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# Impact of Soy Flour Processing Methods on its Functional, Proximate and Antioxidant Properties

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*This work was carried out in collaboration among all authors. ‘Author A’ designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. ‘Author B’ and ‘Author C’ managed the analyses of the study. ‘Author C’ managed the literature searches. All authors read and approved the final manuscript.*

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## ABSTRACT

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| This research studied the impact of selected standard processing methods on the functional, proximate and antioxidant properties of soy flours was carried out. Three soy flour samples were prepared using three processing methods (PM1 –PM3). PM1 involves soaking of clean soy bean seeds, dehulling, sundrying (2-3 days), milling, cooling and packaging; PM2 involves soaking with (0.5% NaHCO3), decanting, washing, dehulling, dehusking, sundrying (2-3 days), milling, cooling and packaging; PM3 involves soaking, blanching (100 ᵒC for 5min), draining, cooling, dehulling, sundrying (2-3 days), milling sieving and packaging. The flours were analyzed for functional, proximate and antioxidant properties. Sample means were separated using Duncan’s multiple range comparison test (DMRCT) to determine which sample differed significantly from each of the three (3) soy powder samples. The swelling power, water absorption capacity, oil absorption capacity, bulk density, foam capacity, emulsion capacity, moisture, protein, fat, fibre, ash and carbohydrate contents ranged from 12.50-14.25%, 86.54-88.14%, 70.26-80.63%, 0.70-0.85 g/cm3, 24.50-36.12%, 19.55-24.56%, 7.15- 7.20%, 29.40-36.80%, 7.93-9.11%, 6.51-7.00%, 7.81-9.13%, 32.0-39.97 and 346.51-359.47 kcal/100 g. Soy flour from processing method one (PM1), a traditional soymilk production method modified to produce soy flour significantly affected some of the proximate parameters such as fat, carbohydrate and energy value. Interestingly, processing (modified) method three (PM3) substantially affected some of the proximate (protein, fibre, ash), antioxidant and all functional properties except for oil absorption capacity.  |

*Keywords: Antioxidant properties; foam capacity; functional properties; processing methods; proximate properties; swelling power.*

**1. INTRODUCTION**

In the last ten years, soybean has started gaining research attention due to its nutritional benefits to both human and animal (Liu 2000). Notably, soybean because of its substantial contribution to human nutrition has gained popularity and this is traceable to its richness in protein amounting to 40% and oil about 20% (Enwere1998; Fabiyi 2006; Javed et al., 2024).

Outstandingly, soybean costs low and nutritionally valuable in terms of its dietary protein (McArthur et al.,1988). In proportion, its protein is relatively higher and cost less when compared with that of beef (18%) and other food materials such as chicken, fish, groundnut having 20%, 18% and 23%, respectively (IITA, 1990).

The major hitch known with soybean has to do with content of anti-nutritional factors which are relatively high and is known to be trypsin inhibitors (Brinda et al., 2017; Ibanez et al., 2020) and can be removed when it is subjected to unit operations (heating) involved in its processing to value added food products such as soymilk (Liu, 2012).

An array of food products (soybean oil, soy flour, tofu) has been produced from soybean by Asian countries (Tyug et al*., 2*010). Different processing methods including soaking and grinding, fermentation, pasteurization, thermal treatment, high pressure processing, traditional methods etc are used for the production of these products either to inactive spoilage microorganisms and extend the shelf-life of soymilk or decrease anti-nutritional factors that are found in soy (Yen et al., 1994).

The major unit operation involved in the processing of soybean are soaking (to aid removal of the husk), drying, grinding, sieving and eventual cooking to get slurry after which the resultant cooked slurry is vacuum-dried to get a grade A soymilk powder (Tyug et al*.,* 2010). What is regarded as waste in during this production process should not be discarded but be thrown away but be ultimately be used in the production of feed animal (Osthoff et al., 2010).

Several efforts have been made in the production or processing of soymilk with little or no success in the keeping quality of the product. Soy flour/powder is now the alternative to the aqueous extract of soymilk which closely resembles cow milk in appearance and composition but with short shelf life. Soy flours are prepared using different processing methods to ensure availability of soy protein (soymilk) (Mohajan et al., 2018). However, the effect of these methods on the nutrient and mineral/vitamin compositions, quality and sensory acceptability of the soy flours/powder is not well understood and therefore, should be assessed. It is on this premise that this study was conducted to assess the quality of soy flours as influenced by processing methods.

**2. MATERIALS AND METHODS**

**2.1 Materials**

Soybean seeds (Var: 1904 - 6F) was obtained for this experiment. All the chemicals and reagents (NaHCO3, CCL4, Olive oil etc) needed for use were made available at the University of Mkar, Mkar, Benue state for the preparation of soy flour samples.

**2.2 Methods**

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## 2.2.1 Sample preparation of soy flours

*2.2.1.1 Processing method one - (PM1)*

This is a traditional soymilk processing method described by Iwe, (1991) but slightly modified to produce soy flour. Cleaned sorted and washed whole soybean seeds was soaked in water overnight at room temperature, dehulled manually, sundried for two-three days and milled into soya flour (Fig. 1) using attrition Mill, model No. R175A at rated speed of 2600 rpm. The soy flour obtained was packed into an air-tight container.

*2.2.1.2 Processing method two - (PM2)*

This is a soymilk processing method involving use of sodium bicarbonate described by Afroz et al. (2016) but slightly modified to produce soy flour. In this method, dry whole soybean seeds were cleaned, washed and soaked in a solution of water + 0.5% sodium bicarbonate (NaHCO3) overnight. Removal of soaking water of husk was simultaneous done. The dehulled bean was sundried to its original moisture content and milled into soy flour using an attrition mill model N0 R175 at rated speed of 2600 rpm and packed into air-tight container for further use (Fig. 1).

*2.2.1.3 Processing method three – (PM3)*

This is a soymilk powder processing method described by Cheryl (2021) but with some modifications to produce soy flour. In this method, washed soybean seeds were soaked overnight in a stainless steel container. The following morning the bean was blanched at 1000 C for 5min (modifications) and then removed and placed in a bowel ¾ filled with cold water. The skins or husks were removed manually. The dehulled beans were sundried for 2-3 days and milled in an Attrition mill model No R175 at rated speed of 2600 rpm. The fine soy flour were separated from the spent grains and kept in a container that prevent access to gaseous exchange through the barrier of the packaging materials before being used (Fig. 1).

**2.3 Functional Properties of the Flours**

**2.3.1 Swelling capacity (mL)**

The swelling index with which the swelling capacity was estimated was determined as shown in equation 1 and 2 as indicated by Ukpabi and Ndimele (1990).

SI = $\frac{weight after soaking- weight before soaking}{weight of sample}$ (1)

Swelling capacity was calculated from the result of swelling index as follows:

Swelling capacity = $\frac{weight of wet gel}{weight of sample}$ x 100 (2)

**2.3.2 Water absorption capacity (WAC, %)**

This was determined as reported by Onwuka (2005) and the relation in equation 3 was used to estimate the water absorption capacity in percentage (%).

WAC% = $\frac{Amount of water added-free water}{weight of sample x density}$ x 100 (3)

**2.3.3 Oil absorption capacity**

This was determined as reported in the procedure of Onwuka (2005) for estimating the rate at which oil is absorbed. The relation in equation 4 is employed in its estimation.

OAC% = $\frac{Amount of oil added-free oil}{weight of sampl x density of corn oil }$ x 100 (4)

**2.3.4 Emulsion activity/stability**

Neto et al. (2001) successfully explained in his report steps involved in the estimation of emulsion activity and stability. The relation in equation 5 was instrumental in estimating the content

Emulsion activity (%) = $\frac{H2}{H1}$ X 100 (5)

where,

H1= Initial height of solution before emulsification

H2=height of the emulsified layer

Emulsion stability is often expressed in ratio as stated in equation 5.

**2.3.5 Foam Capacity and stability**

The foam capacity and its stability were estimate according to standard analytical procedures of AOAC, (2006). The relations with which the foam capacity and foam stability were determined are as stated in equations 6 and 7.

Foam capacity (%) = $\frac{V1-VO}{VO}$ X 100 (6)

Where, VO andV1 are the volume before whipping and after

Foam stability = $\frac{Foam volume after timing}{initial foam volume}$ x 100 (7)

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**Fig. 1. Flow chart showing the three processing methods used in the preparation of soy flour**

**2.3.6** **Bulk density (g/ml)**

The soy flours’ bulk density was estimated following the procedure outlined by Onwuka (2005). The relation stated in equation 8 was used in estimating the bulk density.

Bulk density (g/ml) = $\frac{weight of sample}{volume of sampl after tapping}$ x 100 (8)

**2.3.7 Proximate compositions**

All the proximate parameters such as moisture content, crude protein, crude ash, crude fat and crude fibre were all determined as outlined in the standard analytical procedures of AOAC, (2005).

**2.3.8 Determination of carbohydrate content**

The differential method was adapted in determining the carbohydrate content by Serah et al. (2015). All the parameters summed up to be 100%, therefore when all other parameters are known, it becomes easier to estimate carbohydrate content by difference.

 % carbohydrate = 100- sum of % (moisture + fat +protein +fibre +ash) (15)

**2.3.9 Determination of energy value Kcal/100 g**

Food energy containing samples were assesses based on the fact that three (3) critical nutrition parameters are valuable in deriving energy and these are carbohydrate, protein and lipid. The factors employed in its assessment are as stated in equation 16, and are expressed in kcal/100g (Aunyachulee and Chutinan,2019; Alimi et al., 2024b).

Energy value (%) = [(4C) + (4P) + (9F)] kcal/100 g (16)

Where C, P and F are carbohydrate, protein and fat content present in the food material, respectively.

**2.4 Anti-oxidants Properties**

**2.4.1 Estimation by ferrous reducing antioxidant power (FRAP)**

This assessment was done in accordance with the principles outlined by Benzie and Strain (1996). The principle was pivoted on the fact that the reaction of the electron donating antioxidants at low PH results into the formation of coloured complex usually the reduction of Fe3+ TPTZ complex (colourless) to Fe2+- tripyridyl triazine.

**2.4.2 Determination by 1, 1-Diphenyl -2- picryl hydrazyl**

The potential of free radical to scavenge on soy powder during its processing has been outlined and reported by Brand-william et al. (1995) with some adaptations. This scavenging capacity is estimated by the relations in equation 17.

$1, 1-Diphenyl -2- picryl hydrazyl \left( \% \right) = \frac{AB-AA}{AB}x100 (17)$

**2.4.3 Determination by 3 2, 2- Azinobis (3 ethyl)–benzothiozoline-6-suphonic acid) ABTS assay (AA)**

Procedure in step-wise manner by Re et al*.* (1999) outlined the estimation of the scavenging potential of free radicals and their effect on the soy powder samples. This estimation is possible by using the relations in equation 14.

ABTS scavenging effect % = $\frac{AB-AA}{AB}$ x 100

(14)

**2.4.4 Determination by oxygen radical absorbance scavenging capacity (ORAC)**

Ronald et al*.* (2003) outline the method for estimating the oxygen radical absorbance scavenging capacity which was used in this experiment. The standard (Trolox) used subtracted from the under the curve for blank and comparison was subsequently done.

## 2.5 Statistical Analyses

Data from the experiment were analyzed using SPSS (version 21) significant difference between pairs of treatments at 5% level of significance was done applying least significant difference (LSD).

**3. RESULTS AND DISCUSSION**

**3.1 Functional Composition of the Soybean Flours**

The parameters depicting the end use of food material such as flour known as functional properties for soybean are shown in Table 1.

Substantially (P<0.05) different functional properties were obtained under processing methods (PM1, PM2 and PM3). Quality parameters of prime importance that depicts the end of flour is known as the functional properties (Alimi and Alimi, 2019). The swelling power of the soybean flour ranged from 12.50 to 14.25%, with PM3 having the highest while PM1 had the lowest. The processing method three (PM3) was substantially (p<0.05) higher compared to PM2 in terms of the swelling power.

Swelling capacity is used as an index to assess the extent of interaction between the swollen starch granules and water (Alimi et al., 2024a). The ability to absorb water resulting to the swelling of starch granules for soybean flours has the range 86.54 - 87.95%, with PM3 having the while the lowest value was recorded in PM1. There was substantial increase in the ability of soybean flour to absorb water with range 86.54 - 88.14%. This result of this study is similar with the findings of Akubor and Onimawo (2003) who reported that increase in the ability of soy flour to absorb water which could be attributed to high protein content of soy flour which possibly explains why the propensity to absorb water by all the soy flour samples from the processing methods in this study were proportional to their protein contents.

The soybean flours were significantly (p<0.05) different in terms of oil absorption capacity (Table 1). The propensity of the soybean flours to absorb oil has a range 70.26 - 80.63%, with PM1 having the highest while PM3 had the lowest. The ability of flour to retain oil is known as oil absorption capacity, this enhances the flavour and improves the textural properties of food (Alimi et al., 2024a).

Soy flour prepare using PM2 and PM3 were not the same(p>0.05) statistically with respect to bulk density, having the 0.70 - 0.85 g/cm3, with PM1 having the least while PM2 and PM3 had the highest. Bulk density is said to be important to dietary bulk and packaging requirements and could be affected by particle size and initial moisture content of flour (David et al*.,* 2015; Chandra et al*.*, 2015).

The foam capacity of soy flours prepared using the three different standard processing methods were not the same, with range 24.50 - 36.12%, PM1 have the least while PM3 had the highest (Zhu et al., 2017).

The emulsion capacity of soy flours prepared using the three different standard processing methods were not the same (p<0.05), having a range of 19.55 - 24.56%, PM3 have the highest while the lowest was recorded for PM1.

**3.2 Proximate Composition of the Soybean Flours**

The proximate composition of the soy flour prepared using three different standard processing methods are shown in Table 2. The soy flours were not different (p>0.05) in terms of moisture content but were significantly (p<0.05) different in terms of protein contents. The highest protein content was recorded in soy flour processed with PM3 while the lowest was recorded in soy flour processed with PM1. The soy flours from processing methods (PM1 – PM3) yielded substantially (p<0.05) different protein contents due to different processing treatments. According to Ogbemudia et al. (2018) the protein values obtained could be of benefit to take care of people suffering from kwashiorkor. According to the report of research by Serah et al*.* (2015) which indicated the amount of soy possess enough quantity of protein that is needed by the rats, establishing the fact soybean is a good source of protein. Therefore, soy flour could be incorporated into food products such as biscuit, bread, pasta and some other cereal products as an economical protein supplement.

The range of the fat contents is 7.93-9.11%, carbohydrate (32.0-39.97%) and energy value (346.51-359.47 Kcal/100 g), with flour sample from PM1 recording significantly (p<0.05) highest fat, carbohydrate and energy value. Food meant to be consumed should be rich in carbohydrate as this help in the supply of energy that is needed for doing work (Zelalem et al*.,* 2019). The complete oxidation of glucose to carbon dioxide and water by the brain requires carbohydrate as noted by Zelalem et al*.* (2019).

**3.3** **Antioxidant Properties of the Flour**

The characteristics of antioxidant components of soy flour prepared using three different standard processing methods are presented in Table 3. There were significantly (p<0.05) different antioxidant properties. PM3 had the highest (DPPH) % inhibition level of antioxidant properties and free radicals with mean score of 41.02 followed by PM2 with score of 39.13. The ferric reducing antioxidant power of the soy

Table 1. Functional properties of soy flours

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Properties (%) /Method** | **PM1** | **PM2** | **PM3** | **SE** |
| Swelling capacity  | 12.50 ±0.04c  | 12.94 ±0.02b  | 14.25 ±0.00a  | 0 .33 |
| Water absorption capacity  | 86.54 ±0.02c  | 87.95 ±0.02b  | 88.14 ±001a  | 0 .32  |
| Oil absorption capacity  | 80.63 ±0.01a  | 72.96 ±0.01b  | 70.26 ±0.01c  | 1.96 |
| Bulk density (g/cm3)  | 0.70 ±0.00b  | 0.85 ±0.01a  | 0.85 ±0.01a  | 0.03 |
| Foam Capacity  | 24.50 ±0.00c  | 34.15 ±0.00b  | 36.12 ±0.01a  | 2.27 |
| Emulsion capacity  | 19.55 ±0.00c  | 23.71 ±0.01b  | 24.56 ±0.01a  | 0.98 |

*Means with the same superscripts in the same row are not significantly (p>0.05) different (n=2.000); Means are values of two replicates*

Table 2. Proximate composition of soy flours

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Composition (%)  | PM1 | PM2 | PM3 | SE |
| Moisture | 7.20±0.00a | 7.20 ±0.00a | 7.15 ±0.07a | .00 |
| Protein | 29.40±0.00c | 33.96±0.00b | 36.80±0.01a | .00 |
| Fat | 9.11±0.01a | 8.0± 0.00b | 7.93±0.01c | .01 |
| Fibre | 6.51±0.01c | 6.95±0.00b | 7.00±0.00a | .01 |
| Ash  | 7.81±0.06c | 8.16±0.01b | 9.13±0.00a | .01 |
| CHO | 39.97±0.04a | 35.74±0.02b | 32.0 ±0.06c | .03 |
| Energy value (kcal/100 g) | 359.47±0.04a | 350.78±0.03b | 346.51±0.32c | .03 |

*Means with the same superscripts in the same row are not significantly (p>0.05) different (n=2.000) Values are mean ± SD*

Table 3. Antioxidant properties of soy flours

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Method** | **Ferric reducing Antioxidant Power (FRAP)**  | **DPPH Radical scavenging Activity RSA (% inhibition)** | **Trolox equivalent antioxidant capacity TEAC (µM/gDW)** **ABTS** | **ORAC (µM Trolox equivalent (TE)/g)**  |
| PM1 | 0.25 ±0.00c | 31.71 ±0.62c | 28.26 ±0.01b | 28.95 ±0.01a |
| PM2 | 0.38 ±0.01b | 39.13 ±0.04b | 28.95 ±0.01a | 24.20 ±0.01b |
| PM3 | 0.45 ±0.01a | 41.02 ±0.00a | 26.34 ±0.01c | 22.45 ±0.00c |
| SE | 0.04 | 1.80 | 0.01 | 1.16 |
| LSD | - | 1.14 | - | - |

*Means with the same superscripts in the same column are not significantly (p>0.05) different. (n=2.000); Means are values of two replicates*

samples ranged from 0.25 to 0.45 with PM3 recording significantly (p<0.05) highest in FRAP. The results of ABTS indicated that soy flour processing method two (PM2) had significantly (p<0.05) highest scavenging capacity of 28.95 followed closely by PM1 28.26. The standard processing method PM1 has substantially high oxygen radical absorbance capacity (TE)/g of 28.64 as indicated in Table 3. oxidation of lipid and proteins in cells has been noted to be prone to damage by superoxide, hydroxyl, and nitric oxide radicals (Dong ping et al*.,* 2017).

**4. CONCLUSION**

Soy flour from processing method one (PM1), a traditional soymilk production method modified to produce soy flour significantly affected some of the proximate parameters such as fat, carbohydrate and energy value. Interestingly, processing (modified) method three (PM3) significantly affected protein, fibre, ash, some antioxidant properties such as ferrous reducing antioxidant power (FRAP) and 1, 1, diphenyl-2-pycryl- hydrazyl (DPPH) % inhibition and all functional properties except for oil absorption capacity.

**DISCLAIMER (ARTIFICIAL INTELLIGENCE)**

Author(s) hereby declared that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

Competing interests

Authors have declared that no competing interests exist.

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