Effects of Fermentation on the Nutritional Quality of QPM and Soybean Blends for the Production of Weaning Food

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Abstract

The purpose of this work was to study the effects of fermentation on quality protein maize (QPM) and soybean blends with respect to the nutritional quality including physico-chemical and functional properties; microbiological and sensory analyses, minerals and antinutrients composition. Quality protein maize-soybean blend flours were fermented for 24 and 48 hrs by natural and controlled fermentations. In contrary concentration of tannins and phytate were reduced significantly due to the fermentation process. Micronutrients increment in (mg/100 g) for P, Fe and Zn was 32.57 to 61.9; 3.98 to 7.20 and 2.61 to 4.21, respectively were revealed. Fermentation significantly (p<0.05) decreased the antinutrients which resulted a significant increase in micronutrients. Microbiological result revealed significant reduction of undesirable coliform count and increment of LAB with increase in fermentation time. Sensory quality result showed that gruel prepared from the fermented blended flours at 24 hrs of fermentation time and ~250 μm particle size was found acceptable. In line with the result of this study, natural and controlled fermentation uniformly reduced antinutrients composition and improved the nutritional quality of the weaning blends via increased energy and nutrient densities. Fermentation of cereal and legume blends is low-cost and safe technique to save life of children suffering from protein-energy malnutrition.

Keywords: Antinutrients; Fermentation; Micronutrient; Nutritional quality; Blends; Weaning food

Introduction

Fermentation is widely applied in the processing of cereals for the preparation of a wide variety of dishes in Africa and it contributes to the development of acceptable texture, flavour and improves the nutritional quality, digestibility and safety of foods [1]. Fermentation has also been identified to significantly improve the nutritional value (protein quality) of maize-based foods and as well reduce their antinutrients [2].

The high dependence on maize as a staple food in tropical Africa, coupled with the low nutritive value of the commodity has led to the investigation of simple traditional methods in the improvement of the chemical and functional qualities of maize-based foods [3]. The use of legumes such as soybean has been successfully used to increase the nutritional value of cereal foods. Soybean has high protein content and constitutes the natural protein supplements to staple diets. Protein quality is synergistically improved in cereal-legume blends because of the lysine, tryptophan and methionen contributed by the quality protein maize [4]. Soybean products are frequently incorporated into products used for the treatment or prevention of malnutrition. Enriching weaning foods with soy is a convenient, inexpensive, and highly effective way to upgrade the quality of traditional weaning foods and to provide the nutrition a growing child needs. Soy works together with grain proteins to achieve an overall increase in the value of the protein. Adding even small quantities of soy can greatly increase protein content and quality of weaning foods [5]. However, raw ingredients such as cereals and legumes that are used to prepare fermented foods contain significant amounts of antinutreints. These components may decrease the nutritional value of foods by interfering with mineral bioavailability and the digestibility of proteins and carbohydrates [6].

There is urgent need for provision of weaning foods rich in protein, low-cost and suitable for provision of infants’ nutritional needs in order to reduce child malnutrition which is a major global health problem [7]. Blends with a cereal-legume ratio of 70: 30 have been introduced in many communities for use in the preparation of complementary foods with augmented protein quality. These foods should meet World Health Organization estimated energy and nutrient needs from complementary foods [8]. The rationale of this study was to investigate the influence of fermentation on antinutrients composition, functional properties, physico-chemical and sensory characteristics of QPM-based soybean blended weaning foods in order to increase energy, nutrient densities and mineral bioavailability in a weaning gruel.

Materials and Methods

Research materials collection and sample preparation

Bako Hybrid Quality Protein Maize (BHQPY-545) and soybean (Afagt) varieties were collected from Bako and Hawassa Agricultural Research Centers; respectively. All experiments were performed at Addis Ababa Institute of Technology, Food Engineering laboratory, Ethiopian Health and Nutrition Research Institute and Kality Food Processing Share Company. Quality protein maize and soybean samples were prepared according to the method described [9].

Blend formulation

Formulations of high-protein-energy weaning blends were based on the material balance method, targeting 18% protein, 59% carbohydrates [10] and minimum energy value of 380 kcal per 100 g dry matter, according to WFP requirement specifications for particularly the age group of 6 to 18 months. Therefore, the blend ratio of 82:18; QPM: soybean was formulated.

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Natural and controlled fermentation process

Fermentations (NF, CF) were performed using the methods described [11]. After fermentation the samples were dried and grounded and finally placed into plastic containers and stored at 4°C.

Proximate composition and determination of energy value

After blend formulation both NF and CF fermentation process for 0, 24 and 48 hours with particle size distribution of <500 μm and <250 μm. The dried samples were analyzed for the parameters such as protein, crude fat and crude fiber using the method of AOAC and total carbohydrates were calculated by difference [12].

Determination of nutrient and anti-nutrient concentration

The micronutrients: iron, zinc and phosphorus were analyzed using atomic absorption spectoscopy. The antinutrients such as tannins and phytate were determined by the modified Vanillin assay [10].

Determination of functional properties of blends

Bulk density was determined by the method of Narayana [13]; dispersibility in water was determined by the method of Kulkarni [14]; water and oil absorption capacities were determined according to method described by Nwosu, Coffman [15,16]; respectively.

Microbiological analysis

Analyses of mold and yeast; aerobic plate count (APC), coliform count, fecal coliform count and E. coli were done using standard methods of ISO: 7954 [17].

Sensory quality assessment

The prepared weaning foods as gruel from famix provided from FAFA, QPM-Soybean blend flour and blend fermented for 24 and 48 hrs were evaluated by semi-trained panelists. The gruels were prepared by mixing about 10 g of blend flour dissolved in 200 ml distilled water and cooked at 92°C for 15 min. The panelists were asked to rank the gruel on the basis of appearance (color), odor, and taste using a nine point hedonic scale (where 1 = dislike extremely and 9 = like extremely). Overall acceptability of the samples was also rated on same scale with 9 = extremely acceptable and 1 = extremely unacceptable [18].

Results and Discussion

Effect of fermentation on proximate composition and energy value of QPM-soybean blends

Fermentation time significantly (p<0.05) affects protein content of blend flour (Table 1). As fermentation time increased from 0 to 24 and 48 hrs, protein content of blends were 14.71%, 17.43% and 17.52%; respectively. The protein content of blends before and after fermentation (0,12 and 24 hrs) was higher than the minimum protein requirement (14%) of WFP specification for corn-soya blend and within the range (16.00%-19.97%) reported by Lalude [19].

As fermentation time increases the fat content of fermented blends increases from 8.42% to 10.9% with respect to the fermentation time of 0, 24 and 48 hrs (Table 1). This is due to the removal of soluble carbohydrates during fermentation. According to the findings of Amankwah the fat content of formulation of weaning food from fermented maize, rice, soybean and fishmeal was 9.38% and 8.75% [20]. The experimental value is within the range with this value. The value of crude fat content of the blend before and after fermentation (8.42%, 10.2%, and 10.9% to 8.86%) is higher than the value of WFP specification for the minimum requirement of 6% fat of corn-soya blend. And it is comparable with the value of famix infant food (≥7%). The fat content of current study is also within the range with the value (9.0%) of Nutrend- commercially sold Nigerian weaning food.

The value of crude fiber after fermentation (4.49%, 5.32% for particle size distribution of <250 μm and 5.19%, 4.67% for particle size distribution of <500 μm for natural fermentation) and (5.91%, 5.63%) for particle size distribution of <250μm and (6.88%, 5.96%) for particle size distribution of through <500 μm for controlled fermentation) at 24 and 48 h fermentation time respectively (Table 1). Fermentation time is significantly (p<0.05) decreased the crude fiber contents of blend. According to high fiber contents of weaning foods may inhibit mineral absorption and reduce the digestibility of proteins in foods [21]. According to WHO specification the maximum requirement is (5%). Therefore, the experimental values of the blends after fermentation are close to this value. Fermentation time had significantly (p<0.05) decreasing effect on the total carbohydrates content of blend (66.63%, 64.16%, 63.9%) and (66.63%, 61.91%, 60.49%) for particle size distribution of <250 μm and <500 μm at 0, 24 and 48 h fermentation time respectively as can be seen from the This is possibly due to the degradation of carbohydrates by microorganisms and the decreasing effect of fermentation upon the amount of Non-Digestible Carbohydrates (NDC) which are fibers (Table 1). All the experimental values of blend before and after fermentation are comparable with values (67.21% and 63.32%) research findings by Mbata for fermented maize flour and Bambara groundnut-maize fortified flour [22]. It was reported that the total carbohydrate content of maize-soybean blend for the production of Ogi is (61.76%); that is lower than the value of the current study [23].

In the case of controlled fermentation, the calorific values of the blends are decreased, even though there are some fluctuations in natural fermentation. This may be due to the value difference of the nutrients (protein, fat and carbohydrate) by the effect of fermentation process. The calorific value of the blend shown in the Table 1, are in agreement with the researcher reported by Griffith [24]. The calorific value for weaning food revealed in the range of 395 to 509 kcal which is in agreement with the value (398.9 kcal) for “Nutrend” (Nestle, Nigeria-weaning diet) obtained commercially and the research finding reported by (441 kcal) [19]. The value is also within the range provided by WHO specification (380 kcal) for the weaning food from corn-soya blend.

Influence of fermentation process on antinutrients reduction of QPM-soybean blends

During the preparation of many fermented foods, tannins are reduced before the fermentation step because of their presence in the seed coats of raw ingredients. In several fermented foods, the seed coat or testa is removed from the substrate before fermentation. Consequently, the antinutrients potential caused by the presence of tannins is of little concern [11]. Fermentation process further reduced tannins content of the blend (Table 2). Therefore, fermentation time significantly (p<0.05) affected tannins content of the blend. The phytate concentration present in raw materials and foods of plant origin are suggested to be a major factor responsible for lowering the availability of minerals and some proteins as reported [11]. Fermentation duration, type and particle size distribution greatly affects the composition of antinutrients.

Effect of reduction of antinutrients on micronutrients composition of blends

The P, Fe, Zn and Ca concentrations of the blends before fermentation were (32.57 mg/100 g, 3.98 mg/100 g, 2.61 mg/100 g,
and 34.08 mg/100 g) respectively. In the case of NF, for particle size distribution of <250 μm and <500 μm P of the blend is (32.57 mg/100 g, 61.90 mg/100 g, 61.20 mg/100 g) and (32.57 mg/100 g, 59.60 mg/100 g, 55.30 mg/100 g) during 0, 24, and 48 h, respectively. From the results, fermentation time had significantly (p<0.05) decreasing effect on the viscosity of the blend. A prolonged time of starch gelatinization was observed while preparing gruels of fermented blend. This possibly is due to the degradation of starch granules during fermentation and this might cause for the reduction of starch swelling while cooking. As indicated in Table 4, the viscosity of the blend is (5.31×10^{-3} Pa.s) and the viscosity of blend gruel during fermentation of 24 h and 48 h is (4.38×10^{-3} Pa.s, 4.21×10^{-3} Pa.s); respectively. From the results, fermentation time had significantly (p<0.05) decreasing effect on the viscosity of the blend. A prolonged time of starch gelatinization was observed while preparing gruels of fermented blend. This possibly is due to the degradation of starch granules during fermentation and this might cause for the reduction of starch swelling while cooking.

**Table 5:** Viscosity of QPM-soybean blends

| Fermentation type and particle size distribution | Flour samples | Carbohydrates (%) | Calorific value kcal/100g |
|-----------------------------------------------|--------------|-------------------|---------------------------|
| Before fermentation                           | Blend        | 66.63± 0.02       | 400.81                    |
| (<250 μm), NF                                 | Blend24      | 66.90± 0.28       | 412.67                    |
|                                              | Blend48      | 65.01± 0.08       | 404.01                    |
| (<500 μm), NF                                 | Blend24      | 66.74± 0.03       | 412.63                    |
|                                              | Blend48      | 67.97± 0.05       | 404.01                    |
| (<250 μm), CF                                 | Blend24      | 64.16± 0.11       | 410.75                    |
|                                              | Blend48      | 63.90± 0.42       | 387.08                    |
| (<500 μm), CF                                 | Blend24      | 61.91± 0.36       | 387.54                    |
|                                              | Blend48      | 60.49± 0.06       | 384.69                    |

Values in the same column with different superscripts for each type of analysis are significantly different (P < 0.05).

Where BDLP: Below detection limits

**Table 2:** Content of tannins and phytate of blends

| Fermentation type and particle size distribution | Flour Samples | Tannins (mg/100g) | Phytate (mg/100g) |
|-----------------------------------------------|--------------|-------------------|-------------------|
| Before fermentation                           | Blend        | 3.10± 0.21        | 5.05± 0.08        |
| (<250 μm), NF                                 | Blend24      | 3.10± 0.04        | 3.10± 0.04        |
|                                              | Blend48      | 3.00± 0.02        | 2.10± 0.02        |
| (<500 μm), NF                                 | Blend24      | 4.98± 0.06        | 5.05± 0.08        |
|                                              | Blend48      | 4.98± 0.06        | 5.05± 0.08        |
| (<250 μm), CF                                 | Blend24      | 8.94± 0.04        | 138.65± 0.46      |
|                                              | Blend48      | 8.94± 0.04        | 138.65± 0.46      |

Effect of fermentation viscosity of QPM-soybean blends

As indicated in Table 4, the viscosity of the blend is (5.31×10^{-3} Pa.s) and the viscosity of blend gruel during fermentation of 24 h and 48 h is (4.38×10^{-3} Pa.s, 4.21×10^{-3} Pa.s); respectively. From the results, fermentation time had significantly (p<0.05) decreasing effect on the viscosity of the blend. A prolonged time of starch gelatinization was observed while preparing gruels of fermented blend. This possibly is due to the degradation of starch granules during fermentation and this might cause for the reduction of starch swelling while cooking.

Effect of fermentation viscosity on weaning food

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**Table 3:** Effect of fermentation on the nutritional quality of QPM and soybean blends for the production of weaning food

| Fermentation type and particle size distribution | Flour samples | Crude protein (%) | Crude fat (%) | Crude fiber (%) |
|-----------------------------------------------|--------------|------------------|---------------|----------------|
| Before fermentation                           | Blend        | 14.72±0.03       | 8.42±0.01     | 7.33±0.02      |
| (<250 μm), NF                                 | Blend24      | 17.43±0.02       | 10.20±0.14    | 4.49±0.08      |
|                                              | Blend48      | 17.52±0.01       | 10.90±0.28    | 5.32±0.01      |
| (<500 μm), NF                                 | Blend24      | 17.57±0.06       | 10.20±0.07    | 5.19±0.06      |
|                                              | Blend48      | 17.85±0.04       | 10.80±0.21    | 4.67±0.06      |
| (<250 μm), CF                                 | Blend24      | 17.30±0.21       | 9.40±0.14     | 5.91±0.02      |
|                                              | Blend48      | 17.67±0.04       | 8.50±0.35     | 5.63±0.05      |
| (<500 μm), CF                                 | Blend24      | 17.72±0.01       | 9.36±0.06     | 6.88±0.04      |
|                                              | Blend48      | 19.44±0.05       | 8.86±0.02     | 5.96±0.04      |
cause for the reduction of starch swelling while cooking. According to Plahar in terms of starch stability, fortification with soy flour is a cause for strengthening of the starch granules [26].

**Impact of fermentation on functional properties of QPM-soybean blends**

**Bulk density and dispersibility:** As fermentation time increase from 0 to 24 and 48 h, the bulk density significantly decreases for QPM-Soybean blends. The bulk density of QPM before fermentation is (0.84 and 0.92) and after fermentation of 24 and 48 h is (0.62, 0.59 and 0.63, 0.6) for particle size distribution between <250 and <500 μm; respectively. The bulk density of QPM before fermentation is significantly (p<0.05) higher than that of after fermentation. The values (0.66 and 0.61) of the current research finding is in agreement with that of reported Lalude [19], of a weaning food prepared from sorghum based and Nutrend. Similarly, the values are comparable with value (0.68) reported by Mesfin [27]. As fermentation time increased the value of dispersibility significantly (p<0.05) increased from 0.65%, 69% to 66%, 67% and 65%, 69% for fermentation time of 24 and 48 h and particle size distribution of <250 and <500 μm; respectively. The values of both bulk density and dispersability of QPM and blend are higher than the values (0.55 gm/ml and 32.93%) reported by Ahima respectively [28].

**Water and oil absorption:** As indicated in Table 5, water absorption of both QPM and blend is significantly (p<0.05) decreased when fermentation time increased from 0 to 24 h and from 0 to 48 h. Ahima reported that the water absorption of flour from commercially sold floury maize and Maize-soy flour blend is (194.65% and 172.98% respectively) [28]. This result indicated that the water absorption of QPM and blend is lower than that of the researches findings. In the current study the unfermented and fermented blends found to contain comparable amount of water absorption with (134%) that is reported by Emmanuel, cowpea-fortified nixtamalized food. Oil absorption capacity of QPM (1.4 ml, 2.3 ml) is higher than of blend (1.2 ml, 1.7 ml) with particle size distribution of <250 μm and <500 μm. Similarly, QPM fermented for 24 hrs and 48 hrs (4.2 ml, 5.2 ml and 4.8 ml, 5.0 ml) is significantly higher than that of fermented blend (2.2 ml, 3.0 ml and 1.9 ml, 2.8 ml) for particle size of <250 μm and <500 μm respectively. These results indicated that blending QPMF with soybean flour significantly (p<0.05) decreased the oil absorption of the blend. Therefore, blend ratio and fermentation time has significantly (p<0.05) a decreasing effect on the oil absorption. The values obtained from current study are higher than that of (1.22 ml-2.23 ml) reported by Mensah of extrusion cooking of full-fat soy flour [29]. Similarly, oil absorption of both fermented QPMF and blend are higher than the value (1.82 ml, 1.44 ml) for different varieties reported by Assefa of improved varieties of soybean in Ethiopia [30].

**Sensory quality attributes of value added products**

The average results of taste evaluation by the panelist for the control, fermented for 24 h, unfermented blend and fermented for 48 h were 8.9 (like extremely); 7.8 (like very much); 7.2 (like moderately) and 5.8 (like slightly) respectively. The overall acceptability of gruel fermented for 24 h with the average value of 8.5 is not significantly different (p>0.05) compared with that of the control. The result of the others ranged 5.9 to 7.1. This might be due to all the products are evaluated without Flavoring agents. Similar results on weaning and commentary food products were report by Abbey, Beruk [31,32] (Table 6).

**Microbiological quality of blends**

In Table 7 mould count, yeast count, Aerobic Bacteria plate Count (APC), coliform count and others are shown. The result indicated that as fermentation time increased, the undesirable microorganism, mould count, decreased significantly for both type of fermentation. The molds isolated in the current study are commonly present as contaminants and do not appear to play any significant important role in the fermentation process. This shows clearly that the importance of fermentation in the aspect of food preservation.

At 0 hr fermentation time, the yeast count was found to be <1×10⁵ cfu/g, which is considered to be no yeast colonies in the count, but during 24 and 48 h fermentation, the values were increased upon both Natural and Controlled Fermentation. This shows that fermentation time significantly (p<0.05) affect the yeast count. The coliform count at the start of fermentation was found to be 4.3×10⁵ cfu/g and upon increasing fermentation time, 24 and 48 h the count was decreased to 3.1×10⁴ cfu/g and 3.2×10⁴ cfu/g in the case of NF and almost eliminated (<1×10⁴ cfu/g) in the case of CF. The expected decrease or elimination of coliform is in agreement with the value (2.85 cfu/g, 0 cfu/g, 0 cfu/g) for 0, 24 and 48 hrs fermentation time reported by Mbata of fermented maize flour fortified with barbana groundnut [22]. The aerobic bacteria plate count, (APC) as shown in the Table 7 is 1.8×10⁵ cfu/g at 0 h fermentation time and (2.6×10⁴ cfu/g, 2.4×10⁴ cfu/g) in the case of NF and (6.7×10⁴ cfu/g, 5.7×10⁴ cfu/g) for CF during 24 and 48 hrs fermentation time; respectively. Aerobic plate counts taken at 24 h

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**Table 3:** Micronutrient composition of blends.

| Fermentation type and particle size distribution | Flour Samples | P (mg/100g) | Fe (mg/100g) | Zn (mg/100g) | Ca (mg/100g) |
|-------------------------------------------------|---------------|-------------|-------------|-------------|-------------|
| Before fermentation                              | Blend         | 32.57 ± 0.40 | 3.98 ± 0.06 | 2.61 ± 0.08 | 34.08 ± 0.17 |
| <250 μm, NF                                      | Blend24       | 61.90 ± 0.64 | 7.20 ± 0.14 | 4.21 ± 0.15 | 24.91 ± 0.64 |
| <500 μm, NF                                      | Blend48       | 61.20 ± 0.14 | 4.74 ± 0.03 | 3.81 ± 0.08 | 21.34 ± 0.24 |
| <250 μm, CF                                      | Blend24       | 59.60 ± 0.42 | 6.22 ± 0.16 | 3.98 ± 0.06 | 24.92 ± 0.65 |
| <500 μm, CF                                      | Blend48       | 55.30 ± 0.21 | 3.42 ± 0.03 | 3.65 ± 0.06 | 22.53 ± 0.57 |
| Before fermentation                              | Blend         | 32.57 ± 0.40 | 3.98 ± 0.06 | 2.61 ± 0.08 | 34.08 ± 0.17 |
| <250 μm, NF                                      | Blend24       | 61.90 ± 0.64 | 7.20 ± 0.14 | 4.21 ± 0.15 | 24.91 ± 0.64 |
| <500 μm, NF                                      | Blend48       | 61.20 ± 0.14 | 4.74 ± 0.03 | 3.81 ± 0.08 | 21.34 ± 0.24 |
| <250 μm, CF                                      | Blend24       | 59.60 ± 0.42 | 6.22 ± 0.16 | 3.98 ± 0.06 | 24.92 ± 0.65 |
| <500 μm, CF                                      | Blend48       | 55.30 ± 0.21 | 3.42 ± 0.03 | 3.65 ± 0.06 | 22.53 ± 0.57 |

Values in the same column with different superscripts for each type of analysis are significantly different (P < 0.05).

| Blend with respective fermentation time | Viscosity (Pa.s) |
|----------------------------------------|-----------------|
| Blend                                  | 5.31×10⁻⁶ ± 0.22 |
| Blend24                                | 4.38×10⁻⁶ ± 0.26 |
| Blend48                                | 4.21×10⁻⁶ ± 0.15 |

Values in the same column with different superscripts for each type of analysis are significantly different (P < 0.05).
Conclusions

In this research, it was observed that fermentation process significantly changed the nutritional value of the weaning blends by reducing antinutrients. The maximum reduction of phytate due to fermentation effect can lead to the increment of the bioavailability of micronutrients. Subsequently, 47.4% (P), 47.9% (Ca), 44.7% (Fe) and 38% (Zn) increment was observed. The fermentation process affected reducing antinutrients. The maximum reduction of phytate due to fermentation effect can lead to the increment of the bioavailability of micronutrients. Subsequently, 47.4% (P), 47.9% (Ca), 44.7% (Fe) and 38% (Zn) increment was observed. The fermentation process affected

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