Fermentation Conditions and Blending Ratios Effect on Nutritional Composition of Kocho-Fababean Blended Product

Aklilu Dolla^{1*} Solomon Abera²

School of Public Health, Wolaita Sodo University, Wolaita Sodo Ethiopia P.O.Box 138
Haramaya University, Diredawa, Ethiopia P.O.Box 138

Abstract

A study was conducted at Wolaita zone SNNPR, Ethiopia with the objective of evaluating the effect of fermentation conditions and blending ratios on nutritional composition of kocho- fababean blended product. The experiment consisted of a completely randomized design in factorial arrangements with 25, 15 and 5% fababean blending ratios in kocho and 30 day fermentation conditions. The fermentation conditions included (co-fermentation of fababean and kocho for 30 days, fababean added on the 15th day and fababean added after 30 days kocho fermentation). Fababean at 25% kocho showed a significant effect (p<0.001), that resulted in an increased protein, lipid, fiber and ash content from 1.95, 0.39, 2.15, 1.92% to 12.50, 5.04, 2.88, 2.37% but reduced moisture and carbohydrate content from 9.74, 82.31% to 9.64, 67.57%, respectively. All fermentation conditions improved the nutritional value. The moisture, protein, fat, fiber, ash, carbohydrate content of this products (15% fababean added on the 15th day of kocho blend subjected to fababean added on the 15th day of kocho fermentation) were 9.59, 8.71, 3.07, 2.67, 2.11, 73.85 %, respectively. Thus, 15% fababean per 100 g kocho blend subjected to fababean added on the 15th day of kocho fermentation.

Keywords: Kocho, Fababean, Blending ratios, Fermentation conditions, Nutritional compositions

Introduction

Enset (*Ensete ventricosum (Welw.*), *Cheesman*) is endemic to Ethiopia and cultivated as important food crop in southern, southwestern and central parts of the country (Spring *et al.*, 1996). Enset, consisting of more than 100 varieties, is a perennial herbaceous monocot banana-like large (grow 4 to 8 m which even sometimes reach up to 11 m in height) plant belonging to the family Musaceae, genus of banana (Randy *et al.*, 2007). According to CSA, (2008) enset occupies 233,492 ha of land in Ethiopia, of which 71.6, 28.2 and 0.2% were grown in SNNP'RS, Oromia and Gambela regions respectively. It is a major crop for peoples indigenous to southern Ethiopia (Kippie, 2002).

Enset plant is used to make many locally known food products among these are unleavened kocho bread (simply called kocho), bulla porridge (*genfo*), thick cooked bulla gruel (atmit) and a shredded flake made of a mixture of kocho and bulla (*firfir*). Kocho, Bulla and Amicho are well known raw materials obtained from the enset plant but the most common is Kocho. All products of enset have low protein content, which results in protein malnutrition and kwashiorkor in the country especially in Enset consumers regions (Kippie, 2002).

Ethiopia is a country, which has high production of Enset plantation. Even though, the production is high, the utilization of Enset plant is significant. Kocho by its self is a good source of starch and provides high calorie. The approximate content of protein per 100 g dry matter kocho is 1.1-2.8g. As many as 7 million people consume the low-protein Enset products as staple or co-staple foods, sometimes with Vitamin A foods but commonly without the needed protein supplement (Asnaketch, 1997). The implication of heavy dependence on these poor nutrition crops may have serious implication on the physical and mental health of the Enset consuming and planting people.

Pulse crops, account for the highest grain production of Ethiopia. Most grown legumes worldwide are fababean, peanut, peas, chickpeas and lentils (Salunkhe and Kadam, 1989). They are the basis of non-meat national dishes, are a vital protein supplement to the cereal diet. Faba beans are good source of protein of adequate nutritional quality. They provide sufficient quantities of essential amino acids. (Frias *et al.*, 2000).

The blending of kocho with different cereals and legumes results in the improvement of its nutritional value with high quality protein from cheap sources could be a major step in the region to avoid diseases caused by protein energy malnutrition. Thus, this research intends to achieve the improvement of nutritional value of kocho by supplementing it with faba bean.

Materials and Methods

Study Area Description

Sample preparation, blending and fermentation were conducted in Wolaita zone of SNNPR, Ethiopia. It is located at a distance of 390 km to southwest from the capital city of the country, Addis Ababa. Laboratory analysis of the product was conducted at Food Science and Process engineering Laboratory of Haramaya University.

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Experimental Design

A factorial design (3x3) (two factors each at three level) was used for this study. The independent variables considered are blending ratios and co-fermentation durations of the kocho-faba bean blend.

Chemical Analysis

Determination of Crude Protein

The protein content of the sample was determined on the basis of total nitrogen content by micro kjeldhal method of crude nitrogen determination (AOAC, 2000) using the official method 979.09. *Nitrogen (%)* = $\frac{v_{HCl} \ln L x N_{HCl} (Ca \ 0.1) x 14.00 x 100}{V_{HCl} (Ca \ 0.1) x 14.00 x 100}$

Where: V_{HCl} is volume of HCl in L was consumed to the end point of the titration, N_{HCl} is the normality of HCl (used often is 0.1N). W₀ is sample weight on dry matter basis and 14.00 is the molecular weight of nitrogen. The % of nitrogen was converted to % of protein by using appropriate conversion factor (% protein = $6.25 \times \% N$ for baked products).

Determination of crude fat

Ether extract as an estimate of crude lipid was determined using soxhlet extraction method (AACC, 2000) official method 30.10. The solvent then was evaporated by heating on a steam bath. The flask containing the extracted fat was dried on a steam bath to a constant weight.

Crude fat percent by weight = $\left(\frac{W_2 - W_1}{W}\right) X 100$

Where: W_1 = weight of the extraction flask (g)

- W_2 = weight of the extraction flask plus the dried
- crude fat (g) W = weight of sample products (g)

Determination of Crude fiber content

The crude fiber content was determined by the non enzymatic gravimetric method as described in AACC (2000) method No. 32-10. The total crude fiber was expressed in percentage as:

Crude Fiber (%) = $\frac{(W_1 - W_2)}{W} \times 100$

Where: W_1 = weight of coarse porosity crucible and

residue before ignition (g).

- W_2 = weight of coarse porosity crucible and
 - residue after ignition (g).

W = weight of sample product (g).

Determination of total carbohydrates

Total carbohydrates were determined by difference

%C = 100 - [%M + %P + %F + %Fb + %A]

Where: C- Carbohydrate content, M- Moisture content, P- Protein content, F- Fat content, Fb- Fiber content and Ash content.

Determination of moisture content

To determine the moisture content of the products, AOAC, (1995) method 925.09B was used. A crucible was dried in an oven at 105° C for 1 hour and placed in desiccators to cool. The weight of the crucible (W₁) was determined. Samples was weighed in the dry crucible (W_2) and dried at 50-60^oC to constant mass for long durations and then finally was dried at 130°C for 1 hr and after cooling in a desiccators to room temperature it was again weighed (W₃). The moisture content was determined using:

Moisture content in percent = $\left(\frac{W_2 - W_3}{W_2 - W_1}\right) \times 100$

Determination of ash content Ash content of the products was determined according to AACC (2000) Method No.08-01. The total ash was expressed as percentage on dry matter basis as:

Total ash (%) =
$$\frac{M_3 - M_1}{M_2 - M_1} \times 10$$

Where: $(M_2 - M_1)$ sample weight in g on dry basis

 $(M_3 - M_1)$ sample weight of ash in g on dry basis.

Data Management and analysis

Triplicate data was subjected to analysis of variance (ANOVA) to investigate the effect of fermentation of kocho and duration of kocho-faba bean blend. The statistical analysis system (SAS Institute and Cary, NC, USA) was used for descriptive statistics of kocho-faba bean blended final product nutritional composition. Duncan's multiple range test (DMRT) was used for multiple mean comparison at probability level of ($p \le 0.05$).

Result and Discussions

Nutritional compositions of Kocho and fababean

The nutritional compositions of kocho and fababean flour used in this study were given in Table 1. The protein content of kocho investigated was 1.95%, and is in agreement with the value (1.95 g/100 g) reported by Pijls *et al.*, (2006). Ash, fiber and fat contents of kocho investigated was in close agreement with the value reported by Pijls *et al.*, (2006), 1.92%, 2.15% and 0.39% respectively. The carbohydrate content of kocho investigated was 82.31%, which is less than with the value (96.50%) reported by Pijls *et al.*, (2006), this may be due to different enset types and sample environment may be used by the researchers.

Parameters	Kocho	Fababean
Moisture content (g)	9.74±0.92	6.97±0.04
Ash (g)	1.92 ± 0.14	4.38 ± 0.02
Fiber (g)	2.15±0.16	7.79±0.01
Protein (g)	1.95 ± 0.05	40.71±0.08
Fat (g)	$0.39{\pm}0.07$	18.44 ± 0.56
Carbohydrate (g) **	82.31±3.26	21.71±0.45
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Values are means \pm *standard deviation,* **=*determined by difference*

Effect of blending ratios and fermentation conditions on nutritional composition

The result (Table 2) showed significant (P<0.001) difference on moisture content for each of the blending ratios and fermentation conditions. The moisture content of the blend decreased from 12.62% of separately fermented (control sample) kocho flour to 9.64% of the 25% blend with increasing sample proportion of fababean. This is due to the lower amount of moisture content in fababean than in kocho (Table 1). Fermentation conditions also caused difference in moisture content varying from 9.74% recorded for sample where fababean flour was blended at the end of kocho fermentation to 10.06% of the blended done on the 15th day of fermentation.

There was a considerable effect of fababean proportion on protein content (P<0.001). Table 2 shows with increasing the fababean proportion from 0 to 25%, the protein value of blend products increased from 1.95% to 12.50%. The change in protein value was large along the blend with increasing fababean proportion due to the high amount of protein in fababean and lower amount of protein in kocho (Table 1). Fababean flour is used to increase the protein content of protein-deficient foods (Pardeshi and Chattopadhyay, 2008).

Fermentation conditions showed a change in protein content and the highest value was recorded in the kocho-fababean blends conducted after the kocho was removed from the pit with value of 8.68 g/100 g (Table 2). This is due to the synthesis of certain amino acids in both kocho and fababean by the fermenting microorganisms during the separate fermentation of fababean and kocho rather than the co-fermentation of the blends. Fermenting microorganisms can synthesize certain amino acids and improve protein quality (Blandino *et al.*, 2003).

Fermentation conditions did not affect fat content. Fat content is not affected by fermentation of cereallegume food blends (Sharma and khetarpaur, 1997). Table 1 shows a significant difference (P<0.001) for the blending ratios on fat content. The fat content of the blend increased with increasing the proportion of fababean (Table 2). Addition of fababean flour to kocho increased the fat content. It increased from 0.39% in the control kocho sample to 1.51% in the 5% blend, 3.04% in the 15% blend and 5.04% in the 25% blend.

The crude fiber content was significantly different (P < 0.001) for each of the blending ratios and fermentation conditions. Fermentation conditions showed an effect on crude fiber content of the blends that results 2.46% for addition of fababean flour to kocho on the 1st day of processing and 2.82% for addition of fababean to kocho after the kocho was removed from the pits at the 30th day (Table 2). This is due to the solubilization of fiber by microbial enzymes. The results of this study were in accordance with those reported for cereal-legume based food mixtures fermented with *L. acidophilus* (Binita and kumar, 1996). The crude fiber content of the blends increased from 2.02% of sole kocho to 2.88% with addition of 25% fababean since fababean has high crude fiber content than kocho (Table 1). Addition of 5% and 15% fababean resulted in increasing the fiber content to 2.39 and 2.77%.

Table 2 shows no considerable difference on ash content due to fermentation conditions. This is due to the insignificance effect of fermentation on ash content. Ash content is not affected by fermentation of food mixtures (Binita and kumar, 1996). Ash content of the blends was significantly affected (P<0.001) due to blending ratios. Ash content of the products showed high score with increasing fababean proportion from 1.77% for (control kocho) to 2.37% for 25% fababean blend, this may be due to the high amount ash content in fababean than in kocho (Table 1).

It can be seen in Table 2 that significant difference (P<0.001) for each of the blending ratios and fermentation conditions on carbohydrate content. Fermentation conditions showed a slight change on carbohydrate

content that shows a decrement from 73.71% for the addition of fababean flour to kocho at the 1st day of processing to 73.42% that was blended after the 30th day of processing. This may be caused be an increment of the protein and fiber content since carbohydrate is calculated by difference. The carbohydrate content of the blends decreased from 81.25% of kocho to 67.57% for 25% fababean blend; this is due to the higher carbohydrate content of kocho than in fababean (Table 1).

FC	MC (g)	Protein (g)	Fat (g)	CHO** (g)	Fiber (g)	Ash (g)
F_1	10.00 ± 2.15^{b}	8.42±3.51°	3.19±1.50 ^a	73.71±5.56 ^a	2.46±0.28°	2.15±0.17 ^a
F ₁₅	10.06±1.61 ^a	8.51±3.42 ^b	3.22±1.47 ^a	73.33±5.01°	2.74 ± 0.40^{b}	2.16±0.18 ^a
Fx1	9.74±1.69°	8.68±3.51ª	3.23±1.50 ^a	73.42±5.59 ^b	2.82±0.47 ^a	2.15±0.18 ^a
DMRT (P=0.05)	***	***	NS	**	***	NS
CV (%)	0.49	2.20	0.84	0.32	2.56	1.09
BR (g)						
F _{x2}	12.62±0.05ª	1.95 ± 0.02^{d}	$0.39{\pm}0.03^{d}$	81.25±0.06 ^a	$2.02{\pm}0.03^{d}$	1.77 ± 0.03^{d}
B_1	10.35 ± 0.81^{b}	4.26±0.24°	1.51±0.03°	79.51±1.95 ^b	2.39±0.18°	1.98±0.06°
B_2	9.81±0.93°	$8.84{\pm}0.40^{b}$	3.04 ± 0.04^{b}	73.43±1.83°	2.77 ± 0.35^{b}	2.16 ± 0.08^{b}
B ₃	9.64 ± 0.69^{d}	12.50±0.20 ^a	5.04±0.02 ^a	67.57±2.16 ^d	2.88±0.45ª	2.37±0.06ª
DMRT	***	***	***	***	***	***
(P=0.05)	101 101 101			-114 4 4	-110 -	101 101 10 ⁴
CV (%)	0.49	2.20	0.84	0.32	2.56	1.09

Table 2. Effect of blending ratios and fermentation conditions on nutritional composition per 100 g.

Values are Means \pm standard. Values followed by different letters with in a column indicate significant difference $(P < 0.05")^*$, **= value is significantly different at P = 0.01 and ***= value is significantly different at P = 0.001, NS = Non-significant. Note FC=Fermentation Conditions $(F_1, F_{15} \text{ and } F_{x1} \text{ are fababean addition at } 1^{st}, 15^{th} \text{ and } 30^{th}$ day of processing respectively, BR=Blending Ratios (fababean proportion $(B_1 = 5, B_2 = 15 \text{ and } B_3 = 25 \text{ g} \text{ fababean flour}/100 \text{g}$ blend flour, F_{x2} = sign indicates without fababean addition that was used as a control)), MC=Moisture Content, CHO** = Carbohydrate (**determined by difference), CV= Coefficient of Variance and DMRT=Duncan's multiple-range test

Interaction effects of Fermentation conditions and blending ratios on nutritional composition

Variation for moisture content was highly significant (p<0.001) due to the interaction of fermentation conditions and blending ratios (Table 3). The lowest (9.57%) and the highest (11.02%) moisture content was found in 25% fababean blend which was blended on the 15th day of fermentation and in 5% fababean blend which was blended on the 15th day of fermentation, respectively.

There was a significant (p<0.01) effect on crude protein content due to the interaction of fermentation conditions and blending ratios. The results represented that the highest (12.60%) protein content was found in 25% fababean blend which was blended after the kocho was removed from the pit on the 30th day of fermentation and the lowest (4.13%) value was obtained in 5% fababean blend which was blended on the 1st day of processing which was subjected to the longest fermentation. The protein content increased from 1.95% (the control) to 12.47%, 12.60% and 12.45 (%) for 25% fababean blend which was blended on the 15th day of kocho fermentation, after the kocho was removed from the pit after the 30th day of fermentation and on the 1st day of processing for backed kocho products, respectively (Table 3). The increase in protein content of the blend with increasing fababean proportion is due to the high amount of protein content (40.71%) in fababean than kocho as shown in Table 1. Addition of fababean flour to low protein foods has been known to increase the protein content of protein-deficient foods (Otegbayo *et al*, 2002).

There was a significant (p<0.001) effect on fat content due to the interaction of fermentation conditions and blending ratios (Table 3). The highest (5.04%) fat content was found in 25% fababean blend which was blended after the kocho was removed from the pit on the 30^{th} day of fermentation. The lowest (1.50%) value was obtained in 5% fababean blend which was blended on the 1^{st} day of processing and subjected to the longest fermentation. Blending increased fat content since fababean has high fat content than in kocho (Table 1).

The interaction of fermentation conditions and blending ratios had significant effect (p<0.001) on fiber content of kocho based products (Table 3). The highest fiber content of 3.19% was recorded in 25% fababea blend which was blended on the 15th day of kocho fermentation. The lowest fiber content of 2.29% were observed in 5% fababean blend which was blended on the 1st day of processing which was subjected to the longest fermentation(Table 3).

Fermentation conditions and blending ratios interaction had a significant difference (p < 0.01) on ash content (Table 3). The highest ash content of 2.38% was for 25% fababean blend which was blended on the 15^{th} day of fermentation and subjected to underground fermentation. The lowest ash content of 1.99% was resulted in

5% fababean blend which was blended on the 1st day of processing which was subjected to the longest fermentation (Table 3). Blending has the higher effect on the ash content of kocho-faba bean-wheat blend biscuit (Kalekristos, 2010).

The lowest carbohydrate content of (67.12%) was observed in 25% fababean blend which was blended after the kocho was removed from the pit on the 30th day of fermentation. The highest carbohydrate content of (79.86%) was found in 5% fababean blend which was blended after the kocho was removed from the pit on the 30th day of fermentation (Table 3). It can be seen in Table 3 that the kocho bread shows high amount of Carbohydrate (83.98%). Carbohydrate content of kocho-based product was increased with increasing fababean ratios, since fababean has lower amount of carbohydrate content than kocho (Table 1).

Table 3. Interaction effects of fermentation conditions and blending ratios on nutritional composition per 100 g.								
FC	BR	M.C (g)	Protein (g)	Fat (g)	CHO** (g)	Fiber (g)	Ash (g)	
F_1	B_1	10.23±2.46 ^b	4.13 ± 0.40^{d}	1.50±0.02 ^e	79.86±1.14 ^b	2.29±0.27 ^{fe}	1.99±0.06°	
F_1	B_2	10.12±2.41°	8.69±0.10 ^c	3.04 ± 0.03^{b}	73.55±1.01°	2.49±0.02 ^{cde}	2.11 ± 0.07^{b}	
F_1	B_3	9.65 ± 1.90^{f}	12.45±0.21ª	5.03±0.03ª	67.88 ± 0.89^{d}	2.61±0.03 ^{cde}	2.37 ± 0.06^{a}	
F ₁₅	B_1	11.02 ± 0.68^{a}	4.34 ± 0.02^{d}	1.52±0.02 ^e	78.76 ± 0.27^{b}	2.37±0.06 ^e	1.99±0.07°	
F15	B_2	9.59±1.84 ^g	8.71±0.10 ^c	3.07 ± 0.02^{b}	73.85±0.66°	2.67±0.35 ^{de}	2.11±0.09 ^b	
F ₁₅	B_3	9.57±1.81 ^g	12.47±0.05 ^a	5.02 ± 0.04^{b}	67.36 ± 0.80^{d}	$3.19{\pm}0.04^{a}$	$2.38{\pm}0.06^{a}$	
F _{x1}	B_1	9.80±1.93 ^d	4.32±0.03 ^d	1.50±0.02 ^e	79.89 ± 0.77^{b}	2.51±0.02 ^{cde}	1.98±0.07°	
F _{x1}	B_2	9.73±1.81 ^e	9.13±0.61 ^b	3.01 ± 0.02^{d}	73.19±0.59 ^b	2.83 ± 0.70^{bc}	2.11±0.09 ^b	
F _{x1}	B_3	9.71±1.66 ^e	12.60 ± 0.26^{a}	5.04±0.03ª	67.12±1.09 ^d	3.14±0.17 ^{ab}	2.37±0.61ª	
F _{x2}	-	$9.80{\pm}3.08^{d}$	1.91±0.05 ^e	$0.39{\pm}0.02^{f}$	83.98±2.99ª	$2.02{\pm}0.03^{f}$	1.89±0.13°	
DMR								
(P=0.0)		***		***		***	**	
P=0.055			***		***			
CV		0.49	2.20	0.84	0.32	2.56	1.09	

Values are Means \pm standard deviation. Values followed by different letters with in a column indicate significant difference (P < 0.05'')*, ***= value is significantly different at P= 0.001. Note: FC=Fermentation Conditions (F₁, F₁₅ and F_{x1} are fababean addition at the 1st, 15th and 30th day of processing respectively, BR=Blending Ratios (fababean proportion (B₁= 5, B₂= 15 and B₃= 25 (g) per 100g kocho flour)), MC= Moisture Content, CHO **= Carbohydrate (**determined by difference), CV= Coefficient of Variance and DMRT= Duncan's multiple-range test

Conclusions

The study clearly showed that blending ratios and fermentation conditions had effect on the quality of kochofababean blended product. Fababean with proper treatments like blanching and fermentation is suitable for blending with low protein containing foods like kocho to improve nutritional value. Fermentation of kochofababean blends had improved the availability of nutrient for the benefit of the consumer. Generally the present result suggests blending 15% fababean with 85% kocho on the 15th day of kocho fermentation and subjecting the blend to underground fermentation shows better results for increased nutritional content of kochofababean blended products is the best recommended for application.

Recommendations

• Nutritional problems in the developing countries have frequently been identified as protein energy malnutrition. It is advised that kocho, normally known for its protein deficiency, be blended with fababean to solve the problem.

- Effect of blending ratios and fermentation conditions on the status of microbial activity need to be studied.
- Other processing methods and equipments (improved) for kocho-based bread deserve to be studied.

• Industrial utilization of enset for manufacture of convenient local recipes including fortified products deserves to be studied and promoted.

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