Vol. 12(4), pp. 40-44, May 2018
DOI: 10.5897/AJBR2016.0881
Article Number: DB1B2A357442
ISSN: 1996-0778

http://www.academicjournals.org/AJBR

ACADEMIC JOURNALS
expand your knowledge

African Journal of Biochemistry Research

Full Length Research Paper

Biochemical characterisation of soybean germplasms with respect to bioavailable iron and zinc, vitamin A and crude protein

Titilayo Akande[1]*, Francis N. Onyezili[2] and Joseph Ikwebe[3]

[1]Department of Biological Sciences (Biochemistry Unit), College of Science, Federal University of Agriculture, Makurdi, Benue State, Nigeria.
[2]Department of Biological Sciences (Biochemistry Unit), College of Science, Federal University of Agriculture, Makurdi, Benue State, Nigeria.
[3]Department of Biochemistry, Federal University, Wukari, Taraba State, Nigeria.

Received 22 January, 2016; Accepted 25 April, 2016

Soybean germplasms were assessed for bioavailable zinc (Zn) and iron (Fe), vitamin A and crude protein using standard methods. Thirty soybean germplasms were investigated. Results were analyzed using analysis of variance (ANOVA) and showed statistically significant differences in their Zn, Fe and vitamin A concentrations, as well as in crude protein content ($P < 0.05$). The Zn content ranged from 5.74 ± 0.04 to 18.15 ± 0.05 mg/100 g. TGX 1987-62F had the highest amount of Zn (18.15 ± 0.05 mg/100 g), while TGX 1485-1D had the least Zn concentration (5.74 ± 0.04 mg/100 g). The concentration of the Fe ranged from 6.65 ± 0.05 to 11.58 ± 0.05 mg/100 g with TGX 1987-62F showing the highest (11.58 ± 0.05 mg/100 g) and TGX 1987-10F the least (6.65 ± 0.04 mg/100 g) Fe concentrations. Investigation of the vitamin A content showed that TGX 1951-4F had the highest (8.83 ± 0.03 mg/L), while TGX 1989-53F had the least (2.48 ± 0.01 mg/L) vitamin A levels. Crude protein content ranged from 35.40 ± 0.09 to 57.92 ± 0.02%. TGX 1989-11F (57.92 ± 0.02%) showed the highest crude protein content, while TGX 1990-55F showed the least crude protein content (35.40 ± 0.09%). The germplasm TGX 1987-62F consistently showed high levels of crude protein, Zn and Fe concentrations, but moderate levels of vitamin A, whereas TGX 1989-11F and TGX 1951-4F had the highest levels of crude protein and vitamin A, respectively. No direct correlation with earlier observed genetic and morphological variabilities were observed in terms of bioavailable Fe and Zn, vitamin A and crude protein levels in these soybean germplasms. These genotypes can be widely used in the food industry for the commercial production of high quality protein diet for both human and animal consumption, and can be recommended as good sources of the micronutrients Fe, Zn and vitamin A.

Key words: Soybean, diversity, micronutrients, crude protein, iron, zinc, vitamin A.

INTRODUCTION

Soybean [*Glycine max.*(L.) Merrill] is a leguminous annual vegetable belonging to the pea family Fabaceae that grows in the tropical, subtropical, and temperate regions of the world. Nigeria is the leading producer of soybean in sub-Saharan Africa, accounting for 90% of the 437,115 metric tonnes production (FAO, 2012). China and United States of America are the world's largest producers (FAO, 2011). Soybean represents a major source of edible vegetable oil and proteins, containing more than 36% protein, 30% carbohydrates, 20% oil and

excellent amounts of dietary fibres, vitamins and minerals (IITA, 2009). Soybean protein provides all the essential amino acids in the amounts needed for human health. The amino acids profile of soy protein is nearly equivalent in quality to meat, milk, and egg protein.

Malnutrition, particularly protein deficiency is prevalent in many parts of Africa as animal protein is beyond the reach of the majority of the population. Alternative protein sources can be found in many leguminous crops, but soybean is the only available crop that provides an inexpensive and high quality source of protein comparable to meat, poultry and eggs (IITA, 2009). But, alongside protein malnutrition is also micronutrients malnutrition which perhaps is equally devastating. Jajda et al. (2015) recently defined micronutrients as essential elements needed in small amounts for adequate human nutrition and include iron (Fe) and zinc (Zn). Deficiency of these micronutrients has a significant impact on children as well as on the economic development of communities and nations. These deficiencies can lead to serious health problems, including wasting, reduced resistance to infectious diseases, Fe deficiency anaemia, blindness, lethargy, reduced learning capacity, mental retardation and in some cases, death (Singh, 2004; Black et al., 2008).

Zn for example, is widely recognized as an essential micronutrient with a catalytic role in over 100 specific enzymes in human metabolism (Cousins, 2004). It is one of the most ubiquitous of all trace elements involved in human metabolism and plays multiple roles in the perpetuation of genetic materials including transcription of DNA, translation of RNA, and ultimately in cellular division.

Fe is one of the trace minerals that play vital roles in the body. In foods, Fe occurs in two forms- ferrous and ferric, but the absorption form of Fe is only in the ferrous state. Fe is important in reactions involving energy release in the body (oxidation and reduction reactions). It is a component of oxygen-carrying compounds e.g. haemoglobin and myoglobin. If Fe deficiency is not corrected, it leads to anaemia, which is the most common nutritional disorder in the world. The prevalence of Fe and Zn deficiency in resource-poor regions is exacerbated by a reliance on staple food crops which have low bioavailability of the minerals (Akomo et al., 2016).

Vitamin A plays an important role in vision, bone growth, reproduction, cell division and cell differentiation, and also helps to regulate the immune system (Ross, 1998). Vitamin A promotes healthy surface linings of the eye and the respiratory, urinary and intestinal tracts (Semba, 1998). It also helps to maintain the integrity of skin and mucous membranes, which function as barriers to bacteria and virus infections (Ross, 1998). Deficiency of vitamin A in children affects their ability to resist diseases such as diarrhoea, measles and acute respiratory infections. Other consequences of vitamin A deficiency include night blindness and susceptibility to infection by disease-causing microorganisms, among others (Ross, 1998).

Soybean is hailed as the most protective bean because it has the highest protein content amongst plant products and is the only vegetable food that contains all eight essential amino acids. Investigations of the vitamins and minerals levels in soybean have been documented. Khalid et al. (2008) reported that calcium (Ca) content of soybean ranged from 268.75 to 293.0 mg/100 g, copper (Cu) ranged from 1.2 to 1.37 mg/100 g, Fe from 9.04 to 13.32 mg/100 g, magnesium (Mg), 261.0 to 296.0 mg/100 g, manganese (Mn), 3.38 to 4.94 mg/100 g, sodium (Na), 11.9 to 15.11 and Zn ranged from 3.75 to 4.02 mg/100 g. A significant amount of micronutrients, especially Fe, Zn and Ca are complexed with anti-nutritional factors like phytates, oxalates, tannins and polyphenols (Paul et al., 2004), consequently, inhibiting their bioavailability and bioabsorbability (Norhaizan and Nor Faizadatul, 2009; Hunt, 2003) in foods. Legumes like soybean are however reputed to be particularly high in phytates and oxalates (Weaver and Plawecki, 1994).

Earlier, Omoigui et al. (in press) had assessed the genetic and morphological characteristics of the 30 genotypes used in this study using RAPD markers and 10 morphological characters. The results had revealed 70% polymorphism. Cluster analysis for both genetic and morphological assessment placed the germplasms into 5 groups at an agglomerate coefficient of 0.6 (similarity level) and average distance of 0.75, respectively. A significant lack of correlation was observed between the RAPD markers and the morphological traits evaluated. Although, both analyses grouped the soybean genotypes into 5 clades, the composition of the individual clusters were very variable and divergent with the correlation between the largest clusters in the molecular and morphological characterisation at only 35 to 42%. Hence, the current work was intended to evaluate the extent to which these genetic and morphological variabilities affect the concentrations of crude proteins and the essential bioavailable micronutrients, Fe and Zn, and vitamin A in the soybean germplasms under investigation.

MATERIALS AND METHODS

Soybean germplasms (30) (Table 1) used in this study were obtained from Molecular Biology Laboratory, University of Agriculture, Makurdi, Benue State, Nigeria. They represented several independent breeding sources from International Institute of

*Corresponding author. E-mail: titilayomi2013@gmail.com. Tel: +2348020790765.

Tropical Agriculture- IITA.

Determination of Zn and Fe content

Dried seeds of the soybean germplasms were ground into fine powder using mortar and pestle in the first instance, and then with an electric blender (Binatone). The ground materials were then passed through a 1 mm sieve and stored in air tight glass containers at room temperature until required. The ground soybean materials were digested using wet digestion method. 1 g of each sample was placed in 300 Kjeldahl digestion flasks and to it was added 10 ml of 95% concentrated nitric acid, 0.5 ml of sulphuric acid (98%) and 1 ml of 70% perchloric acid. The mixture was then digested initially at a moderate heat and later more vigorously until evolution of white fumes. The digest was then allowed to cool, filtered and transferred to a 50 ml volumetric flask. The volume of the solution was made up to the mark with distilled water, and then transferred to a bigger flask. Zn and Fe concentrations were determined using Unicam Atomic Absorption Spectrophotomer, model 290B following A.O.A.C (1990) with absorbance read at 213 and 248 nm, respectively.

Determination of vitamin A

One gram each of ground soybeans was placed in test tubes. 5 ml of propanol (98%) was added and allowed to stand for 10 min to extract properly. The samples were filtered and 1 ml of each filtrate was transferred into a cuvette and absorbance read at 325 nm using a UV - Visible spectrophotometer following A.O.A.C (1990). The absorbance was proportional to the vitamin A content in the sample.

Determination of crude protein content

Total protein content was determined by Kjedahl method (A.A.O.C.,1998). 2 g of each sample was weighed into 100 ml Kjedahl flask and a few anti-pumping granules were added. 8 g of a mixed catalyst (CuSO4 and Na2SO4) (anhydrous) and 30 ml of concentrated sulphuric acid were added. The flask was placed on a Kjeldahl digestion flask rack and heated until a clear solution was obtained. At the end of the digestion, the flask was cooled and the digest was transferred to a 100 ml volumetric flask and made up to the mark with distilled water. 5 ml of the digest was measured into a Markham semi micro nitrogen steel tube and 7 ml of 50% NaOH solution was cautiously added. The digest was steam-distilled liberating ammonia into a 100 ml conical flask containing 5 ml of 1% boric acid and a drop of methyl red-methyl blue indicator was added until the colour changed from pink to green. 20 ml was then collected and titrated with 0.1 M HCl. The end point was indicated by a colour change from green to pink and the volume (V) of the acid for each sample was noted. Percentage nitrogen per sample was calculated using the Equation 1:

$$\% \text{ Nitrogen} = \text{Titre} \times 0.0014 \times 100/\text{Weight of sample (2 g)} \quad (1)$$

The crude protein was calculated as:

$$\text{Protein (\%)} = 6.25 \times \% \text{ Nitrogen} \quad (2)$$

Statistical analysis

All the measurements were analysed statistically using the Statistical Package for Social Sciences (SPSS) version 12.0 and the results expressed as mean ± standard error of the mean (SEM). Analysis of the differences in the measured parameters was performed using one way analysis of variance (ANOVA). Statistically significant difference was determined at $P < 0.05$.

RESULTS AND DISCUSSION

The nutritional compositions with reference to bioavailable Zn and Fe, vitamin A and crude protein of thirty soybean germplasms were determined. The Zn content of the soybean germplasms ranged from 5.74 ± 0.04 to 18.15 ± 0.05 mg/100 g. TGX 1987-62F had the highest amount of Zn (18.15 ± 0.05 mg/100 g), while TGX 1485-1D had the least Zn composition (5.74 ± 0.04 mg/100 g) as shown in Table 1. Each entry represents a mean of three readings. The concentration of Fe ranged from 6.65 ± 0.05 to 11.58 ± 0.05 mg/100 g TGX 1987-62F had the highest Fe concentration (11.58 ± 0.05 mg/100 g) while TGX 1987-10F recorded the least Fe concentration (6.65 ± 0.04 mg/100 g). The values for Zn and Fe of the soybean germplasms were higher than the values for Zn (8.4 ± 0.2 mg/100 g) and Fe (9.3 ± 0.2 mg/100 g) reported by Odumodu (2010). Vitamin A content assessment of the soybean germplasms showed that TGX 1951-4F had the highest vitamin A content (8.83 ± 0.03 mg/L), while TGX 1989-53F had the least vitamin A content (2.48 ± 0.01 mg/L). Each entry also represents a mean of three readings.

Crude protein analysis showed values ranging from 35.40 ± 0.09 to 57.92 ± 0.02%. TGX 1989-11F (57.92 ± 0.02%) showed the highest crude protein content, while TGX 1990-55F showed the least crude protein content (35.40 ± 0.09%). Generally, the soybean germplasms had significantly higher protein content than reported in the literature, even recently (37.53 to 40.20%) by Ari et al. (2012). In addition, this result gives credence to the report of Maidala et al. (2013) who reported crude protein levels ranging between 40.13 and 56.66%.

The genotype TGX 1987-62F consistently showed high levels of crude protein, Zn and Fe concentrations, but moderate levels of vitamin A (3.013 ± 0.02 mg/L). Similarly, the highest level of crude protein was observed in the genotype TGX 1989-11F, but the same showed only moderate concentrations of Fe, Zn and vitamin A. Furthermore, TGX 1951-4F had the highest levels of vitamin A observed; the genotype was moderate for Fe, Zn and crude protein.

The higher values of Fe, Zn, vitamin A and crude protein observed in this study may be as a result of the soybean germplasms being hybrids developed and released by IITA for yield improvement, better nutritional content and resistance to biotic and abiotic stresses.

The soybean germplasms showed statistical differences that were highly significant ($P< 0.05$) in their Zn, Fe and vitamin A concentrations, as well as in crude protein content as shown in Table 2. A clear trend observed is that the soybean germplasms investigated showed ample variation of Fe, Zn and vitamin A

Table 1. Zn, Fe, vitamin A and crude protein concentrations in the germplasms.

Genotype	Fe (mg/100 g) [Mean ± SEM]	Zn (mg/100 g) [Mean ± SEM]	Vit A (mg/L) [Mean ±SEM]	Crude protein (%N) [Mean ± SEM]
TGX 1989-11F	7.45 ± 0.05^{bc}	7.64 ± 0.04^{e}	5.05 ± 0.09^{n}	57.92 ± 0.02^{t}
TGX 1990-110FN	9.45 ± 0.05^{d}	10.25 ± 0.05^{l}	2.67 ± 0.01^{b}	50.20 ± 0.01^{o}
TGX 1989-42F	10.35 ± 0.05^{e}	11.55 ± 0.05^{n}	5.40 ± 0.02^{o}	39.87 ± 0.02^{b}
TGX 1990-95F	6.85 ± 0.05^{a}	7.35 ± 0.05^{c}	5.89 ± 0.03^{r}	44.54 ± 0.02^{j}
TGX 1989-45F	9.22 ± 0.02^{d}	10.56 ± 0.04^{m}	3.77 ± 0.01^{g}	51.50 ± 0.50^{q}
TGX 1989-53FN	8.78 ± 0.02^{cd}	14.50 ± 0.00^{t}	2.48 ± 0.01^{a}	41.84 ± 0.04^{e}
TGX 1993-4FN	9.32 ± 0.02^{d}	12.25 ± 0.05^{r}	4.46 ± 0.01^{kl}	48.54 ± 0.20^{m}
TGX 1989-75FN	7.41 ± 0.01^{a}	11.79 ± 0.01^{q}	4.17 ± 0.04^{j}	53.80 ± 0.01^{r}
TGX 1990-78F	10.25 ±0.05^{e}	11.05 ± 0.05^{n}	7.55 ± 0.01^{s}	44.14 ± 0.02^{i}
TGX 1990-114FN	9.10 ± 0.10^{d}	12.55 ± 0.05^{s}	3.64 ± 0.08^{f}	48.28 ± 0.03^{m}
TGX 1987-62F	11.58 ± 0.05^{f}	18.15 ± 0.05^{u}	3.01 ± 0.02^{c}	45.53 ± 0.03^{l}
TGX 1448-2E	9.22 ± 0.03^{d}	10.11 ± 0.01^{k}	4.63 ± 0.02^{l}	43.73 ± 0.03^{h}
TGX 1989-40F	8.75 ± 0.05^{cd}	9.53 ± 0.03^{j}	3.54 ± 0.04^{e}	49.20 ± 0.01^{n}
TGX 1990-52F	7.55 ± 0.05^{bc}	11.53 ± 0.03^{o}	3.05 ± 0.05^{c}	45.32 ± 0.01^{l}
TGX 1989-48FN	7.75 ± 0.05^{bc}	11.64 ± 0.04^{o}	4.26 ± 0.02^{j}	55.92 ± 0.02^{s}
TGX 1990-40F	9.10 ± 0.10^{d}	7.53 ± 0.03^{d}	4.37 ± 0.03^{k}	50.58 ± 0.02^{p}
TGX 1989-49FN	9.64 ± 0.04^{d}	11.64 ± 0.04^{p}	3.46 ± 0.01^{d}	44.87 ± 0.02^{k}
TGX 1990-57F	7.62 ± 0.02^{bc}	11.21 ± 0.01^{n}	3.53 ± 0.05^{e}	39.62 ± 0.01^{b}
TGX 1989-68FN	8.70 ± 0.10^{cd}	8.81 ± 0.01^{g}	4.04 ± 0.02^{i}	42.51 ± 0.01^{f}
TGX 1990-46F	8.53 ± 0.03^{cd}	7.62 ± 0.02^{e}	4.10 ± 0.02^{ij}	40.35 ± 0.03^{c}
TGX 1990-55F	7.75 ± 0.05^{bc}	7.61 ± 0.01^{e}	4.16 ± 0.00^{j}	35.40 ± 0.09^{a}
TGX 1987-10F	6.65 ± 0.05^{a}	7.55 ± 0.05^{d}	3.84 ± 0.00^{h}	43.37 ± 0.01^{g}
TGX 1835-10E	8.10 ± 0.10^{c}	8.93 ± 0.03^{h}	5.70 ± 0.01^{q}	40.08 ± 0.02^{b}
TGX 1485-1D	9.10 ± 0.10^{d}	5.74 ± 0.04^{a}	4.21 ± 0.00^{j}	41.78 ± 0.03^{e}
TGX 1945-1F	7.20 ± 0.10^{b}	6.72 ± 0.02^{b}	5.46 ± 0.04^{p}	44.17 ± 0.04^{i}
TGX 1951-3F	8.10 ± 0.10^{c}	6.82 ± 0.02^{b}	4.88 ± 0.01^{m}	45.03 ± 0.03^{k}
TGX 1935-3F	6.90 ± 0.60^{a}	7.85 ± 0.05^{f}	4.10 ± 0.06^{i}	40.87 ± 0.01^{d}
TGX 1904-6F	8.05 ± 0.05^{b}	7.12 ± 0.02^{bc}	4.48 ± 0.00^{kl}	53.80 ± 0.01^{r}
TGX 1951-4F	9.45 ± 0.05^{d}	9.35 ± 0.05^{i}	8.83 ± 0.03^{t}	45.55 ± 0.05^{l}
TGX 1955-4F	10.40 ± 0.10^{f}	7.62 ± 0.02^{e}	5.07 ± 0.00^{n}	44.54 ± 0.02^{j}
CV	14.43	27.34	29.56	11.50
LSD	0.86	0.10	0.095	0.275

Means on the same column with different superscript are statistically different from each other at ($P < 0.05$). SEM: Standard error of means; CV: coefficient of variation; LSD: least significant difference; Soybean varieties with the same superscript(s) show no statistical difference while soybean varieties with different superscript(s) are statistically different at $P < 0.05$.

Table 2. Analysis of variance for nutritional composition of soybean genotypes.

Source of variation	DF	Fe	Zn	Vitamin A	Crude Protein
Genotype	29	2.9629**	14.4575**	3.5339**	66.2081**
Error	30	0.1785	0.0025	0.0022	0.0181
Total	59	-	-	-	-

** Highly significant at $P < 0.05$; DF = Degree of freedom.

accumulation as well as crude protein. The variations in the investigated parameters consistently correlated with the observed genetic diversity of the germplasms earlier studied. Evaluation of the genetic diversity of the soybean genotypes (Ikwebe et al., in press) using RAPD markers had grouped them into 5 clusters, the compositions of which were very variable and divergent. Although, previous studies (Karr-Lilienthal et al., 2004; Nahashon et al., 2011)

have demonstrated that variations in nutritional quality (Fe, Zn and proteins) of soybean could be a function of geography, growing conditions, etc., the soybean germplasms in the present study were grown in the same controlled environment. Hence, it would appear that the various germplasms inherently possess different abilities to accumulate certain nutrients. It must however, be mentioned that, soybean is generally reputed to be high in phytate and oxalate (Weaver and Plawecki, 1994) which potentially could affect demonstrable levels of Fe and Zn (phytate levels especially, is the subject of a follow up to this article). Nevertheless, the focus here relates to measurable levels of unchelated Fe and Zn which are bioavailable.

CONCLUSION

The concentrations of crude proteins and the essential micronutrients, Fe, Zn and vitamin A in the 30 soybean germplasms studied were very varied and showed no direct correlation with the earlier observed genetic and morphological variabilities. The genotype TGX 1987-62F consistently showed high levels of crude protein, Zn and Fe concentrations, but moderate levels of vitamin A, whereas TGX 1989-11F and TGX 1951-4F had the highest levels of crude protein and vitamin A, respectively. The results also demonstrate that significant levels of Fe and Zn remain unchelated by anti-nutritional agents like phytic acid, and are hence, bioavailable. Nevertheless, an understanding of the phytate-chelated Fe and Zn is required for a more holistic picture of the total Fe and Zn in the germplasms. The genetic and morphological variabilities earlier observed also affect the concentrations of crude proteins and the essential bioavailable micronutrients, Fe and Zn, and vitamin A in the soybean germplasms under investigation. These genotypes can be widely used in the food industry for the commercial production of high quality protein diet for both human and animal consumption, and can be recommended as good sources of the micronutrients Fe, Zn and vitamin A. Also, this study would serve as a bench mark for the identification of potential parents for breeding programme. Public enlightenment on the nutritional importance of these soybean germplasms studied would help enhance their acceptability.

Conflict of interests

The authors have not declared any conflict of interests.

REFERENCES

Akomo PO, Egli I, Okoth MW, Bahwere P, Cercamondi I, Zeder C, Njage PMK, Owino VO (2016). Estimated iron and zinc bioavailability in soybean-maize-sorghum ready to use foods: effect of soy protein concentrate and added phytase. Journal of Food Processing Technology 7(2):1-5.

Ari MM, Ayanwale BA, Adama TZ, Olatunji EA (2012). Evaluation of the chemical composition and anti-Nutritional factors (ANFs) levels of different thermally processed soybeans. Asian Journal of Agricultural Research 6:91-98.

Association of Official Analytical Chemists (AOAC) (1990). Official Method of Analysis, 15th Edition, Washington D.C., USA.

Association of Official Analytical Chemists (AOAC) (1998). Official Method of Analysis, 17th Ed. Washington D.C., USA.

Black RE, Allen LH, Bhutta ZA, Caulfield LE, De Onis M, Ezzati M, Mathers C, Rivera J (2008). Maternal and child undernutrition: global and regional exposures and health consequences. The Lancet 371(9608):243-260.

Cousins RJ (2004). Zinc, Present Knowledge in Nutrition, 7th Ed. Washington, DC. ILSI Press.

Food and Agriculture Organisation of the United Nations (2011). FAO Statistics Division, FAOSTAT database.

Food and Agriculture Organisation of the United Nations (2012). FAO Statistics Division, FAOSTAT database.

Hunt JR (2003). Bioavailability of iron, zinc, and other trace minerals from vegetarian diets. American Journal of Clinical Nutrition 78:633-639.

International Institute of Tropical Agriculture (IITA) (2009). Genetic Resources for Tropical Agriculture, Pamphlet. P 32.

Jajda HM, Patel KG, Patel SR, Solanki VH, Patel KN, Singh S (2015). Comparative efficacy of two standard methods for determination of iron and zinc in fruits, pulses and cereals. Journal of Food Science and Technology 52(2):1096-1102.

Karr-Lilienthal LK, Grieshop CN, Merchen NR, Mahan DC, Fahey GC Jr (2004). Chemical composition and protein quality comparisons of soybeans and soybean meals from five leading soybean-producing countries. Journal of Agricultural Food Chemistry 52:6193-6199.

Khalid AI, Elsiddig AEE, Elfadi EB (2008). Minerals composition of soybean (*Glycine max*. L.) seeds as influenced by *Brady rhizobium* inoculation and chicken manure or sulphur fertilization. Pakistan Journal of Nutrition 7(6):793-800.

Maidala A, Doma UD, Egbo LM (2013). Effects of different processing methods on the chemical composition and anti-nutritional factors of soybean [*Glycine max* (L.) Merrill]. Pakistan Journal of Nutrition 12(12):1057-1060.

Nahashon SN, Kilonzo-Nthenge AK (2011). Advances in soybean and soybean by-products in monogastric nutrition and health, soybean and nutrition, In Tech, El-Shemy H (Ed.). ISBN: 978-953-307-536-5. Available at: http://www.intechopen.combooks/soybean-andnutrition/advances-insoybean-and-soybeanby-products-in-monogastricnutrition-and-health

Norhaizan ME, Nor Faizadatul Ain AW (2009). Determination of phytate, iron, zinc, calcium contents and their molar ratios in commonly consumed raw and prepared foods in Malaysia. Malaysian Journal of Nutrition 15(2):213-222.

Odumodu CU (2010). Nutrient and anti-nutrients content of dehulled and soybean. Continental Journal of Food Science 4:38-45.

Omoigui LO, Akande T, Ikwebe J (2016). Morphological variability and molecular characterisation of soybean genotypes using RAPD markers. African Journal of Biotechnology (in press).

Paul I, Turner RE, Ross D (2004). Nutrition. Jones and Barlett Publishers.

Ross AC (1998). Vitamin A and public health: challenges for the next decade. Proceeding of Nutrition Society 57:159-165.

Semba RD (1998). The role of vitamin A and related retinoids in immune function. Nutrition Review 56:538-548.

Singh M (2004). Role of micronutrients for physical growth and mental development. Indian Journal of Pediatrics 71(1):59-62.

Weaver CM, Plawecki KL (1994). Dietary calcium: adequacy of a vegetarian diet. American Journal of Clinical Nutrition 59:1238-124.

Akande Titilayo: *Department of Biological Sciences (Biochemistry Unit), College of Science, Federal University of Agriculture, P.M.B.2373, Makurdi, Benue State, Nigeria.*