 38% over the best OPVs and local varieties. The local varieties as the poorest yielders may be due to genetic composition of the genotypes. It has also been reported by many researchers that QPM varieties widely grown in many African countries produce higher grain yield compared with the currently released normal maize varieties for most grain yield characters (Vasal 2000, 445-450; Sallah et al. 2004, 95-104 and Olakojo et al. 2007, 97-104). Among the genotypes tested, highest grain yield of 4.7 t ha^{-1} from ART98-SW5-OB was recorded within the hybrids, 4.4 t ha^{-1} (CML178 and CML181), 3.7 t ha^{-1} in the OPVs, and 3.3 t ha^{-1} (SUWAN-1-SR (DMR)) among the checks. However, hybrid ART98-SW5-OB which ranked first for grain yield out-yielded the OPV checks by more than 44%. The genotypes also differed significantly for yield contributing characteristics among all the genotypes with inbred lines ranked the highest followed by hybrids, OPVs and the local checks on average values. The cob weight ranged from 39-48 gm in the inbreds, and between 39 and 45 g in both hybrids and OPVs. The checks also had the least cob weight range of 39-40 g.

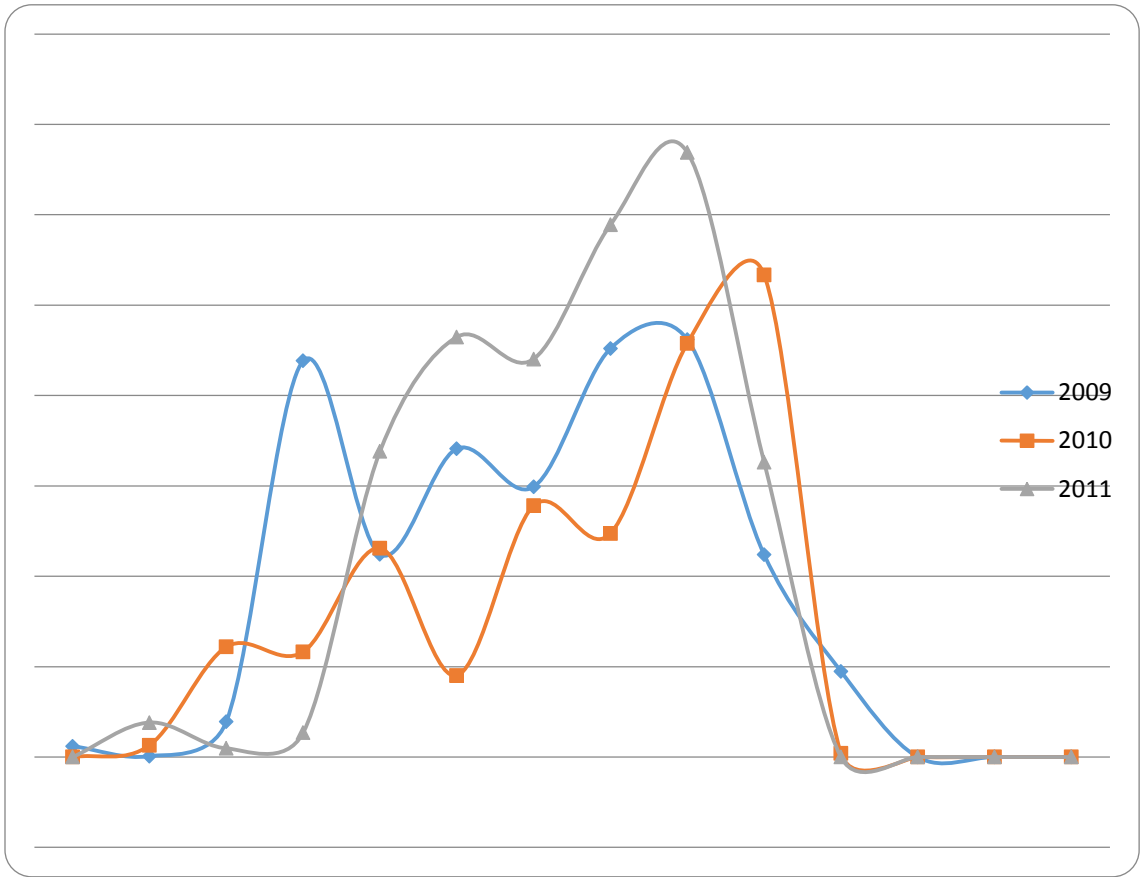

Figure 1: Monthly rainfall distribution (mm) pattern (3 years) at Oke Oyi, Ilorin from 2009 to 2011

Source: Meteorological Department of Lower Niger River Basin Development Authority Ilorin, Nigeria

Correlation coefficients analysis of grain characteristics

Genotypic correlation coefficients between grain characteristics showed that kernel number per cob has the most positive correlation (r=0.671**) with grain yield followed by kernel rows per cob (r = 0.556**), cob diameter (r=0.543**) and cob weight (r=0.452**) (Table 7). Crude protein content recorded negative non-significant association with all the traits studied including grain yield except cob weight and kernel rows per cob. The correlation between 1000-kernel weight and cob weight was positive and significant (r=0.571**). It seems that by increasing cob weight due to more accumulation of photo-assimilates, the greatest portion of assimilates were relocated to grains, so that grain weight increased. Number of kernels per row was positively correlated with grain yield. One thousand kernel weight that was positively correlated with grain yield confirmed the findings of Dwivedi et al. 1997, 175-177; Jin and Wang 1997, 23-26; Gautam et al. 1999, 1169-1171; Prakash et al.2006, 91-98 and Golam et al. 2011, 6147-6154. Khayatnezhad et al. 2010; 96-99 also reported that 500-grain weight had the most highest correlation with grain yield. Kernel rows per cob and kernel number per cob that were positively correlated with grain yield was also confirmed by Saha 1985, 240-246 and You et al. 1998, 3-4.

Table 6: Performance of 22 quality protein maize (*Zea mays* Li.) genotypes and two local checks for grain yield characteristics evaluated in 2009-2011 cropping seasons in Oke Oyi, Ilorin, Nigeria

Genotype	Grain Yield (t/ha)	Kernel number per cob (no)	Kernel number per row (no)	Kernel rows per cob (no)	Kernel depth (cm)	Cob length (cm)	Cob diameter (cm)	Cob weight (g)	1000 kernel weight (g)
OPVs									
Obatanpa	3.5	300	23	18	18.5	6.3	15.3	0.45	295.5
EV8766-SR	3.6	302	21	17	20.3	5.9	16.4	0.39	282.4
EV8363-SR	3.6	312	22	19	19.7	6.0	15.2	0.40	297.4
Pool 18-SR	3.8	298	20	20	16.8	5.8	16.1	0.42	267.4
Pool 19-SR	3.7	302	19	16	19.8	6.2	16.0	0.39	298.6
Hybrids									
Mama-ba	4.1	299	25	18	19.4	6.0	16.0	0.44	289.6
CIDA-ba	4.0	306	24	19	21.5	5.7	15.9	0.45	296.2
Dada-ba	4.7	301	25	17	18.9	5.9	16.2	0.44	289.1
ART98-SW6-OB	4.0	304	23	17	17.7	6.1	15.9	0.45	299.5
ART98-SW5-OB	4.9	307	26	18	16.8	6.3	15.3	0.39	278.4
ART98-SW4-OB	4.8	304	23	17	19.8	6.2	16.3	0.41	287.6
TZPB-OB	4.7	300	25	19	19.7	5.8	15.5	0.45	297.4
ILEI-OB	4.1	301	24	19	17.4	6.1	16.1	0.39	295.6
Inbred lines									
CML176	4.1	300	19	20	19.5	5.7	15.4	0.39	289.9
CML177	4.1	299	23	16	19.4	6.1	16.1	0.38	296.9
CML178	4.5	302	25	17	16.8	5.9	15.7	0.43	300.3
CML181	4.5	298	24	18	17.7	6.0	16.0	0.45	275.0
CML437	4.2	301	20	19	17.7	5.9	15.8	0.39	302.4
CML490	4.2	304	23	19	17.4	6.2	15.9	0.40	297.6
CML491	4.2	297	22	17	17.5	5.9	15.4	0.43	267.0
CML492	4.2	303	23	16	16.5	5.9	15.2	0.39	298.6
CML493	3.6	302	24	17	19.7	6.0	15.7	0.48	300.9
Local checks									
Oba-Super 1	3.7	278	19	19	18.8	6.2	14.7	0.39	286.2
SUWAN-1-SR(DMR)	3.5	262	18	19	19.2	5.8	15.4	0.40	256.3
Mean	4.1	299	23	18	18.6	6.0	15.7	0.42	289.4
CV %	11.6	2.05	0.11	5.73	3.04	0.07	0.01	11.98	0.11
LSD (0.05)	0.45*	10.04*	3.07*	1.26*	1.11*	0.09*	0.94*	1.03*	12.24*

*, Significant F test at 0.05 level of probability

Number of kernels per row and kernel rows per cob that have positive genetic correlations with grain yield in this study were also observed by previous workers (Wang et al. 1999, 211-222; Yousuf et al. 2001, 387-388 Liu 2009, 13-15 and Kashiani et al. 2010, 78-84). The high correlation of grain yield with the number of rows per cob recorded in this study was also reported by other researchers (Mohammadi et al. 2003, 1690-1697). A highly significant and positive correlation between grain yield and cob length was also noticed by Rafiq et al. 2010, 35-38. In this study the correlation between grain yield and kernel depth was not significant. This suggests that cob length is a more important yield component than kernel depth in contributing to final grain yield in maize. Correlation of grain yield with grain number per row noticed in this study was also reported by Marefatzadeh and Hamidi 2010, 1-6 and Khodarahmpour et al. 2012, 3099-

3105. The present study agreed with the results of significant correlations between yield and number of rows per cob, number of kernel per row and 1000-grain weight, number of kernel per row and number of rows per cob observed by earlier researchers (Viola et al. 2003, 22-25; Ross et al. 2006, 301-313; Khazaei et al. 2010, 14-19; Rafiq et al. 2010, 35-38; Orlyan et al. 1999, 9-12; Zirehzadeh et al. 2011, 853-857) also suggested that most important traits influencing grain yield are number of kernel per row. Cob length that showed positive and significant correlation with kernel number per row in this study indicates that increase in cob length will increase the kernel number per row. Ultimately as a result of increased kernel number per row, kernel number per cob will also increase, thereby increasing the grain yield. A positive correlation between cob length and cob weight recorded in this study is in accordance with Amin et al. 2003; 181-190 and Abou-Deif 2007, 86-90. Results of this study therefore showed that, kernel number per cob, kernel number per row, kernel row per cob, kernel depth, cob weight, cob diameter, cob length and 1000-kernel weight could be used as important traits for prediction of grain yield. This finding was also in agreement with the results of earlier researchers (Choucan et al. 2007, 543-562; Jafari et al. 2009, 10-17; Bello et al. 2010, 2633-2639; Golbashy et al. 2010, 2714-2719; Beiragi et al. 2011, 32-37). Actually, yield components have effects on each other in positive ways, which may be due to the same genes controlling these traits.

Table 7: Genotypic correlation coefficients between grain yield, other yield characteristics and crude protein of 22 quality protein maize (*Zea mays* L.) genotypes and two local checks evaluated in 2009-2011 cropping seasons in Oke Oyi, Ilorin, Nigeria

Parameters	**Grain yield (t/ha)**	**Kernel number per cob (no)**	**Kernel Number per row (no)**	**Kernel rows per cob (no)**	**Kernel depth (cm)**	**Cob length (cm**	**Cob diameter (cm)**	**Cob weight (g)**	**1000 kernel weight (g)**	**Crude protein %**
Grain Yield (t/ha)	-									
Kernel number per cob (no)	0.671**	-								
Kernel number per row (no)	0.124	0.798**	-							
Kernel rows per cob (no)	0.556**	0.673**	-0.157	-						
Kernel depth (cm)	0.065	-0.541**	-0.413	-0.174	-					
Cob length (cm)	0.235	0.765**	0.641**	0.007	-0.089	-				
Cob diameter (cm)	0.543**	0.114	-0.312	0.651**	0.276	0.121	-			
Cob weight (g)	0.452**	0.032	-0.231	0.63*	0.431**	0.234	0.879**	-		
1000 ernel weight (g)	0.067	-0.53**	-0.653**	0.177	0.751**	-0.125	0.567**	0.571**	-	
Crude protein %	-0.031	-0.0086	-0.064	0.091	-0.0097	-0.098	-0.0076	0.056	-0.076	-

*, ** Significant at < 0.05 and < 0.01 level of probability, respectively

Table 8: Path coefficient analysis showing the direct and indirect effects of grain yield characteristics and crude protein of 22 quality protein maize (*Zea mays* L.) genotypes and two local checks evaluated in 2009-2011 cropping seasons in Oke Oyi, Ilorin, Nigeria

Trait	Kernel number per cob (no)	Kernel number per row (no)	Kernel Rows per cob (no)	Kernel depth (cm)	Cob length (cm)	Cob diameter (cm)	Cob weight (g)	1000 kernel weight (g)	Crude protein %
Kernel number per cob (no)	-5.142	-4.366	-1.957	-2.312	-4.952	-4.584	-3.541	-2.871	0.076
Kernel number per row (no)	-2.758	-4.641	0.095	-1.461	-3.675	-1.572	-2.712	-1.978	0.175
Kernel rows per cob (no)	0.579	-0.597	3.154	-0.357	-0.654	0.414	-0.571	-0.487	0.265
Kernel depth (cm)	-0.672	-0.076	-0.045	-0.56	-5.842	-0.561	-0.095	-0.047	-0.431
Cob length (cm)	11.076	12.432	-3.175	-0.169	13.067	-0.671	-0.573	10.112	-2.672
Cob diameter (cm)	-0.842	-0.712	-0.153	-1.831	0.879	-1.087	-1.946	-0.895	0.198
Cob weight (g)	6.573	8.679	-2.679	-0.352	10.071	-0.345	-0.572	9.451	-0.045
1000 kernel weight (g)	-2.637	-1.963	0.632	-1.739	-3.452	-3.453	-4.621	-4.105	0.117
Crude protein %	-0.009	-0.065	0.085	-0.038	-0.075	-0.214	-0.456	-0.045	0.823
Genotypic correlation coefficient	0.046	0.745	0.387	0.078	0.163	0.543	0.034	0.456	0.084

Path analysis of grain characteristics

Since, the significance of simple correlation among yield parameters cannot give enough reasons for cause/effect phenomena, path coefficient analysis for determination of direct and indirect effects is essential (Bello et al. 2010, 2633-2639). This is because the characteristics that are interrelated do not exist by themselves, but are linked to other yield attributes. The path coefficient analysis not only specifies the effective measure of direct and indirect causes of association, but also depicts the relative importance of each factor involved in contributing to the final product of yield. In the present study, cob length has a maximum positive direct effect on grain yield (13.067) followed by kernel rows per cob (3.154) and crude protein (0.823) (Table 8). Positive and highly significant direct effect of cob length for grain yield was reported by other workers (Nemati et al. 2009, 194-198; Selvaraj et al. 2011, 209-223; Selvaraj et al 2011, 209-223) also noticed a favourable influence of kernel rows per cob on grain yield. Direct effect of crude protein on grain yield was reported by Baheeruddin et al. 1999, 85-89. In this study, kernel number per cob, kernel number per row, kernel depth, cob diameter, cob weight and 1000 kernel weight recorded negative direct effects on grain yield even though genotypic correlation coefficients on grain yield were positive, as previously reported by Manivannan (1998, 293-294), and Selvaraj et al (2011, 209-223) (Table 6). The indirect effect of kernel number per row through kernel rows per cob and crude protein content were positive, and through kernel number per cob, kernel number per row, kernel depth, cob length, cob diameter, cob weight and 1000 kernel weight were negative (Table 7). Negative indirect effect of kernel number per cob on grain yield was noticed on all the characteristics studied except via crude protein. Positive indirect effect of cob length on grain yield was noticed for kernel number per cob, kernel number per row and 1000

kernel weight but it was negative through kernel rows per cob, kernel depth, cob diameter, cob weight and crude protein. A highly significant and positive direct effect of cob length on grain yield was indicated by previous researchers Nemati et al. (2009, 194-198) and Selvaraj et al. (2011, 209-223). Cob length due to involvement of direct and indirect effects through total kernel number has high correlation with kernel yield in this study. Ross et al. (2006, 301-313) earlier suggested that cob length is a component effecting kernel yield, therefore, genetically controlled recognition of ear length and additional traits correlate to kernel yield can be of help to hereditary and diversity recognition of kernel yield. The authors further stressed that kernel numbers per ear have the best direct effect on kernel yield, therefore maize breeders must give importance to kernel and row number as selection indicators in yield breeding. The authors also suggested that kernel numbers per cob is the best trait affecting kernel yield. In the present study, positive indirect effects of cob weight on grain yield were recorded through kernel number per cob, kernel number per row, cob length and 1000 kernel weight; it was negative through the remaining parameters. However the indirect influence of crude protein on grain yield via kernel rows per cob was only positive, though other parameters were negative.

CONCLUSION

From the present study, QPM genotypes evaluated were generally of considerable lysine and tryptophan contents. The QPM hybrids seem superior for the essential amino acid contents followed by inbreds and OPVs, with the local checks being most inferior. This phenomenon is of the same trend for grain yield. The best QPM hybrids for grain yield (Dada-ba, ART98-SW5-OB, ART98-SW4-OB and TZPB-OB) had percentage lysine and tryptophan advantage of 34% compared with the local checks. Similarly, these hybrids out-yielded other genotypes with yield advantage of 10, 24 and 26% over the best inbreds, OPVs and checks respectively. Most of the QPM hybrids and inbred lines evaluated have superior performance for grain yield compared with open pollinated and local varieties. The QPM hybrids that combined high yield performance and essential amino acids (especially hybrids Dada-ba, ART98-SW5-OB, ART98-SW4-OB and TZPB-OB) could be assessed in different agro-ecologies to identify those that could be released to farmers in each environment. The inbreds (especially CML 178 and CML 181) that were superior for grain yield, lysine and tryptophan contents could also be introgressed for further breeding programs. However, correlation studies showed that grain yield had a positive association with all the yield attributes except crude protein content which was negatively correlated. Kernel number per cob had maximum correlation with grain yield followed by kernel rows per cob, cob diameter and cob weight. Direct selection of these characteristics might be rewarding for yield improvement since they revealed the true relationships with grain yield. The direct effect for crude protein was positive but the correlation was negative; in such a situation direct selection for this parameter should be practiced to reduce the undesirable indirect effects.

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