ISSN 1819-1894 / DOI: 10.3923/ajar.2012.
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# Evaluation of the Chemical Composition and Anti Nutritional Factors (ANFs) Levels of Different Thermally Processed Soybeans

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## ABSTRACT

Chemical composition, amino acid profile, anti nutritional factors (ANFs) and protein quality factors of different thermally (extrusion, cooking, toasting and roasting) processed soybeans (*Glycine max*) for broiler diets were evaluated in this study. Dry Matter (DM) percentage ranged from 79.00 to 91.25% while Crude Protein (CP) percentage ranged from 12.51 to 28.34%. Crude Fibre (CF) values were highest with toasting (28.34) while cooking gave the lowest value (12.51). Ether Extract (EE) highest value was obtained in cooked soybeans (19.50%) while the least (9.72%) was obtained in extruded soybeans. Total ash percentages ranged from 4.27 to 4.46% while NFE percentage ranged from 13.75 to 26.31%. Ca and P percentage values were high (1.08 and 0.33%) in roasted and extruded respectively. Amino acid profiles (g/100 g protein) values for lysine ranged from 2.40 to 5.1, while cystine had values ranging from 0.64 to 0.88. Methionine value were highest (1.02 and 1.14) in cooking and roasting and least in extrusion (0.52) while phyenylalanine values ranged from 3.06 to 4.26. Reduction due to treatment was observed in Trypsin Inhibitor Activity (TIA) and phytic acid and cooking methods was highest for TIA (85%), extruded soybeans (60.59%), toasted and roasted soybeans were similar (52.44% and 53.75%). Reduction in phytic acid was highest with roasted soybean (71.7%), followed by 70.73% in cooked soybeans, toasted soybean was least (48.12%). Urease assay values ranged from 0.02 to 0.09 (ΔpH) and cooked soybean had highest protein solubility index of 83.40%. Phytic acid and Trypsin Inhibitor Activity (TIA), phytic acid and percentage reduction in TIA, Protein Solubility Index (PSI) and phytic acid, TIA and percentage reduction in TIA were correlated. Protein quality and TIA inactivation was more preserved and effective with cooking method.

**Key words:** Anti nutritional factors, amino acid profile, proximate composition, thermal processing, soybeans

## INTRODUCTION

The necessity of heat treatment before feeding full fat soybean diets to poultry have been documented (ASA, 1997). This is due to the presence of several Anti-Nutritional Factors (ANFs) in variable amounts in the raw soybean grain. The ANFs in soybean consist mainly of heat labile (trypsin inhibitors, lectins, goitrogens, phytates) and heat stable (oligosaccharides) factors (Feng *et al*., 2007; Gharaghani *et al*., 2008; Soetan and Oyewole, 2009; Ebrahimi-Mahmoudabad

and Taghinejad-Roudbaneh, 2011). These ANFs compounds interfere with metabolic processes and nutrient availability thereby, leading to the low acceptance and utilization of soybean products (Caprita *et al*., 2010; Coulibaly *et al*., 2011).

Thermal processing of soybean is acknowledged to be very successful in enhancing the nutritional value of soybean and in reducing these ANFs. These processes are however affected by many and varied reports on the influence temperature-time combinations on the ANFs and amino acids profile of soybean among other constraints.

Several thermal and hydro thermal processing techniques of soybean that are aimed at improving the nutritive values and removing ANFs have been documented. These include: dry heating (Papadopoulos, 1987; ASA, 1997; Mridula *et al*., 2008), toasting (Tamiyu, 2001), cooking (Kaankuka *et al*., 1996), extrusion (Asiedu, 1989; ASA, 1997), autoclaving (Balogun, 1989) and infrared (Horani, 1987; Ebrahimi-Mahmoudabad and Taghinejad-Roudbaneh, 2011; Rathnayaka, 2012).

The lack of standardisation of cooking time and temperature regimes and high technology required for either autoclaving, extrusion, micronization, infrared and other thermal based processing methods as well as the energy demand for these processes and the effect of heat on the nutrient content of the full fat soybeans posed serious challenge to average feed processors and small scale poultry farmers.

Some deleterious effects of heat treatment on the nutrient and amino acid composition of soybean have also been reported (Sadiku and Jauncy, 1997) as nutritional losses are often associated with thermal treatments of soybean with very 10°C increase in temperature (Dworschak, 1980; Wang *et al*., 1997). It was for these reasons (Cheva-Isarakul and Tangtaweewipat, 1995) compared different thermal and hydrothermal processes on anti-nutritional factors availability in full fat soybeans and their utilization by broilers.

Steaming was observed to be more effective than roasting in inactivation of TIA and therefore opined that steamed full fat soybeans at steaming time above five minutes could be efficiently used for broilers without detrimental effects. Mild to moderate heating was reported by Miller (2002) to cause loss of sulphydryl groups, formation of disulphide cross links, racemisation of L to D-aspartic acid and reduced digestibility of all amino acids. Some negative effects on production characteristics were also observed when broilers were fed infrared processed full fat soybeans at 28 and 22% inclusion rates in the broiler starter and finisher levels, respectively (Horani, 1987).

This study therefore, evaluates the effects of different thermal and hydrothermal processing methods on the proximate composition, amino acid profile, protein quality factors and some ANFs in soybeans (*Glycine max).*

## MATERIALS AND METHODS

**Seeds collection and processing:** Soybeans seeds *(Glycine max)* were procured from a local market in Lafia metropolis of Nasarawa State, Nigeria. The collected seeds were cleaned by winnowing and hand picking of stones and debris.

**Experimental treatment:** The raw soybeans were subjected to three thermal and hydrothermal processing methods viz: cooking , toasting and roasting (dry heating). Each of these processing methods served as experimental treatment groups to be compared with control (soybean cake

processed through treatment and obtained from Grand Cereals in Jos). The different thermal and hydothermal processes are described as thus:

**Soybeans cake (extrusion processing):** The soybeans cake procured from Grand Cereals in Jos, Plateau State served as the control. This soybeans cake was derived from oil extraction from the whole soybeans. This procedure as described by ASA (1997) involved the initial conditioning of the soybeans before forcing the product through a die. A high accompanying temperature was created by the friction (dry extrusion) or partly by steam injection (wet extrusion).

**Cooking:** The raw soybeans were sorted to ensure homogeneity of product. The soybeans was cleaned and poured into tower aluminium pot containing 50 litres of water per each batch of 50 kg of soybeans. The soya beans was allowed to cook at 100°C for 30 min according to the methods described by Kaankuka *et al.* (1996).

**Toasting:** The cleaned raw soybeans were poured into a hot metal dry pan (common driers). The soybeans were toasted at an approximate temperature of 100°C for 30 min. This is a modification of methods of Cheva-Isarakul and Tangtaweewipat (1995). The dried soybeans were spread to cool before grinding.

**Roasting (dry heating):** In order to reduce the over thickness of the soybeans seed and to allow for even spread of heat in the dry heating process, a modification of the method of Cheva-Isarakul and Tangtaweewipat (1995) was adopted.

The experimental soybeans samples were cleaned and pounded using pestle and mortar in order to increase the surface area of the seeds. One thousand (1000) grams of the seeds were weighed into a tray and roasted (dry heating) in batches using hot plate (Gallenkamp) Muffle Furnace size 2 at a set temperature of 100°C for 30 min. The dry fried seeds were allowed to cool before packaging and grinding.

## ANALYTICAL PROCEDURES

**Proximate analysis:** Proximate composition of each of the processed soybeans samples from the four thermal processing methods were determined according to AOAC (2000) methods. The parameters determined include; moisture, crude protein, crude fibre, lipids and ash content while nitrogen free extract was obtained by difference.

**Determination of amino acid profile:** The amino acid profile of the differently processed soybeans was determined using the methods described by Spackman *et al.* (1958). The soybeans samples were dried to constant weights, defatted, hydrolyzed and evaporated in a rotary evaporator and then loaded into the Technicon sequential multisample amino acid analyzer (TSM).

**Trypsin inhibitor activity (TIA):** Trypsin inhibitor activities of the raw and soybeans subjected to the different processing methods were determined according to the method described by Gupta and Deodhar (1975) and Hammerstrand *et al.* (1981). The methods shared the same principles of determining trypsin inhibitors in soybeans products based on the tryptic hydrolysis of synthetic substrate, benzoyl-DL-arginine-p-nitroanilide (BAPA).

**Phytic acid determination:** The phytic acid in the soybeans samples subjected to the different processing methods was determined according to the modified method described by Wheeler and Ferrel (1971) and Steward (1974).

**Protein solubility index (PSI):** The protein solubility index method described by Araba and Dale (1990), was adopted to ascertain the protein quality of the raw and processed soybeans subjected to the different processing methods.

**pH methods of urease assay:** The determination of pH of the differently processed soybeans samples was done using urease assay as a measure of protein quality (Dudley-Cash, 2004). This was based on the hydrolysis of urea by the enzyme Urease present in soybeans to produce carbon dioxide and ammonia.

## RESULTS AND DISCUSSION

The chemical composition of thermal treated soybean (test ingredient) is presented in Table 1. The dry mater percentage ranged from 79.00 to 91.25% while crude protein percentages range from 12.51 to 28.34%. Crude fibre values were highest with toasting (28.34) while cooking gave the lowest value (12.51). The highest value of ether extract was obtained in cooked soybeans (19.50%) while the least (9.72%) was obtained in extruded soybeans. The total ash percentages ranged from 4.27 to 4.46% while NFE percentage ranged from 13.75 to 26.31%. The highest Ca and P percentage values were 1.08 and 0.33% in roasted and extruded, respectively.

The results of the proximate and chemical composition of thermally processed soybeans are consistent with the reports of HNIS (1989) and Ensminger *et al.* (1990), who observed variations in the proximate composition of soybeans subjected to different processing methods. Similar variations were also reported by Qin *et al.* (1996) and Tamiyu (2001) for different thermally processed soybeans. The proximate values obtained in this experiment are within the range reported in NRC (1994) table for heat and mechanical extruded soybeans.

Amino acid profiles (g/100 g protein) of the different thermal processing of soybean are presented in Table 2. Values for lysine ranged from 2.40 to 5.1 while cystine had values ranging from 0.64 to 0.88. Methionine value were highest (1.02 and 1.14) in cooking and roasting and least in extrusion (0.52) while the phyenylalanine values ranged from 3.06 to 4.26.

The results obtained for amino acid profile indicates that processing method affected the overall amino acid profile of soybeans an earlier observed by OECD (2001). The values obtained in this study for essential amino acids for all the thermal processing methods are within the range reported

Table 1: Effect of thermal processing on the chemical composition of soybean

| | Chemical composition (%) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Thermal methods of processing | Dry matter | Crude protein | Crude fibre | Ether extract | Total ash | NFE | Ca | P |
| Extrusion | 79.00 | 40.20 | 19.50 | 9.72 | 4.27 | 26.31 | 0.45 | 0.33 |
| Cooked | 89.83 | 39.27 | 12.51 | 19.27 | 4.39 | 24.56 | 0.56 | 0.29 |
| Toasted | 91.25 | 35.47 | 28.34 | 18.03 | 4.41 | 13.75 | 0.44 | 0.28 |
| Roasted | 90.57 | 37.53 | 24.29 | 16.92 | 4.46 | 16.8 | 1.08 | 0.29 |

Table 2: Amino acid profile of thermal processed soybean

| | Amino acids values (g/100 g protein) | | | |
|---|---|---|---|---|
| Amino acids | Extrusion | Cooking | Toasting | Roasting |
| Lysine | 2.40 | 5.30 | 3.60 | 5.71 |
| Histidine | 3.21 | 2.60 | 3.00 | 2.90 |
| Arginine | 4.52 | 4.95 | 4.48 | 4.72 |
| Aspartic acid | 11.80 | 11.57 | 10.49 | 10.60 |
| Threonine | 1.47 | 2.99 | 2.80 | 2.49 |
| Serine | 2.45 | 2.59 | 1.90 | 2.38 |
| Glutamic acid | 12.01 | 11.28 | 14.94 | 12.01 |
| Proline | 3.45 | 3.47 | 3.08 | 3.34 |
| Glycine | 2.71 | 3.21 | 3.35 | 2.84 |
| Alanine | 3.47 | 3.70 | 3.04 | 3.14 |
| Cystine | 0.71 | 0.64 | 0.70 | 0.88 |
| Valine | 1.69 | 2.65 | 2.85 | 3.05 |
| Methionine | 0.52 | 1.01 | 0.88 | 1.14 |
| Isoleucine | 1.81 | 2.41 | 2.32 | 2.89 |
| Leucine | 2.94 | 6.80 | 6.00 | 6.73 |
| Tyrosine | 1.69 | 2.90 | 2.63 | 2.49 |
| Phenylalanine | 3.31 | 3.60 | 3.06 | 4.26 |

Table 3: Effect of thermal processing on the values of anti-nutritional factors of soybeans

| Parameters | Raw soya | Extrusion | Cooked | Toasted | Roasted |
|---|---|---|---|---|---|
| Trypsin Inhibitor Activity TIA (mg $k^{-1}$) | 15.35 | 6.05 | 2.30 | 7.30 | 7.10 |
| Reduction in TIA (%) | 0.00 | 60.59 | 85.02 | 52.44 | 53.75 |
| Phytic Acid (mg/100 g) | 345.00 | 102.00 | 113.90 | 178.90 | 97.61 |
| Reduction in PA (%) | 0.00 | 70.73 | 67.25 | 48.12 | 71.71 |
| Urease Assay (ΔpH) | 0.03 | 0.06 | 0.09 | 0.02 | 0.06 |
| Protein Solubility Index PSI (%) | 85.74 | 76.20 | 83.40 | 77.40 | 64.80 |

earlier by OECD (2001). Cooking group grace the cumulative best amino acid profile which indicates that proteins are less denatured by hydrothermal processing. This is in agreement with the report of Balloun (1980), who presented better essential amino acids values for solvent extracted soybeans than extruded soybeans.

The effect of different thermal processing methods on the levels of anti-nutritional factors is shown in Table 3. All the thermal processing methods reduced trypsin inhibitor activity and phytic acid.

However, the percentage reduction of TIA was highest with cooking methods (85%). This value was closely followed in extruded soybeans (60.59%). The reduction proportions for toasted and roasted soybeans were similar (52.44 and 53.75%, respectively). Similarly, reduction in phytic acid was highest with roasted soybean (71.71%) and closely followed by 70.73% in cooked soybeans while toasted soybean gave the least reduction in phytic acid (48.12%). Urease assay values ranged from 0.02 to 0.09 (ΔpH) and the highest protein solubility index of 83.40% was obtained in cooked soybean.

The correlation coefficients of anti-nutritional factors and assessment test factors are presented in Table 4. The following factors and assessors were significantly ($p<0.05$) correlated: phytic acid and trypsin inhibitor activity, phytic acid and percentage reduction in trypsin inhibitor activity,

Table 4: Correlation coefficients between anti-nutritional factors in thermally processed soybean

| Parameters | Correlation | p-value |
|---|---|---|
| Phytic acid vs. TIA | 0.9 | <0.05 |
| Phytic acid vs. reduction in TIA | 0.9 | <0.05 |
| TIA v reduction in TIA | 0.1 | <0.01 |
| Phytic acid vs. PSI | 0.9 | <0.05 |

protein solubility index and phytic acid while trypsin inhibitor activity and percentage reduction in trypsin inhibitor activity were also significantly ($p<0.01$) correlated.

The reduction in trypsin inhibitor activity observed in this experiment was consistent with the report of Cheva-Isarakul and Tangtaweewipat (1995), who indicated that steaming was more effective than roasting in TIA inactivation while phytic acid reduction is best achieved through roasting. Protein solubility index (PSI) and urease assay are both quality indices indicating the degree of heating and indirect determinants of TIA levels in processed soybeans (Soetan and Oyewole, 2009; Thacker and Kirkwood, 1990; Araba and Dale, 1990). The PSI and UA values of 83.40% and ΔpH of 0.09 recorded in cooking was an indication of less denaturation of nutrients through cooking as observed by ASA (1997) and Araba and Dale (1990) and supported by the report of Caprita *et al.* (2010). The high correlation between anti-nutritional factors parameters observed in this study was similarly reported by Qin *et al.* (1996). Protein quality assessment factors were more preserved in cooking method and TIA inactivation was also for effective with this method compared to other thermal processing.

## ACKNOWLEDGMENTS

The authors sincerely acknowledgment the support of Dr. Ojobi of Biological Sciences Laboratory, University of Jos, Mr. Sylvester and Mallam Aliyu of the Microbiology and Biochemistry Laboratories of the National Veterinary Research Institute (NVRI) Vom, Mr. Yau Agade and Munir Mohammed of the College of Agriculture, Lafia Animal Science Laboratory. The financial support received from FIMs services for this research is acknowledged.

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