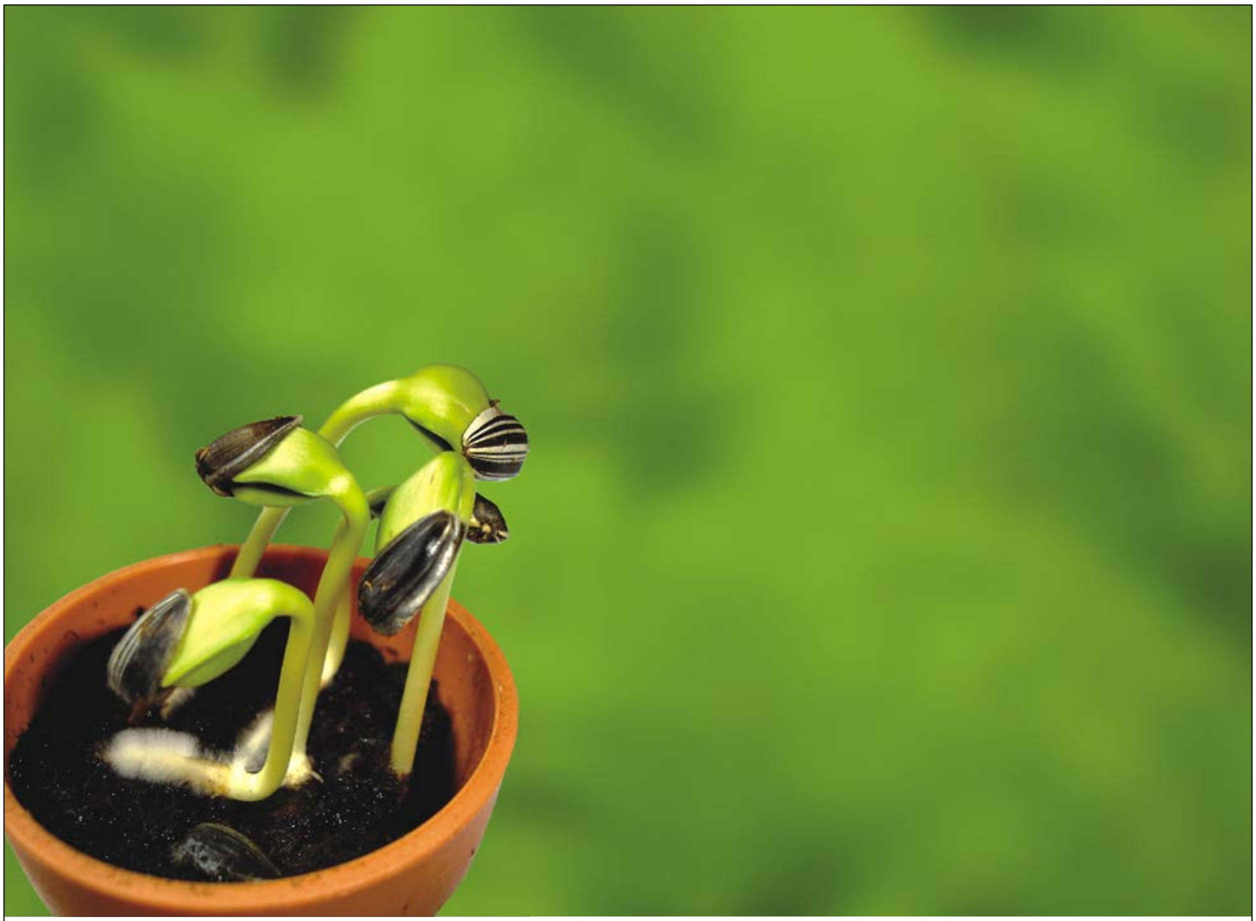

International Journal of
Plant Breeding and Genetics

ISSN 1819-3595

Academic
Journals Inc.

www.academicjournals.com

International Journal of Plant Breeding and Genetics 5 (4): 317-327, 2011
ISSN 1819-3595 / DOI: 10.3923/ijpbg.2011.317.327

Nutritional Analysis of Quality Protein Maize Varieties Selected for Agronomic Characteristics in a Breeding Program

[1,2]K. Mbuya, [4]K.K. Nkongolo and [1,3]A. Kalonji-Mbuyi
[1]Faculty of Agronomy, University of Kinshasa, P.O. Box 117, Kinshasa 11, RD-Congo
[2]National Institute for Research and Agronomic Studies (INERA), P.O. Box 2037, Kinshasa 1, RD-Congo
[3]Regional Nuclear Energy Center, Kinshasa (CRENK), P.O. Box 868, Kinshasa XI, RD-Congo
[4]Department of Biological Sciences, Laurentian University, Sudbury, Ontario, P3E 3C6, Canada

Corresponding Author: K.K. Nkongolo, Department of Biological Sciences, Laurentian University, Sudbury, Ontario, P3E 3C6, Canada

ABSTRACT

Nutritional characteristics of QPM genotypes that have been released are limited. A breeding program has been initiated in 2008 in DR-Congo using varieties selected from several agro-ecological regions. The objective of the present study was to establish a nutritional profile including amino acid and carotenoid content of selected QPM from the DR-Congo breeding program. Six Quality Protein Maize (QPM) and seven normal maize varieties were evaluated for agronomic characteristics and disease reaction. The grain amino acid and carotenoid concentrations were evaluated. The impact of QPM diet on chick's weight was also determined. QPM Longe 5 produced the highest grain yield in several trials in famer's fields. Lysine content of QPM-SR-SYNTH and QPM Longe 5 showed significant increase of 33 and 37%, respectively, over the other maize varieties. There was a 50% increase in tryptophan in QPM Longe 5 compared to normal maize varieties. More importantly, the two QPM varieties provide proteins with a better amino acid balance than the normal maize varieties. The level of the carotenoids analyzed was significantly higher in the maize MUS-1 variety with yellow endosperm compared to all the genotypes with white endosperm. The total carotenoid content in MUS-1 was over 250 fold compared to QPM and other maize varieties. The use of QPM in poultry resulted in a 50% increase in body weight compared to normal maize over a 9 week-period. A breeding program combining the benefits of QPM and the high level of carotenoid of yellow maize endosperm should produce superior QPM varieties for human and animal nutrition.

Key words: Participatory breeding, amino acid profile, carotenoid, weight gain, DR-Congo

INTRODUCTION

Maize is a major cereal crop for human nutrition, worldwide. Several million people, particularly in the developing countries, derive their protein and calorie requirements from maize. In many developing countries of Latin America, Africa and Asia, maize is the major staple food and often the only source of protein because animal protein is scarce and expensive and consequently, unavailable to a vast sector of the population (Vasal, 2000). One of the main nutritional limitations of normal maize grain is its poor nutritional profile because of a deficiency in essential amino acids such as lysine, tryptophan and methionine (Bantte and Prasanna, 2003; Huang *et al.*, 2006). This has been attributed to a relatively higher proportion of prolamines in maize storage proteins which are essentially devoid of lysine and tryptophan. In fact, normal maize varieties proteins contain on an average about 2% lysine which is less than one-half of the concentration recommended for human nutrition by the Food and Agriculture Organization (FAO). In addition, methionine is the third limiting amino acid in normal maize used in non-ruminant diets after lysine and tryptophan and it is the first limiting amino acid in the legumes (Scott *et al.*, 2004). Thus, even in a complete diet often consisting of maize and legume, methionine levels can be nutritionally limiting. Quality Protein Maize (QPM) varieties developed by the international maize and wheat improvement (CIMMYT) in the late 1990's produces 70 to 100% more lysine and tryptophan and yields more grain than the most modern varieties of tropical maize (Bjarnason and Vasal, 1992).

Presently, more than 23 countries in developing countries have released QPM varieties for large-scale cultivation on area over 2.5 million ha. In sub-Saharan Africa, 17 countries are growing QPM on around 200,000 ha with Ghana alone accounting for about 70,000 ha, Obatanpa being the major cultivar (Akande and Lamidi, 2006; Sofi *et al.*, 2009). Most of the QPM varieties developed in Africa have been agronomically characterized and their adaptation to targeted areas is well documented. But nutritional characteristics of these varieties are lacking because of a high cost for protein and amino acid analyses. The impact of QPM in animal and human nutrition has been conducted in several countries in poultry, pigs and children. In DR-Congo, evaluation of introduced QPM varieties was initiated recently in 2008 with farmer participation. This first round of participatory selection resulted in the identification of few elite QPM adapted to local agro-ecological conditions (Mbuya *et al.*, 2010).

The main objectives of this study were (1) to establish a nutritional profile of elite QPM varieties from the DR-Congo breeding program and (2) to select for local release the best QPM variety based on agronomic, nutritional and organoleptic values.

MATERIALS AND METHODS

Field trials: Six-Quality Protein Maize (QPM) and seven normal maize varieties were evaluated for agronomic characteristics and disease reaction in Gandajika in 2008-2009. Gandajika is in the Eastern Kasaï province of DR-Congo with latitude 6° 45' S, longitude 23° 57' E and altitude 780 m.

The source and year of introduction of each variety to DR-Congo are described in Table 1. The community plot was ploughed and ridged at a spacing of 0.75×0.5 m. Gross plot size (experimental unit) was 5 m long and 3 m wide. Two seeds were planted per stand and later thinned to one two weeks after seedling emergence to provide a uniform plant population of 53,333 plant/ha. Manual weeding was carried out as to keep the field clean. The trial was a Completely Randomized Block Design (RCBD) with three replicates at two sites. The fertilizer treatment consisted of the application of 140 kg per ha of NPK (17-17-17) at 14 days after planting followed by 20 kg per ha of nitrogen (urea fertilizer) application 25 to 30 days later. This represents half of the recommended dose of mineral fertilization in the region.

The following data were collected from the two middle rows of each plot: Cob length (in cm), cob size (in cm), kernel rows per cob, kernels per rows and cob aspect using ratings 1-5, where 1 = excellent, 2 = very good, 3 = good, 4 = fair and 5 = poor. At harvest, ears from the middle two rows were harvested together, shelled and grain yield per plot was determined at 14% moisture content from which grain yield per ha was estimated. Grain yield was adjusted to 83% shelling recovery from the de-husked cob weight per plot. Farmers also assessed corn flour quality for local dishes (fufu).

Table 1: Origin, year of introduction and types of the 14 maize varieties evaluated with farmers in Gandajika, RD-Congo

Varieties	Origin/provider	Year of introduction	Type	Category
ECA-QVE 3	CIMMYT-Kenya	2008	QPM	OP*
ECA-QVE 4	CIMMYT-Kenya	2008	QPM	OP
ECA-QVE 6	CIMMYT-Kenya	2008	QPM	OP
DMR-ESR-W**	IITA-Ibadan	1994	NORMAL	OP
AK9331-DMR-ESR-Y	IITA-Ibadan	1994	NORMAL	OP
QPM-SR-SYNTH***	CIMMYT-Kenya	2008	QPM	OP
SUSUMA	CIMMYT-Kenya	2008	QPM	OP
LONGE 5***	NARI-Uganda	2008	QPM	OP
SALONGO-2**	INERA-Gandajika	1976	NORMAL	OP
KASAI-1**	INERA-Gandajika	1976	NORMAL	OP
MUS-1**	INERA-Gandajika	1996	NORMAL	OP
GPS-5	INEAC-Gandajika	-	NORMAL	OP
LOCALE 1	Farmers	-	NORMAL	OP

*OP: Open pollinated varieties, **: Selected improved normal maize varieties, ***: Selected Elite Quality protein maize varieties, CIMMYT: International Maize and Wheat Improvement Center, INERA: National Institute of Agronomic Research and Studies (RD-Congo), INEAC: National Institute of Agronomic Studies (Belgium Congo), IITA: International Institute of Tropical Agriculture NARI: Namulonge Agriculture Research Institute

In 2009-2010, six QPM were evaluated in farmer's field using the recommended fertilization dose: 280 kg per ha of NPK (17-17-17) at 14 days after planting followed by 40 kg per ha of nitrogen (urea fertilizer) application 25 to 30 days later. The agronomic assessment was based on plant aspect, cob aspect and grain yield. In addition, the QPM were observed for the natural symptoms of two main local diseases, Maize Streak Virus (MSV) and Downy mildew (caused by *Perosclerospora (sclerospora) sorghi*). Severity of each of MSV was scored using a rating 1- 5 where, 1 = no symptom, 2 = slight infection, 3 = moderate infection, 4 = severe infection and 5 = very severe. For Down mildew, only the number of affected plants per plot was recorded.

Collected data were subjected to two-way Analysis of Variance (ANOVA) using Genstat discovery version 3 and the least significant differences among means were calculated to identify differences among specific treatments.

Nutritional analysis: Amino acid analyses were conducted at the University of Missouri Agricultural Experiment Station Chemical Laboratories (ESCL). Only the two QPM varieties selected for agronomic characteristics along with four normal maize varieties were analyzed in triplicates. They include QPM Longe 5, QPM-SR-SYNTH, DMR-ESR-W, Salongo 2, MUS-1 and locale-1. The cowpea accession MW 308 was used as a legume reference. The grain amino acid concentration was evaluated using AOC standard method (Method 982.30 E (a, b, c), AOAC, 2006). Crude protein was determined by combination analysis (Method 990.03, AOAC, 2006) using the formula crude protein = N x 6.25. ANOVA (two-way) was used to identify significant variation for each amino acid and crude protein.

The Carotenoid analysis was conducted in Dr. M. Ferruzzi laboratory at Purdue University (Indiana, USA). For this analysis, 12 varieties (6 QPM and 6 normal varieties) included in the initial field trial were considered. Like with amino acid analysis, each sample was analyzed in triplicate. The four carotenoid compound lutein, zeaxanthin and B-cryptoxanthin and B-carotene were determined using High Pressure Chromatography (HPLC). Details of the procedure are described by Kean *et al*. (2007, 2008).

Impact of QPM on chick's grain weight: To assess the impact of QPM in poultry, 750 g of QPM flour were mixed with grinded amaranth leaves and put in feeding troughs, while 500 mg of tetracycline antibiotic and calcium and 400 mg of Vitamin A tablets were put in drinkers in 1 L of water each week. This mixture represented a ration for 5 chicks. When chicks were small, this ration (750 g) was for 4 days and as they were growing it was enough for only 1 day. Five other chicks were fed with the same mixture but 750 g of yellow normal maize was used instead of QPM flour. The ration was replaced every day. Weight gain was recorded once a week for 9 weeks. The data were analyzed using the t-test.

RESULTS AND DISCUSSION

Field trials: A breeding program aimed at selecting and developing QPM adapted to DR-Congo maize growing area has been developed. Significant variations for cob and rachis size, cob length and grain yield were observed (Table 2). Results of 2008-2009 trials revealed that out of 13 genotypes evaluated from 2008 to 2010 including 6 QPM and 7 normal maize, the QPM genotype the QPM-SR-SYNTH and the normal variety AK9331-DMR-ESR-Y produced the highest yield. But subsequent trials in farmer's field at several locations indicated the superiority of QPM Longe 5 over all the normal and QPM varieties (data not shown). In fact, in the 2009-2010 trial of six selected QPM in farmer's fields under chemical fertilization, the genotype QPM Longe 5 produced 5208 kg ha^{-1} followed by QPM-SR-SYNTH with 3811 kg ha^{-1} (Table 3). These data were significantly higher than the grain yield of other tested varieties. The moderate grain yield of the elite QPM Longe 5 in the 2008-2009 was ascribed to a lower germination rate of the seed lot used. Overall, QPM Longe 5 had higher mean grain yield performance over different agro-ecological areas in both rich and poor environments.

There was no significant statistical difference among QPM varieties for maize streak virus ratings (Table 3). With the exception of Susuma, a QPM genotype that showed a higher number of mildew-affected plants per plot, the incidence of mildew was low for all the varieties tested. This

Table 2: Average agronomic performance of 13 QPM and normal maize varieties evaluated under half dose of recommended fertilization regime (140 kg per ha of NPK - 17-17-17 and 20 kg of urea) in community plots in Gandajika, RD-Congo

Varieties	Cob Aspect* 1 - 5	Kernel rows per cob (No.)	Cob size/ diameter (cm)	Cob rachis size (cm)	Cob length (cm)	Adjusted grain yield* (t ha^{-1})	Flour quality* 1-5
ECA-QVE 3	1.8	14.7	4.3	2.6	15.1	3.1	1.5
ECA-QVE 4	1.7	14.7	4.3	2.4	13.7	3.8	1.5
ECA-QVE 6	1.7	14.0	4.3	2.5	13.5	3.2	1.5
DMR-ESR-W	1.7	13.3	3.8	2.1	13.7	2.9	1.5
AK9331-DMR-ESR-Y	1.5	14.0	4.3	2.4	15.0	4.8	1.5
QPM-SR-SYNTH	1.7	14.0	4.3	2.5	16.6	4.2	1.5
SUSUMA	1.5	14.7	4.6	2.4	15.3	3.7	1.0
LONGE 5	1.7	14.0	4.3	2.7	14.8	3.1	1.0
SALONGO-2	1.8	14.7	4.4	2.5	15.0	3.5	1.5
KASAI-1	1.8	16.0	4.6	2.5	15.2	3.4	1.5
MUS-1	1.8	14.7	4.3	2.7	15.1	3.8	1.0
GPS-5	2.2	12.0	3.8	2.2	15.1	2.2	1.5
LOCALE	1.8	10.7	3.7	1.4	14.0	3.5	1.5
LSD (0.05)	0.6	1.4	0.3	0.3	1.1	0.4	-

*1 = best and 5 = worst

Table 3: Average agronomic performance of 6 QPM evaluated under full dose of recommended fertilization regime (280 kg per ha of NPK -17-17-17, and 40 kg of urea) in community plots in Gandajika, RD-Congo in 2009 -2010

Varieties	Plant height (cm)	Plant aspect (1- 5)	Cob aspect (1- 5)	Grain yield (kg ha^{-1})	Mildew infection No. of affected plants/plot	Reaction to MSV* (1 - 5)
LONGE 5	157	2.2	1.8	5208	2	1.3
QPM-SR-SYNTH	147	3.0	2.5	3811	4	1.7
ECA-QVE 3	133	3.0	2.3	3120	4	1.3
ECA-QV4	129	2.8	2.7	3388	4	1.3
ECA-QV6	132	3.0	2.5	3470	5	1.2
SUSUMA	155	2.7	2.2	3225	7	1.3
LSD (0.05)	19.6	0.9	0.7	1048	3.5	0.5

*MSV = Maize streak virus

was consistent with data reported by Mbuya *et al.* (2010). QPM varieties, Longe 5 and Susuma produced flour that was rated of higher quality for local dish "fufu". In fact, the taste of "fufu" prepared with QPM Longe 5 flour was judged by farmers and other consumers as similar to that of MUS 1, a genetically improved maize variety released in the region. Based on agronomic characteristics QPM Longe 5 and QPM-SR-SYNTH were selected for consideration for local release. Considering that the two selected QPM (QPM-SR-SYNTH and QPM Longe 5) are genetically closely related based on recent molecular analysis (unpublished data), only QPM Longe 5 will be considered for release in 2011. Like in most QPM breeding programs the nutritional superiority of QPM varieties developed and released in Africa has not been documented.

Amino acid analysis: The present study was designed to quantitatively measure and to compare the levels and variation of carotenoids, total protein as well as the individual amino acids in six QPM and 7 normal maize varieties. The overall amino acid composition of the maize varieties and the levels of statistical significance obtained from analysis of variance are shown in Table 4. Lysine content of QPM-SR-SYNTH and QPM Longe 5 showed significant increase of 33 and 37%, over the normal maize varieties, respectively (Table 4). The levels of lysine in QPM-SR-SYNTH and QPM Longe 5 were 3.6 and 4.2 g of lysine/100 g of protein, respectively. The data are in accord with the lysine values reported by Kniep and Mason (1991) and Zarkadas *et al.* (2000). These authors reported the lysine values ranging from 4.1 to 4.3% and 4.4 to 4.6, respectively.

In several reports, lysine levels have been associated with tryptophan levels. In the present study, there was a 50% increase in tryptophan in QPM Longe 5 compared to normal maize varieties. But the level of tryptophan in QPM-SR-SYNTH was equal to that found in the normal maize varieties MUS-1, Salongo 2 and Kasai -1.

The other potentially limiting amino acids are threonine, isoleucine and methionine. Threonine level was the same in QPM and normal varieties. Evaluation of methionine content showed a decrease of 28 and 61% for QPM-SR-SYNTH and QPM Longe 5, respectively over the 5 normal maize analyzed. These results are consistent with previous studies that have shown that QPM have lower methionine levels than normal maize varieties (Eggum *et al.*, 1979; Scott *et al.*, 2004). It should be pointed out that methionine is the third limiting amino acid in normal maize after lysine and tryptophan and it is the first limiting amino acid in legumes (Scott *et al.*, 2004). Reduction levels for leucine were 22 and 65%, for QPM - SR - SYNTH and QPM Longe 5 compared to normal varieties. This significant reduction in methionine and leucine in part balances the increase in other amino acids.

Table 4: Total protein and essential amino acid content in quality protein maize (QPM) and normal maize varieties from a RD-Congo breeding program

	Corn varieties						Cowpea	
	Locale-1*	QPM-SR-SYNTH*	Salongo 2*	QPM Longe 5	MUS-1	DMR-ESR-W	MW 387	
Eddential AA	------------------(w/w %)------------------							LSD
Taurine	0.04 (0.3)	0.04 (0.4)	0.04 (0.4)	0.04 (0.5)	0.04 (0.4)	0.04 (0.4)	0.05 (0.2)	0.02
Hydroxproline	0.00 (0.0)	0.00 (0.0)	0.00 (0.0)	0.00 (0.0)	0.00 (0.0)	0.00 (0.0)	0.00 (0.0)	-
Aspartic acid	0.64 (6.0)	0.75 (7.5)	0.66 (6.1)	0.65 (7.4)	0.68 (6.2)	0.66 (6.5)	3.00 (12.3)	0.09
Threonine	0.34 (3.2)	0.35 (3.5)	0.35 (3.3)	0.31 (3.5)	0.36 (3.3)	0.34 (3.3)	0.91 (3.7)	0.09
Serine	0.43 (4.0)	0.40 (4.0)	0.43 (4.0)	0.36 (4.1)	0.47 (4.3)	0.45 (4.4)	1.02 (4.2)	0.11
Glutamic acid	2.10 (19.7)	1.85 (18.4)	2.11 (19.6)	1.53 (17.4)	2.19 (19.8)	1.99 (19.6)	4.25 (17.5)	0.42
Proline	0.98 (9.2)	0.92 (9.2)	1.00 (9.3)	0.88 (10.0)	0.99 (9.0)	0.91 (9.0)	1.16 (4.8)	0.10
Lanthionine	0.00 (0.0)	0.00 (0.0)	0.00 (0.0)	0.00 (0.0)	0.00 (0.0)	0.00 (0.0)	0.00 (0.0)	-
Glycine	0.40 (3.8)	0.42 (4.2)	0.45 (4.2)	0.41 (4.7)	0.43 (3.9)	0.36 (3.5)	1.07 (4.4)	0.08
Alanine	0.85 (8.0)	0.74 (7.4)	0.86 (8.0)	0.58 (6.6)	0.91 (8.2)	0.82 (8.1)	1.10 (4.5)	0.26
Cysteine	0.23 (2.2)	0.23 (2.3)	0.25 (2.3)	0.24 (2.7)	0.23 (2.1)	0.20 (2.0)	0.26 (1.1)	0.09
Valine	0.53 (5.0)	0.54 (5.4)	0.53 (4.9)	0.48 (5.4)	0.53 (4.8)	0.49 (4.8)	1.29 (5.3)	0.40
Methionine	0.23 (2.2)	0.18 (1.8)	0.21 (2.0)	0.14 (1.6)	0.23 (2.1)	0.19 (1.9)	0.38 (1.6)	0.06
Isoleucine	0.40 (3.8)	0.37 (3.7)	0.40 (3.7)	0.30 (3.4)	0.41 (3.7)	0.37 (3.6)	1.07 (4.4)	0.09
Leucine	1.47 (13.8)	1.20 (12.0)	1.47 (13.7)	0.89 (10.1)	1.56 (14.1)	1.41 (13.9)	1.92 (7.9)	0.24
Tyrosine	0.35 (3.3)	0.29 (2.9)	0.33 (3.1)	0.24 (2.7)	0.34 (3.1)	0.33 (3.2)	0.78 (3.2)	0.10
Phynylalanine	0.54 (5.1)	0.47 (4.7)	0.53 (4.9)	0.38 (4.3)	0.57 (5.2)	0.53 (5.2)	1.39 (5.7)	0.09
Hydroxylysine	0.01 (0.1)	0.01 (0.1)	0.01 (0.1)	0.01 (0.1)	0.01 (0.1)	0.01 (0.1)	0.01 (0.0)	-
Ornithine	0.00 (0.0)	0.01 (0.1)	0.00 (0.0)	0.01 (0.1)	0.00 (0.0)	0.00 (0.0)	0.02 (0.1)	-
Lysine	0.27 (2.5)	0.36 (3.6)	0.27 (2.5)	0.37 (4.2)	0.28 (2.5)	0.28 (2.8)	1.75 (7.2)	0.18
Histidine	0.32 (3.0)	0.34 (3.4)	0.35 (3.3)	0.35 (4.0)	0.31 (2.8)	0.29 (2.9)	0.79 (3.2)	0.08
Arginine	0.46 (4.3)	0.51 (5.1)	0.45 (4.2)	0.55 (6.2)	0.45 (4.1)	0.43 (4.2)	1.89 (7.8)	0.09
Tryptophan	0.06 (0.6)	0.06 (0.6)	0.06 (0.6)	0.09 (1.0)	0.06 (0.5)	0.06 (0.6)	0.19 (7.8)	0.02
Total	10.65	10.04	10.76	8.81	11.05	10.16	24.33	-
Crude protein	11.64	10.07	11.68	9.76	12.09	10.80	26.92	-

*The values are expressed in w/w = grams per 100 grams of sample. The numbers in parentheses represent the percent (%) of individual amino acid in the crude protein; **AA = Amino acids

In addition, there was a small but significant increase of arginine content of up to 13 to 16% for QPM-SR-SYNTH and QPM Longe 5, respectively, over normal maize. A sensible decrease of up to 47 and 38% was observed for alanine and glutamic acid. A slight decrease in proline, isoleucine, and tyrosine was also noted. Since crude protein was not significantly different between the QPM and normal maize, these changes in amino acid concentrations represent readjustment of amino acid level between the two groups.

Overall, the total basic acids which include lysine, arginine and histidine constituent 12.1 and 14.4% of the total amino acids for QPM-SR-SYNTH and QPM Longe 5, respectively. These values are considerably lower than the acidic amino acids (aspartic acid and glutamic acid) which represent 25.9 and 24.7% of the total amino acid residues for the two QPM varieties. These results are close to those reported by Zarkadas *et al.* (1995, 2000). In normal maize varieties, the basic amino acid values were lower than those found in QPM and account for 9.4 and 9.9% of the total amino acids for MUS-1 and Salongo 2 varieties, respectively. The acidic amino acid values for normal maize were equal or slightly lower than in QPM, being 20 and 25.7% for MUS-1 and Salongo 2. The significant increase of basic totally charged and hydrophilic amino acids compared to normal maize suggest an increase in nonzein protein and hydrophobicity in QPM (Zarkadas *et al.*, 2000). The concentration of lysine in the maize endosperm has been shown to be highly correlated with the content of a single nonzein protein called the protein synthesis factor EF-1α (Habben *et al.*, 1995; Moro *et al.*, 1996). Genetic selection of genotypes with a high EF-1α content can be used to improve the nutritional quality of maize. A number of proteins have been found to have higher lysine content including β-amylase (5%), protein Z (7.1%), chrymotrypsin inhibitors CI-I (9.5%) and CI-2 (11.5%) (Sofi *et al.*, 2009). With sequencing of maize genome being finished, a large number of marker system are now available that are associated with o2 and endosperm modification phenotype (Bantte and Prasanna, 2003; Lopez *et al.*, 2004). An appropriate application of such markers will greatly enhance the efficiency of selection for improvement of grain protein in maize (Bantte and Prasanna, 2003; Sofi *et al.*, 2009).

Nutritional implication of essential amino acid content: In the present study, lysine content in QPM proteins was significantly higher than normal maize, averaging 35.9 and 42 mg g^{-1} for QPM-SR-SYNTH and QPM Longe 5, respectively (Table 4) but it is still below the recommended FAO/WHO (1991) reference lysine standard value of 58 mg g^{-1} of dietary protein for a 2-5 year child.

Specifically, QPM-SR-SYNTH and QPM Longe 5 supply 62 and 72% of human protein requirements compared to 42% for MUS-1, 43% for Salongo 2 and 43% for locale-1.

Although normal maize is not deficient in isoleucine or threonine, the presence of large amount of leucine in human diet can cause both amino acid imbalances and interference of isoleucine absorption. Considering that niacin requirements increased in humans fed primary maize, excess leucine could be partly responsible for pellagra which is primarily caused by a decrease in niacin or tryptophan intake or by a deficiency in lysine leading to deficiency in niacin (FAO, 1992; Zarkadas *et al.*, 2000). The ratio of leucine/isoleucine found in QPM-SR-SYNTH and QPM Longe 5 were 3.24 and 2.97, respectively, compared to 3.6 for Salongo 2 and locale-1 and 3.8 for MUS-1. This indicates that the two QPM varieties provide proteins with a better EAA balance than is indicated from the amino acid profile. This is consistent with Huang *et al.* (2006) stating that a pleiotropic increase in non-zein proteins is contributing to an improved amino acid balance.

Carotenoid analysis: In addition to the amino acid profile, the levels of carotenoids in maize grain were determined. The interest in dietary carotenoids comes from their anti-oxidant properties and the association between carotenoid deficiencies and many chronic human diseases. The four major carotenoids in corn endosperm are lutein, β-cryptoxanthin, zeraxanthin and β-carotene (Moro *et al.*, 1996). These pro-vitamins A are synthesized into fat soluble Vitamin A in mammals. Among the 12 QPM and normal maize varieties analyzed, only MUS-1 (a normal maize) variety has a yellow endosperm, while the other 11 genotypes have white endosperm. Distribution of the four carotenoid among the 12 genotypes used are presented in Table 5. The level of the four carotenoid analyzed was higher in the normal maize MUS-1 variety with yellow endosperm compared to all the genotypes with white endosperm (Table 5). The total carotenoid content in MUS-1 was over 250 fold compared to QPM varieties and other normal maize varieties released in the region (Salongo-2, Kasai-1 and locale-1). The two QPM (QPM-SR-SYNTH and QPM Longe 5) tested have similar levels of carotenoids than other normal maize with white endosperm with the exception of DMR-ESR-W. This later genotype has the highest carotene content among the white maize. It should be pointed out that the level of zeaxanthin in MUS-1 variety accounts for almost

Table 5: Carotenoid in quality protein and normal corn varieties selected in a RD-Congo breeding program

Varieties	Lutein	Zeaxanthin	β-cryptoxanthin	β-carotene	Total carotenoid
MUS-1	1.2176	3.3829	1.6041	0.7545	6.9591
AK-9331-DMR-ESR-Y	0.4118	1.3588	0.5002	0.1822	2.4530
Locale-1	0.0037	0.0058	0.0075	0.0049	0.0219
QPM-Longe-5	0.0033	0.0025	0.0134	0.0059	0.0251
ECA-QVE 6	0.0074	0.0032	0.0158	0.0051	0.0315
DMR-ESR-W	0.0050	0.0006	0.0116	0.0049	0.0221
Salongo-2	0.0047	0.0029	0.0128	0.0046	0.0250
GPS-5	0.0037	0.0006	0.0118	0.0085	0.0246
ECA –QVE 4	0.0037	0.0025	0.0103	0.0057	0.0222
ECA-QVE 3	0.0061	0.0022	0.0132	0.0038	0.0253
Susuma	0.0021	0.0006	0.0104	0.0066	0.0197
QPM-SR-SYNTH	0.0057	0.0049	0.0119	0.0094	0.0319

Values reported are mg carotenoid/kg maize flour on wet weight basis

50% of total carotenoids. These results are consistent with previous reports and confirm that zexanthin is a major pigment in determining endosperm color (Egesel *et al.*, 2003). The level of lutein and zeaxanthin are important in the prevention of age-related macular degeneration (ARMD). This condition affects the center of the field of vision and the ability to see fine detail. Diets rich in lutein and zeaxanthin are one of the options currently available to delay the onset of ARMD.

Impact of QPM on poultry growth rate and human health: Productivity in the poultry industry depends primarily on genetic potential of the fowl, the physical environment and the quality of food. Genetic selection for fast growth is a continuous process in this industry. To exploit fully the genetic potential of monogastric animals such as pigs and poultry, high nutrient density diets that are balanced in all the nutritive elements are required. Pigs and poultry for example, require a more complete protein than cereals alone can provide, as in case with normal maize that is deficient in lysine and tryptophan. A number of studies have proved that the more potential impact of QPM can be its use in commercial feeds for pigs and poultry as it results in improved growth (Krivanek *et al.*, 2007; Sofi *et al.*, 2009). There are several evidences that QPM can provide a cheaper way of obtaining a balanced animal feed and that effect can be calculated in monetary terms (Krivanek *et al.*, 2007; Sofi *et al.*, 2009). In the present study, dietary replacement of normal maize with QPM resulted in a significantly higher body weight gain at various growth stages (Table 6) of chicks studied. In fact, the use of QPM resulted in 50% increase in body weight compared to normal maize over a 9 week-period. The weights between QPM and normal maize- fed chicks were significantly different based on the t-test at the 0.05 probability level from week 1 to week 9 (Table 6). The results need to be validated at larger scale within the community. Interestingly, the weight gain was similar to that observed in poultry feed with a mixture of normal maize, soybean and fish products. Considering the current soybean and fish price, the use of QPM will result in a significant monetary saving. It is expected that for small farmers in Africa where a balanced nutriment animal feed is not used and maize is the primary or sole feed component, economic impact of substituting QPM for normal maize will be significant.

In light of the effects of QPM on chick's growth rate, a study targeting malnourished children from a maize growing region of the Eastern Kasai (DR-Congo) was initiated. The preliminary

Table 6: Overtime gain weight in chicks fed with ration supplemented with Quality Protein Maize (QPM) and normal maize flour

	Weeks of chicks'feeding									
Type of maize	0	1**	2**	3**	4**	5**	6**	7**	8**	9**
Weight in grams										
Normal maize	100*	250	350	550	650	800	900	950	1000	1150
QPM	100*	450	700	950	1210	1440	1630	1830	1990	2200

*Represents the starting weight before the trial. ** The weight differences between QPM and normal maize fed chicks were statistically significant (at the 0.05 probability level) from week 1 to week 9 based on the T-test

results from a sampling of 30 children in total showed a sensible weight gain and less disease occurrence for children fed with QPM compared to those under normal maize diet. The design and the protocol of the trial need to be reviewed and expanded at larger scale to validate the results. Detailed data of this study will be published in further reports. To date, a number of studies showed the nutritional superiority of QPM over normal maize with respect to indicators such as apparent nitrogen retention and biological value in human (Kies and Fox, 1972; Krivanek *et al*., 2007). But the impact on improving nutritional and health status needs to be demonstrated in target individuals and communities where malnutrition is prevalent, and where maize is a major component of the diet. An earlier study carried out in Ghana showed that QPM fed children (0-15 months) were healthier, suffer fewer fatalities and had growth rates than children fed with normal maize (Krivanek *et al*., 2007; Sofi *et al*., 2009). Similar results were obtained in India, Guatemala and Brazil, Mexico and Ethiopia (Singh *et al*., 1980; Krivanek *et al*., 2007; Sofi *et al*., 2009).

CONCLUSION

In the present study, QPM varieties with higher grain yield compared to normal improved varieties have been identified. The selected QPM genotypes have similar level of protein content but with a higher lysine and a better amino acid balance compared to normal maize. The levels of carotenoid in QPM varieties tested were similar to those found in other normal maize. The local yellow maize MUS had over 250 fold total carotenoid content compared to QPM and normal maize varieties with white endosperm released locally. Dietary inclusion of QPM in poultry increases body weight by 50% compared to normal maize. A breeding program combining the benefits of QPM and the high level of carotenoid of yellow maize endosperm should produce superior QPM varieties for human and animal nutrition.

ACKNOWLEDGMENTS

We thank Dr. Mario Ferruzzi of Purdue University (Indiana, USA) for carotenoid analysis and the University of Missouri Agricultural Experiment Station Chemical Laboratories (ESCL) (USA) for amino acid analyses. We are grateful to Mrs Ramya Narendrula and Dr. Melanie Mehes-Smith for lab assistance. Contribution of participating farmers for field selection of QPM is greatly appreciated. This research was conducted through a partnership between Laurentian University (Ontario, Canada), University of Kinshasa (DR-Congo) and Caritas Congo. The authors are grateful to the Canadian International Development agency (CIDA) for financial support and the Association of Universities and Colleges of Canada (AUCC) for managing the partnership. We would like to express sincere gratitude to Caritas Development Mbuji Mayi and INERA Gandajika for logistical arrangement. We thank the African Development Bank (ADB) for providing a

financial support through the Forum for Agricultural Research in Africa (FARA) and the Association for Strengthening Agricultural Research in Eastern and Central Africa (ASARECA) to study the impact of QPM on chicks' weight gain and children health.

REFERENCES

AOAC., 2006. Official Methods of Analysis. 18th Edn., Association of Official Analytical Chemists, Gaithersburgs, MD.

Akande, S.R. and G.O. Lamidi, 2006. Performance of quality protein maize varieties and disease reaction in the derived-savanna agro-ecology of South-West Nigeria. Afr. J. Biotechnol., 5: 1744-1748.

Bantte, K. and B. Prasanna, 2003. Simple sequence repeat polymorphism in QPM lines. Euphytica, 129: 337-344.

Bjarnason, M. and S.K. Vasal, 1 992. Breeding for quality protein maize (QPM). Plant Breed. Rev., 9: 181-216.

Egesel, C.O., J.C. Wong, R.J. Lambert and T.R. Rochefort, 2003. Gene dosage effects on carotenoid concentration in maize grain. Maydica, 48: 183-190.

Eggum, B.O., E.M. Villegas and S.K. Vasal, 1979. Progress in protein quality maize. J. Sci. Food Agric., 30: 1148-1153.

FAO., 1992. Maize in human nutrition. Report Series 25, Food and Agriculture Organization, Rome, Italy.

FAO/WHO., 1991. Protein quality evaluation. Expert Consultation, Report Series 51, Food and Agriculture Organization/ World Health Organization Nutrition Meetings, Rome, Italy.

Habben, J.E., G.L. Moro, B.G. Hunter, B.R. Hamaker and B.A. Larkins, 1995. Elongation factor 1 α concentrations is highly correlated with the lysine content of maize endosperm. Proc. Natl. Acad. Sci. USA., 92: 8640-8644.

Huang, S., A. Frizzi, C.A. Florida, D.E. Kruger and M.H. Luethy, 2006. High lysine and high tryptophan transgenic maize resulting from the reduction of both 19- and 22-kD α-zeins. Plant Mol. Biol., 61: 525-535.

Kean E.G., G. Ejeta, B. Hamaker and M.G. Ferruzzi, 2007. Characterization of carotenoid pigments in mature and developing kernels of select yellow-endosperm sorghum varieties. J. Agric. Food Chem., 55: 2619-2626.

Kean, E.G., B.R. Hamaker and M.G. Ferruzzi, 2008. Carotenoid bioaccessibility from whole grain and degermed maize meal products. J. Agric. Food Chem., 56: 9918-9926.

Kies, C. and H.M. Fox, 1972. Protein nutritional value of Opaque-2 corn grain for human adults. J. Nutr., 102: 757-765.

Kniep, K.R. and S.C. Mason, 1991. Lysine and protein content of normal and opaque-2 maize grain as influenced by irrigation and nitrogen. Crop Sci., 31: 177-181.

Krivanek, A.F., H. De Groote, N.S. Gunaratna, A.O. Diallo and D. Friesen, 2007. Breeding and disseminating quality protein maize (QPM) for Africa. Afr. J. Biotechnol., 6: 312-324.

Lopez, M., L. Gloverson and B. Larkins, 2004. Genetic mapping of opaque-2 modifier genes. Maize Genet. Newslett., 69: 165-165.

Mbuya, K., K.K. Nkongolo, A. Kalonji-Mbuyi and R. Kizungu, 2010. Participatory selection and characterization of quality maize (QPM) varieties in savannah agro-ecological region of DR-Congo. J. Plant Breed. Crop Sci., 2: 325-332.

Moro, G.L., J.E. Habben, B.R. Hamaker and B.A. Larkins, 1996. Characaterization of the variability in lysine content for normal and opaque-2 maize endosperm. Crop Sci., 36: 1651-1659.

Scott, M.P., S. Bhatnagar and J. Bertran, 2004. Tryptophan and methionine levels in quality protein maize breeding program. Maydica, 49: 303-311.

Singh, J., S. Koshy, K. Agarwai, N.N. Singh, M. Logha and A. Sethi, 1980. Relative efficiency of opaque-2 maize in growth of preschool children. Ind. J. Nutr. Dieter., 17: 326-334.

Sofi, P.A., S. Wani, A.G. Rather and S.H, Wani, 2009. Quality Protein Maize (QPM): Genetic manipulation for nutritional fortification of maize. J. Plant Breed. Crop. Sci., 1: 244-253.

Vasal, S.K., 2000. Quality protein maize story. Proceedings of the Workshop on Improving Human Nutrition through Agriculture: The Role of International Agricultural Research, (IHNA'00), IRRI, pp: 1-16.

Zarkadas, C.G., Z. Yu, R.I. Hamilton, P.L. Pattison and N.G.W. Rose, 1995. Comparison between the protein quality of northern adapted cultivars of common maize and quality protein maize. J. Agric. Food Chem., 43: 84-93.

Zarkadas, C.G., R.I. Hamilton, Z.R. Yu, V.K. Choi, S. Khanizadeh, N.G.W. Rose and P.L. Pattison, 2000. Assessment of the protein quality of 15 new Northern adapted cultivars of quality protein maize using amino acid analysis. J. Agric. Food Chem., 48: 5351-5361.