

## Effect of Different Processing Methods on Proximate and Mineral Compositions, and Concentrations of Some Anti-Nutritional Factors in Kidney Bean (*Phaseolus vulgaris*) Seeds

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**Abstract:** Effect of Different Processing Methods of Kidney Bean (*Phaseolus vulgaris*) Seeds on the Proximate and Mineral Compositions, Concentrations of Some Anti-Nutritional Factors; and Performance of Broiler Chickens was evaluated. Raw Kidney bean seeds contained 90.29% DM, 25.96% CP, 2.49% CF, 2.20% E.E, 3.94% Ash and 65.49% NFE. Proximate composition was significantly affected ( $P<0.05$ ) by the different processing methods. Potassium and Iron were the most abundant macro and micro minerals in Kidney bean seeds. There was a significant ( $P<0.05$ ) decrease in the levels of phosphorus and magnesium when Kidney bean seeds were cooked for 60 minutes; sodium level was not affected by soaking in water for 48 hours and cooking for 60 minutes. Fermentation also resulted in significantly ( $P<0.05$ ) the highest destruction of most of the anti-nutritional factors in Kidney bean seeds. Phytate and trypsin inhibitor were the most destroyed by cooking in water for 60 minutes. Soaking for 24 hours resulted in the destruction of 16.67% TIA, 5.45% phytate, 2.33% tannin, 16.13% oxalate and 1.91% HCN. Fermentation seemed to be most effective in getting rid of some of the TIA, HCN and tannin in Kidney bean seeds; while cooking in water for 60 minutes was most effective in destroying phytic acid. Soaking for 24 hours and then fermenting for 72 hours resulted in the destruction of 33.33% TIA, 34.93% HCN, 22.58% oxalate 16.28% tannin and 10.91% phytic acid. Apart from fermentation, and cooking for 60 minutes; soaking for 48 hours also appeared to be a promising method for processing Kidney bean seeds.

**Keywords:** Proximate, Mineral Compositions, Kidney Bean (*Phaseolus vulgaris*) Seeds

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### INTRODUCTION

The quest for non-conventional feed sources for feeding livestock is an on-going exercise in the developing countries of the world, most especially as the prices of conventional feed stuffs continue to escalate. The livestock producer is faced with the problem of trying to reduce the cost of feeds in order to maximize profit. In developing countries, majority of the populace suffer from protein deficiency, therefore the need to look for cheaper and simple ways of getting the animal protein

required for normal body growth and functions (Ayanwale *et al.*, 2006).

Feed is the most important input in a profitable poultry production, because it accounts for 70-80 % of the total cost of production (Bello, 1984; Ogundipe, 1987 and Kehinde *et al.*, 2006). The use of non-conventional feed ingredient and the search for other feed resources that are not expensive is therefore necessary (Farinu *et al.*, 2006). Non-conventional feedstuff offers the best

alternative in our environment for reduction in feed cost (Dafwang *et al.*, 2001).

Igwebuike *et al.* (2001), Taiwo *et al.* (2004) and Ani and Okafor (2004) suggested that the solution to the problem of animal protein shortage in Nigeria lies in the production of highly prolific animals that are efficient converters of feed to flesh, and the integration of a wide array of cheap and locally available non-conventional feedstuffs at our disposal into well-defined feeding systems to reduce cost.

Kidney bean contains high amounts of protein and energy, and its amino acids content is similar to that of soybean except for a lower level of methionine (Laurena *et al.*, 1991). However like other tropical legumes raw kidney beans contain trypsin inhibitors, amylase inhibitors, haemagglutinins, tannin, phytic acid, and oxalates (Liener, 1989). These anti-nutritional factors negatively affect the nutritive value of the bean through direct and indirect reactions: they inhibit protein and carbohydrate digestibility; induce pathological changes in intestine and liver tissue, thus affecting metabolism; inhibit a number of enzymes; and bind nutrients, making them unavailable (Bressani, 1993). Inclusion of raw kidney beans in the diet of growing animals as the only source of plant protein almost invariably leads to a significant impairment in growth (Ologhobo *et al.*, 1993) and other undesirable physiological and biochemical alterations (Aletor and Aladetimi, 1989). Heat treatment is reported to reduce or totally eliminate anti-nutritional factors in some legumes (Amaefule and Obioha (2001) and Marty and Chavez (1993).

## MATERIALS AND METHODS

The creamy white variety of Kidney bean seeds used in this study were purchased from Akwanga town market, Akwanga Local Government Area of Nasarawa State. Samples were analyzed for their proximate and mineral compositions according to the methods of A.O.A.C. (2005).

The processing methods used were adopted from the works of Matthew *et al.* (2010).

i. Soaking: 1 kg of Kidney bean seeds were soaked in 2.5 litres of water in open containers for 24 hours and a different batch of 1kg Kidney bean seeds were soaked for 48 hours. The water was decanted and replaced after every 8 hours. At the completion of 24 hours and 48 hours respectively, the water was drained and the sample dried under the sun for 5 days on concrete floor.

ii. Fermentation: A different batch of 1kg raw Kidney bean seeds were soaked in 2.5 litres of water for 24 hours and then removed and the water allowed to drain. The soaked seeds were then bagged and put into air-tight polythene bag and allowed to ferment for 3 days (72 hours). The seeds were then sun-dried to practical dryness on a concrete floor for 5 days.

iii. Cooking: To obtain the cooked sample of Kidney bean seeds, 2.5 litres of water was put into a 6-litre silver pot

with a cover and allowed to reach boiling point (100°C) on a kerosene stove with a steady flame. 1kg of the seeds were then poured into the boiling water and allowed to boil or cook for 60 minutes. The pot was then removed from the fire and the water drained, the seeds put on a concrete floor and sun-dried for 5 days.

iv. Raw Kidney bean seeds were milled and used for comparison.

Products obtained from all these processing methods as well as the raw kidney bean seeds were separately milled using a laboratory milling machine with a screen size of 0.5 mm

Samples were analyzed for proximate chemical composition; dry matter (%DM), crude protein (%CP), Crude fibre (%CF), Ether extract (%EE) and Ash using the procedure outlined by A.O.A.C (2005). Nitrogen free extract (%NFE) was calculated by the difference; using the formula:

$$100 - (\%CP + \%CF + \%EE + \text{Ash} + \% \text{Moisture}).$$

The metabolizable energy was calculated from the proximate composition data using the formula described by Pazungu (1985).  $ME = 35 \times \% \text{ crude protein} + 81.8 \times \% \text{ ether extracts} + 35.5 \times \% \text{ nitrogen free extract}$ . Macro kjeldahl method of A.O.A.C. (1990) was used in determining the Nitrogen (N) content and ash content determined as the residue remaining after incinerating the sample at 600°C for 3 hours in a muffle furnace.

Methods described by A.O.A.C. (1990) were also adopted in determining the mineral profile of the samples. Potassium and sodium were determined by flame photometry using the flame photometer at 967 and 589nm, respectively; while calcium, copper, iron, magnesium, manganese and zinc were determined by using the Perkin-Elmer (Model 403) Atomic Absorption Spectrophotometer (AAS).

Tannin content of the samples were determined by Vanillin-HCl procedure which was based on an acid-catalyzed addition of Vanillin to flavonols and their polymers as well as addition to other polyphenolic compounds such as dihydrochalcone and flavones. These reactions were determined colorimetrically at 500nm according to Earp *et al.* (1981).

Phytate was determined by the method of Wheeler and Ferrel (1971) while Trypsin inhibitor was determined by the method of Kakade *et al.* (1974), with modification by Liu and Markakis (1989).

The cyanide content was determined according to the method of Cooke and Maduagwu (1978), as modified by Ikediobi and Fashagba (1985), and spectrophotometric method of Brunner (1984) was used for the analysis of Saponins.

All data obtained from the study were statistically analysed using the general linear model of Statistical Analysis Systems (SAS, 2008).

## RESULTS AND DISCUSSION

Treatments had significant ( $P < 0.05$ ) effect on the kidney bean seeds (Table 1). The DM was significantly ( $P < 0.05$ ) increased by all the processing methods except in the cooked sample. Crude protein increased ( $P < 0.05$ ) by soaking the seeds in water for 24 hours and by fermentation, but reduced ( $P < 0.05$ ) by soaking in water for 48 hours and cooking for 60 minutes. CP in all the treatments ranged from 24.39-38.95 while that of DM, CF, EE, Ash and NFE ranged from 89.88-91.27, 2.06-3.41, 1.89-2.89, 3.37-4.07 and 51.65-67.28 respectively.

The percentage dry matter of raw Kidney bean seeds (90.29%) is similar to those reported by Marzo *et al.* (2002), Audu and Aremu (2011) and Sissay *et al.* (2014). The % DM of Kidney bean seeds increased significantly when the seeds were soaked in water for 48 hours and reduced by cooking in water for 60 minutes. The percent crude protein value (25.96) obtained for the raw Kidney bean seeds in this study was close to 24.70% reported by Emiola (2011). The differences in the percentage crude protein value of Kidney bean seeds reported by various authors could be due to the variety of Kidney bean seeds used, types of soil on which they were grown, as well as the method and the accuracy of the laboratory analysis employed.

Oboh (2006) reported increase in the protein value of the processed kidney bean seeds due to break down of crude protein to amino acids during processing.

There was a significant reduction in the percentage crude fibre of raw Kidney bean seeds with increased soaking time (48 hours). The crude fibre of 2.49 % obtained was lower than 5.1 and 5.0 reported by Arijia *et al.* (2006) and Emiola (2011) respectively. Eke *et al.* (2008) reported that kidney bean seed fell within the acceptable range of 2.7 to 7.9% CF. The importance of fibre in the diet cannot be over emphasized. Fibre helps to maintain the health of gastro intestinal tract, and also prevent colon cancer (Audu and Aremu, 2011).

The 2.20% ether extract obtained in this study for the raw Kidney bean seed compared favourably with the oil content of most under-utilized legume grains (Oke *et al.*, 1996; Amaefule and Obioha, 2001). Total ash content of the raw Kidney bean seeds declined significantly when soaked in water; probably due to leaching of soluble inorganic salts into the processing water.

The low value of ash in the raw seeds may be as a result of the effect of anti-nutrients on its mineral contents, because anti-nutrients could interfere with the bioavailability of minerals (Alonso *et al.*, 2001 and Anigo *et al.*, 2009); however, since anti-nutrients are heat labile, processing can reduce the levels of the anti-nutrient, thereby improving the bio-availability of the minerals as seen in the resultant increase in the ash values of flour from processed kidney bean seeds as reported by the authors.

**Table 1:** Proximate Composition of Differently Processed Kidney Bean Seeds

Parameters	Treatment					SEM	LOS
	1 (Raw)	2 (Sk 24hrs)	3 (Sk 48hrs)	4 (Fmtd)	5 (Cooked)		
DM	90.29 <sup>d</sup>	90.58 <sup>c</sup>	91.27 <sup>a</sup>	90.75 <sup>b</sup>	89.88 <sup>e</sup>	0.02	*
CP	25.96 <sup>c</sup>	30.27 <sup>b</sup>	24.85 <sup>d</sup>	38.95 <sup>a</sup>	24.39 <sup>e</sup>	0.01	*
CF	2.49 <sup>d</sup>	2.66 <sup>c</sup>	2.06 <sup>e</sup>	3.41 <sup>a</sup>	2.79 <sup>b</sup>	0.01	*
EE	2.20 <sup>c</sup>	2.89 <sup>a</sup>	1.98 <sup>d</sup>	2.75 <sup>b</sup>	1.89 <sup>e</sup>	0.01	*
Ash	3.94 <sup>b</sup>	3.68 <sup>d</sup>	3.76 <sup>c</sup>	3.37 <sup>e</sup>	4.07 <sup>a</sup>	0.02	*
NFE	65.49 <sup>c</sup>	60.64 <sup>d</sup>	67.28 <sup>a</sup>	51.65 <sup>e</sup>	66.96 <sup>b</sup>	0.02	*

a,b,c,d,e; Means on the same row with different superscripts differ significantly ( $P < 0.05$ )

DM = Dry matter, CP = Crude protein, CF = Crude fibre, EE = Ether extract, NFE = Nitrogen free extract, SEM = Standard error of means, LOS = Level of significance, Sk 24hrs = Soaked 24hrs, Sk 48hrs = Soaked 48hrs, Fmtd = Fermented.

Raw Kidney bean seeds gave the highest phosphorus (2981.00) and magnesium (374.18) among other treatments, while cooked Kidney bean seeds had significantly ( $P < 0.05$ ) higher potassium (13831.00) and the lowest was obtained in Kidney bean soaked for 48

hours (7171.00). The highest levels of sodium and calcium were obtained in Kidney bean seeds soaked for 24 hours (Table 2). The variation in the mineral composition of kidney bean could be as a result of processing methods and genetic factors which affected

**Table 2: Effect of Different Processing Methods on Some Macro Mineral Levels of Kidney Bean Seeds (Mg /l)**

Mineral	Treatment					SEM	LOS
	1 (Raw)	2 (Sk 24hrs)	3 (Sk 48hrs)	4 (Fmtd)	5 (Cooked)		
P	2981.00 <sup>a</sup>	2771.00 <sup>b</sup>	2241.00 <sup>e</sup>	2281.00 <sup>d</sup>	2491.33 <sup>c</sup>	0.65	*
K	13171.00 <sup>b</sup>	12331.00 <sup>c</sup>	7171.00 <sup>e</sup>	11171.00 <sup>d</sup>	13831.00 <sup>a</sup>	0.58	*
Na	171.00 <sup>c</sup>	201.00 <sup>a</sup>	171.00 <sup>c</sup>	181.00 <sup>b</sup>	171.00 <sup>c</sup>	0.58	*
Ca	237.34 <sup>b</sup>	318.25 <sup>a</sup>	237.37 <sup>b</sup>	237.37 <sup>b</sup>	237.35 <sup>b</sup>	0.01	*
Mg	374.18 <sup>a</sup>	369.76 <sup>b</sup>	347.79 <sup>d</sup>	365.38 <sup>c</sup>	365.38 <sup>c</sup>	0.01	*

a,b,c,d,e; Means on the same row with different superscripts differ significantly (P<0.05)

P = Phosphorus, K = Potassium, Na = Sodium, Ca = Calcium, Mg = Magnesium, SEM = Standard error of means, LOS = Level of significance, Sk 24hrs = Soaked 24hrs, Sk 48hrs = Soaked 48hrs, Fmtd = Fermented.

the accumulation of minerals in the embryo and seed coat. Common bean seeds contained over 94.5% calcium in their seed coat and from 76.0 to 89.7% potassium in their embryo. The levels of phosphorus and magnesium in the seeds were significantly (P<0.05) reduced to 2241.00 and 347.79 by soaking in cold water for 48 hours, possibly due to leaching of minerals into the water. The result revealed a significant decrease in the level of phosphorus and magnesium when the seeds were processed, particularly by soaking in water for 48 hours and cooking in water for 60 minutes; this could be due to the leaching of these minerals into the water. This is in line with Ologhobo (1980) who reported a decrease in the levels of P, Ca, Mg and Na in Ife brown, Aduki and Far V-13 varieties of cowpea when cooked. Aletor and Ojo (1989) and Iyayi and Egharevba (1998) also reported a decrease in the K, Mg, Na and Ca levels of samples of cooked soybean, lima beans and *Mucuna* seeds. This

study revealed that phosphorus is the most abundant mineral in Kidney bean seeds followed by potassium. Kingsley (1995) reported greater portion of potassium in the hull of oil beans which was significantly reduced during cooking due to leaching into the water. Sodium content was the least recorded in Kidney bean seeds.

Treatments significantly (P<0.05) affected iron and zinc contents of kidney bean seeds, but does not have effect on the level of manganese. The highest iron content was obtained in the raw seeds (123.94) and zinc content ranged from 45.32- 130.23 (Table 3). The reason for this variation could be the processing methods and genetic factors which affected the accumulation of minerals in the embryo and seed coat. There was a significant (P<0.05) reduction in the levels of Fe and Zn analyzed. This could be due to the effect of leaching and degree of volatility of the minerals when subjected to heat.

**Table 3: Effect of Different Processing Methods on Some Micro Mineral Levels of Kidney Bean Seeds (Mg/l)**

Mineral	Treatment					SEM	LOS
	1 (Raw)	2 (Sk 24hrs)	3 (Sk 48hrs)	4 (Fmtd)	5 (Cooked)		
Mn	19.35	19.35	19.34	19.35	19.35	0.01	NS
Fe	123.94 <sup>a</sup>	3.84 <sup>b</sup>	0.00 <sup>c</sup>	0.00 <sup>c</sup>	0.00 <sup>c</sup>	0.04	*
Cu	0.00	0.00	0.00	0.00	0.00	0.00	NS
Zn	96.65 <sup>ab</sup>	45.32 <sup>b</sup>	45.32 <sup>b</sup>	130.23 <sup>a</sup>	96.66 <sup>ab</sup>	16.25	*

a,b; Means on the same row with different superscripts differ significantly (P<0.05)

Mn = Manganese, Fe = Iron, Cu = Copper, Zn = Zinc, SEM = Standard error of means, LOS = Level of significance, Sk 24hrs = Soaked 24hrs, Sk 48hrs = Soaked 48hrs, Fmtd = Fermented

The graphical representation of the effect of processing methods of Kidney bean seeds on the percent

destruction of anti-nutritional factors is captured in Figure 1. All the ANFs were found to be higher in the raw seeds

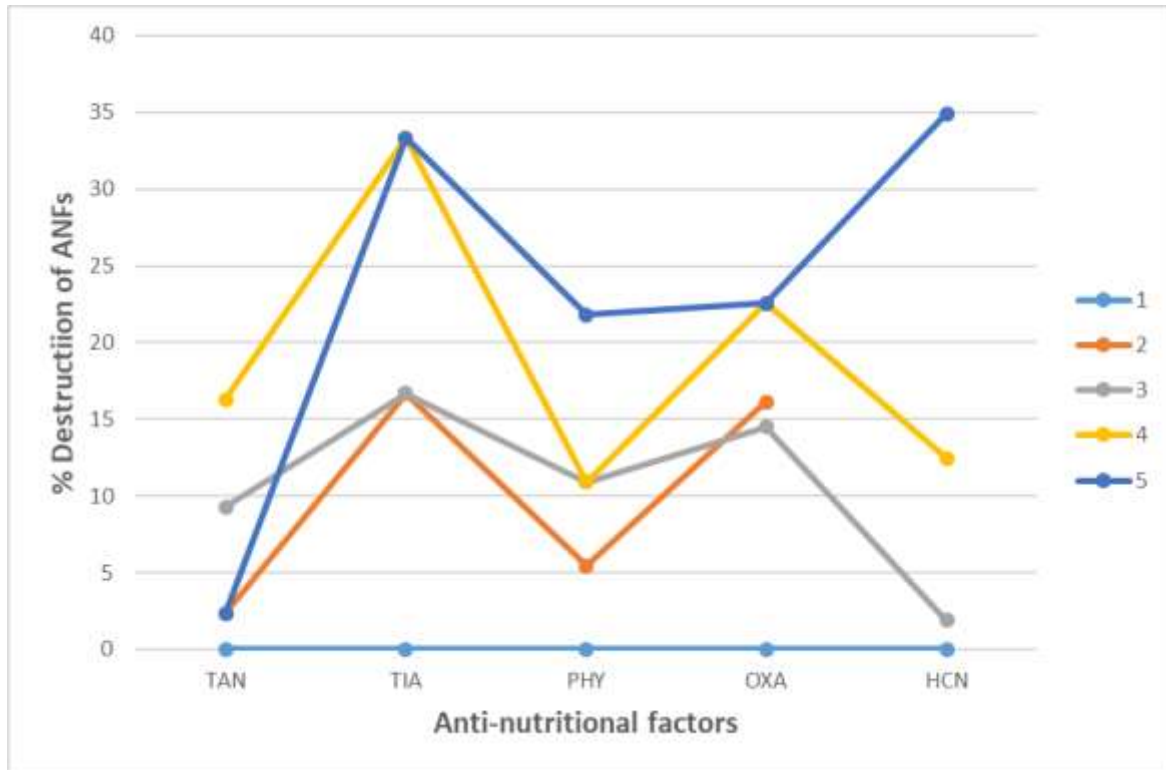


Figure 1: Effect of Processing Methods of Kidney Bean Seeds on the Percent Destruction of Anti-Nutritional Factors

compared to the other treatments (Table 4). Kumar (1998) reported that many anti-nutritional factors are liable to heat and that heat treatment and simple washing with water will alleviate some anti-nutritional factors. The findings also corroborated the work of Nuttapon and

Naiyatat (2009), who reported an overall 94% reduction of mimosine and 99.33% of tannin after processing *Leucaena leucocephala* leaf meal in both, fresh and hot water for 72 and 48 hours respectively. Tannic acid was significantly ( $P < 0.05$ ) destroyed by 16.28% when the

**Table 4:** Effect of Processing Methods on Anti-Nutritional Activities of Kidney Bean Seeds (Mg/100g)

ANFs	Treatment					SEM	LOS
	1 (Raw)	2 (Sk 24hrs)	3 (Sk 48hrs)	4 (Fmtd)	5 (Cooked)		
TAN	0.43 <sup>a</sup>	0.42 <sup>b</sup>	0.39 <sup>c</sup>	0.36 <sup>d</sup>	0.42 <sup>b</sup>	0.00	*
%Destruction	-	2.33	9.30	16.28	2.33		
PHY	0.55 <sup>a</sup>	0.52 <sup>b</sup>	0.49 <sup>b</sup>	0.49 <sup>b</sup>	0.43 <sup>c</sup>	0.00	*
%Destruction	-	5.45	10.91	10.91	21.82		
OXA	0.62 <sup>a</sup>	0.52 <sup>b</sup>	0.53 <sup>b</sup>	0.48 <sup>c</sup>	0.48 <sup>c</sup>	0.01	*
%Destruction	-	16.13	14.52	22.58	22.58		
HCN	2.09 <sup>a</sup>	2.05 <sup>a</sup>	1.83 <sup>b</sup>	1.36 <sup>c</sup>	1.87 <sup>b</sup>	0.05	*
%Destruction	-	1.91	12.44	34.93	10.53		
TIA	0.12 <sup>a</sup>	0.10 <sup>b</sup>	0.10 <sup>b</sup>	0.08 <sup>c</sup>	0.08 <sup>c</sup>	0.00	*
%Destruction	-	16.67	16.67	33.33	33.33		

a,b,c,d; Means on the same row with different superscripts differ significantly ( $P < 0.05$ )

SEM = Standard error of means, LOS = Level of significance, TAN = Tannin, PHY = Phytate, OXA = Oxalate, HCN = Hydrocyanic acid, TIA = Trypsin inhibitor activities, ANFs = Anti-nutritional factors, Sk 24hrs = Soaked 24hrs, Sk 48hrs = Soaked 48hrs, Fmtd = Fermented



seeds were fermented for 3 days and 9.30 % through soaking in water for 48 hours; 21.82% phytic acid was significantly ( $P<0.05$ ) destroyed by cooking in water for 60 minutes. Fermentation led to significant ( $P<0.05$ ) reduction of oxalate from 0.62 to 0.48 mg/100g (22.58 % destruction); similar to the % destruction caused by cooking. The highest % destruction of TIA (33.33 %) was also actualized by cooking the seeds in water for 60 minutes. Cooking resulted in the highest destruction of phytate, oxalate and trypsin inhibitor, while fermentation led to a greater destruction of tannin and HCN. This finding agrees with the work of Matthew *et al.* (2010) who reported cooking of pigeon pea seeds as a viable processing method. Cheftel *et al.* (1993) and Emiola (2004) revealed that cooking was more efficient in reducing tannin and haemagglutinin than decortication. The rate of reduction in the activity of the trypsin inhibitors by cooking for 60 minutes as observed in this study supports the widely held view that the protein inhibitors are easily denatured by heat (Liener and Kakade, 1980).

The different processing methods significantly ( $P<0.05$ ) reduced the phytic acid content of raw Kidney bean seeds especially when cooked for 60 minutes. The phytic acid content in raw Kidney bean seed was significantly ( $P<0.05$ ) reduced from 0.55mg/100g to 0.49mg/100g when the seeds were soaked in water for 48 hours; probably due to its solubility in processing water. Soaking + fermentation of Kidney bean seeds in water for 3 days also significantly ( $P<0.05$ ) reduced the phytic acid content in raw Kidney bean seeds to 0.49mg/100g. Ologhobo and Fetuga (1984) reported soaking in water for 96 hours, followed by germination as an effective method of reducing phytic acid content of raw soybean seeds. The loss of phytic acid according to these authors was due to its solubility in processing water.

This present study revealed that like other grain legumes, Kidney bean seeds contain tannin which were significantly ( $P<0.05$ ) reduced by cooking for 60 minutes, soaking for 24 hours, soaking for 48 hours and soaking + fermentation. This result agrees with the report of other workers who observed that cooking was effective in significantly reducing the tannin content of winged bean (Bressani *et al.*, 1982; Kaankuka *et al.*, 1996). The loss of tannin when the seeds were cooked for 60 minutes could be due to its solubility in water as tannin are known to be water soluble (Kingsley, 1995). Balogun *et al.* (2001) also reported that most anti-nutritional factors in legume seeds

are destroyed after cooking the seeds for a minimum of 30 minutes. Bawa *et al.* (2003) also reported similar result, when Lablab seeds were subjected to boiling at 100°C for 30 minutes. Udedibie and Igwe (1989) and Aletor *et al.* (2002) had earlier suggested that before legume seeds can be incorporated into poultry diets, it should be well processed preferably by cooking for at least 30 minutes to ensure the destruction of most of the anti-nutritional factors.

Cooking appeared to be effective in getting rid of some of the TIA and phytic acid in Kidney bean seeds and the effect is comparable to what was obtained with fermentation; agreeing with Adeyemi and Adeyemi (2000) who earlier observed fermentation as a possible option for processing legume seeds to enhance their full utilization in monogastric diets. Apart from cooking, soaking followed by fermentation appeared to be promising methods for processing Kidney beans (*Phaseolus vulgaris*).

## CONCLUSION

The nutritional profile of Kidney bean affirms it to be an under-utilized legume with a very good potential to be used as protein source in broiler feeds.

Cooking, fermentation and soaking of Kidney bean seeds are effective in reducing Anti-nutritional factors.

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