

Organoleptic And Nutritional Assessment Of Soybean Tempe

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Abstract: This study investigated the organoleptic and nutritional components of soybean tempe. Three different tempe were produced which included tempe inoculated with *Rhizopus oryzae* (Tempe A) and tempe fermented with *Rhizopus oligosporus* (Tempe B) and tempe without inoculation of a fungus. The organoleptic attributes of the tempe snacks produced from soybean in this study revealed that texture was the least quality attribute preferred by the panelists with the lowest scores ranging from (1.98 ± 0.12 to 2.48±0.13) while the taste was most preferred by the panellists with the highest scores (3.38±0.11 and 2.45 ± 0.13^b). Hedonic scale rating of the organoleptic evaluation of tempe fermented with *R. oligosporus* showed that the colour, taste, texture, aroma and general acceptability of tempe fermented with *R. oligosporus* ranged from 2.48±0.13 - 3.38±0.11 while that of tempe fermented with *R. oryzae* ranged from 1.98±0.12 - 2.45±0.13 The nutritional assessment of tempe revealed the nutritional information of soybean tempe. However, considering the proximate composition of tempe, tempe inoculated with *R. oligosporus* (Tempe B) had significantly higher protein content (40.8%) than its corresponding tempe inoculated with *R. oryzae* (Tempe A (38.92%)) and soybean without inoculation (Tempe C (38.90%).

Keywords: Organoleptic, Nutritional properties, Tempe and *Rhizopus oligosporus*

I. INTRODUCTION

Tempe is characteristically fermented soybean (*Glycine max* Linn) meal that is produced through fermentation using the mould *Rhizopus oligosporus*. Tempe has white to cream appearance caused by growth of the mould's mycelia. Nout and Kiers (2005) confirmed that mycelia bond soybeans and this form a compact texture known as cake (tempe). Tempe involves both innate culturing and batch fermentation which helps to bind the soy bean particles to form a cake. Fermented foods have played an important role in human health for hundreds of years; either from plant or animals as they form part of human diet both in the developed and developing countries of the world. These have played relevant roles in the socio economy less developed nations. Most countries have different fermented foods which form the staple diets and the raw materials that are present in that country. This contributes mainly to the protein consumed by less developed nations

most especially the rural areas. Most indigenous fermented foods are usually prepared at the household level (Egwim *et al.*, 2013).

Tempe fermentation process and its retention of the whole bean give it a higher content of protein, dietary fiber and vitamins compared to tofu, as well as firmer texture and stronger flavor (Shurtleff and Aoyagi, 2001). Tempe is used worldwide in vegetarian cuisine; some consider it to be a meat analogue (Bennett and Sammartano, 2008). Tempe is a vegetable protein that is also rich in vitamin having a bold and characteristic flavour midway between nuts and mushrooms which is perfect for preparing stew, Pan – tossed Dishes and meatless rissoles, but also excellent when fried, served in a piquant sauce. Tempe is a health promoting food it is rich in nutrient, active substances and as the best source of vitamin B₁₂ in vegetarian diets (Steinkraus, 1996; Shurtleff and Aoyagi, 2001). Tempe has attracted interests in Netherlands and United States of America but, yet to be popular in Nigeria.

The aim of this study was to evaluate the organoleptic and nutritional properties of soybean tempe fermented with *Rhizopus oligosporus*

II. MATERIALS AND METHODS

SOURCES OF MATERIALS

Yellow seeded variety of soybeans (*Glycine max L. Merr*) was purchased from Uchi market in Auchi, Edo state, Nigeria; processed and used as substrates for the production of tempe. The starter culture used (*Rhizopus oligosporus*) was obtained from the Indonesian Embassy at No. 5 Salt Lake street, Maitama –Abuja, Nigeria. *Rhizopus oryzae* was cultured from rice.

LABORATORY PRODUCTION OF TEMPE

In the production of tempe in the laboratory, the method of Dubey, (2010) and Aderibigbe, *et al.* (2010) was adopted where two thousand grammes (2000 g) of clean dried yellow seeded soybeans were weighed and soaked for 24 h. This was de-hulled manually by rubbing the seed coats from soaked soya beans, subsequently, the steeping water was discarded and the soybeans were washed in fresh tap water. The beans were subjected to heat by autoclaving at a temperature of 110°C for 10 minutes in fresh water at a bean: water ratio of 1: 3; these were allowed to cool and air dried for 45 mins by spreading on a mesh. This was then inoculated with spores' suspension from *Rhizopus oligosporus*, *Rhizopus oryzae* grown on malt extract agar and incubated at 37°C for 72 h. The produced tempe was dried in a hot air oven at 70°C and milled for analysis.

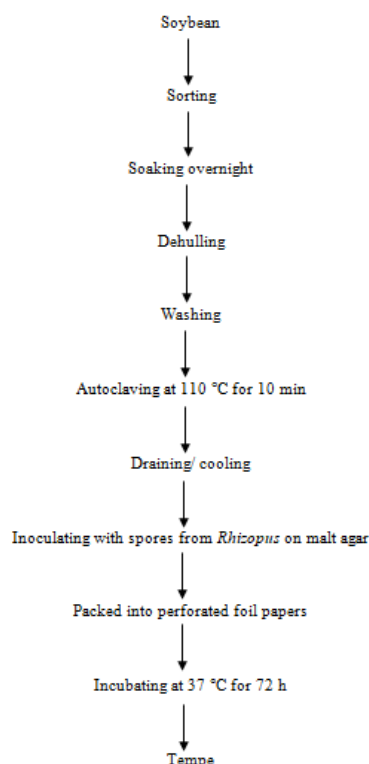


Figure 1: Production of Tempe in the laboratory

PRODUCTION OF TEMPE SNACKS

The tempe produced were prepared using the method of Aderibigbe *et al.* (2010). The tempe was steamed, mixed with spices, kneaded and fried in vegetable oil.

SENSORY EVALUATION

Sensory evaluation was carried out after processing them into snacks by 10 untrained panelists. A 5- point hedonic scale was used to score the colour, taste, texture, aroma and general acceptability of the products; where numerical values of 1 (dislike extremely) through 5 (like extremely) were given (Stone and Sidel, 1992).

PREPARATION OF EXTRACTS FROM TEMPE

An amount of 1 g of tempe was homogenized with 10 ml of distilled water for 3 min. The solution was filtered through a sterile filter paper and this was refrigerated prior to use.

NUTRITIONAL COMPONENTS

After bringing the samples to uniform size, they were analyzed for moisture, protein, fat, ash, fiber and nitrogen free extract by the methods of AOAC (2016). Six parameters were analyzed and these include: moisture, ash, crude fibre, fat, protein and carbohydrates.

DETERMINATION OF MOISTURE

Moisture was determined by oven drying method. 1.5 g of well-mixed sample was accurately weighed in clean, dried crucible (W_1). The crucible was dried in an oven at 100°C for 6-12 h until a constant weight was obtained. Then the crucible was placed in the desiccator for 30 mins to cool. After cooling, it was weighed again (W_2) (AOAC, 2016).

The percent moisture was calculated by following formula:

$$\% \text{ Moisture} = \frac{w_1 - w_2}{\text{Wt of sample}} \times 100$$

Where

W_1 = Initial weight of crucible + Sample

W_2 = Final weight of crucible + Sample

Note: Moisture free samples were used for further analysis.

DETERMINATION OF ASH

For the determination of ash, clean empty crucible was placed in a muffle furnace at 600°C for an hour, cooled in desiccator and then weight of empty crucible was noted (W_1). One gram of each of 1 sample was taken in crucible (W_2). The sample was ignited over a burner with the help of blowpipe, until it is charred. Then the crucible was placed in muffle furnace at 550°C for 2-4 h. The appearances of gray white ash indicate complete oxidation of all organic matter in the sample. After ashing furnace was switch off. The crucible was

cooled and weighed (W_3) (AOAC, 2016). Percent ash was calculated by following formula:

$$\% \text{ Ash} = \frac{\text{Difference in weight of ash} \times 100}{\text{Wt of sample}}$$

Difference in wt. of Ash = $W_3 - W_1$

DETERMINATION OF CRUDE PROTEIN

Protein in the sample was determined by Kjeldahl method. The samples were digested by heating with concentrated sulphuric acid (H_2SO_4) in the presence of digestion mixture. The mixture was then made alkaline. Ammonium sulphate thus formed, released ammonia which was collected in 2% boric acid solution and titrated against standard HCl. Total protein was calculated by multiplying the amount of nitrogen with appropriate factor (6.25) and the amount of protein was calculated. Protein in the sample was determined by Kjeldahl method. 0.5-1.0 g of dried samples was taken in digestion flask. Ten to fifteen ml of concentrated H_2SO_4 and 8 g of digestion mixture i.e. K_2SO_4 $CuSO_4$ (8:1). The flask was swirled in order to mix the contents thoroughly then placed on heater to start digestion till the mixture become clear (blue green in color). It needs 2 h to complete. The digest was cooled and transferred to 100 ml volumetric flask and volume was made up to mark by the addition of distilled water. Distillation of the digest was performed in Markam Still Distillation Apparatus (Khalil and Manan, 1990). Ten milliliters of digest was introduced in the distillation tube then 10 ml of 0.5 N NaOH was gradually added through the same way. Distillation was continued for at least 10 min and NH_3 produced was collected as NH_4OH in a conical flask containing 20 ml of 4% boric acid solution with few drops of modified methyl red indicator. During distillation yellowish color appears due to NH_4OH . The distillate was then titrated against standard 0.1 N HCl solutions till the appearance of pink color. A blank was also run through all steps as above. Percent crude protein content of the sample was calculated by using the following formula:

$$\% \text{ Crude Protein} = 6.25 \times \% \text{ N} \text{ (*Correction factor)}$$

$$\% \text{ N} = \frac{(S - B) \times N \times 0.014 \times D \times 100}{\text{Wt. of the sample} \times V}$$

Where

S = Sample titration reading

B = Blank titration reading

N = Normality of HCl

D = Dilution of sample after digestion

V = Volume taken for distillation

0.014 = Milli equivalent weight of Nitrogen

DETERMINATION OF CRUDE FAT

Dry extraction method for fat determination was implied. It consisted of extracting dry sample with some organic solvent, since all the fat materials for example fats, phospholipids, sterols, fatty acids, carotenoids, pigments, chlorophyll and so on are extracted together therefore, the results are frequently referred to as crude fat. Fats were determined by intermittent soxhlet extraction apparatus. Crude fat was determined by ether extract method using Soxhlet apparatus. Approximately 1 g of moisture free sample was

wrapped in filter paper, placed in fat free thimble and then introduced in the extraction tube. Weighed, cleaned and dried the receiving beaker was filled with petroleum ether and fitted into the apparatus. The water heater was turned on to start extraction. After 4-6 siphoning, ether was allowed to evaporate and beaker disconnected before last siphoning. Extract was transferred into clean glass dish with ether washing and evaporated ether on water bath. Then placed the dish in an oven at $105^\circ C$ for 2 h and cooled it in a desiccators (AOAC, 2016). The percent crude fat was determined by using the following formula:

$$\% \text{ Crude Fat} = \frac{\text{wt of ether extract} \times 100}{\text{Wt of sample}}$$

DETERMINATION OF CRUDE FIBER

A moisture free and ether extracted sample of crude fiber made of cellulose was first digested with dilute H_2SO_4 and then with dilute KOH solution. The undigested residue collected after digestion was ignited and loss in weight after ignition was registered as crude fibre (AOAC, 2016). Zero point one five three grammes sample (W_0) was weighed and transferred to porous crucible. Then the crucible was placed into Dosi-fiber unit and the valve was kept in "OFF" position. After that, 150 ml of preheated H_2SO_4 solution was added and some drops of foam-suppresser to each column. The cooling circuit was opened and turned on the heating elements (power at 90%). When it started boiling, reduced the power at 30% and left it for 30 min. Valves were opened for drainage of acid and rinsed with distilled water thrice to completely ensure the removal of acid from sample. The same procedure was used for alkali digestion by using KOH instead of H_2SO_4 . The sample was dried in an oven at $150^\circ C$ for 1 h. This was then allowed the sample to cool in a desiccator and weighed (W_1). The sample crucible was kept in muffle furnace at $55^\circ C$ for 3-4 h. The sample was cooled in desiccator and weighed again (W_2).

Calculations were done by using the formula:

$$\% \text{ Crude Fibre} = \frac{W_1 - W_2 \times 100}{W_0}$$

STATISTICAL ANALYSIS

All data collected were analyzed statistically using analysis of variance (ANOVA) and Duncan multiple range test. The mean, standard deviation and standard error were determined and least significant difference (LSD) at $p \leq 0.05$ was used to separate the mean using SPSS.

III. RESULTS

The organoleptic properties of the tempe snacks produced from soybean are presented in Table 1. Tempe fermented with *Rhizopus oligosporus* had a more acceptable colour, taste, aroma and general acceptability when compared to tempe fermented with *Rhizopus oryzae* and that without fungus inoculation. The tempe snacks produced is as shown in plates 1 and 2 respectively.

The nutritional components of the tempe is presented in Table 2. The nutritional components of tempe inoculated with *Rhizopus oligosporus* was highest, followed by tempe inoculated with *Rhizopus oryzae* and soybean without inoculation had the least with the exception of carbohydrate where the soybean without any inoculation had the highest value.



Plate 1: Fried Tempe snacks fermented with *Rhizopus oligosporus* used for organoleptic assessment



Plate 2: Fried Tempe snacks fermented with *Rhizopus oryzae* used for organoleptic assessment

Parameters	Value (%)		
	Tempe A	Tempe B	Tempe C
Moisture	5.13± 0.17 ^a	5.13±0.17 ^a	5.10 ± 0.15 ^b
Ash	3.51± 0.14 ^a	3.16±0.13 ^b	2.98 ± 0.15 ^c
Crude fibre	5.26± 0.14 ^b	5.43±0.16 ^a	5.35 ± 0.14 ^a
Fat	10.36 ±0.16 ^b	10.74±0.11 ^a	10.56 ± 0.13 ^a
Protein	38.92± 0.12 ^b	40.83±0.18 ^a	38.90 ± 0.11 ^b
Carbohydrate	35.80 ± 0.12 ^b	35.71± 0.17 ^b	36.85±0.16 ^a

Mean within the row with the same superscript are not significantly different at $p \geq 0.05$.

“±” represent SD

Key: Tempe A = Tempe inoculated with *Rhizopus oryzae*.
Tempe B = Tempe inoculated with *Rhizopus oligosporus*.
Tempe C = Soybean without fungus inoculation.

Table 2: Nutritional components of tempe from soybean fermented with test fungi and control

IV. DISCUSSION

The organoleptic attributes of the tempe snacks produced from soybean in this study revealed that texture was the least quality attribute preferred by the panelists with the lowest scores ranging from (1.98 ± 0.12 to 2.48±0.13) while the taste was most preferred by the panelists with the highest scores (3.38±0.11 and 2.45 ± 0.13^b). Hedonic scale rating of the organoleptic evaluation of tempe fermented with *R. oligosporus* showed that the colour, taste, texture, aroma and general acceptability of tempe fermented with *R. oligosporus* ranged from 2.48±0.13 - 3.38±0.11 while that of tempe fermented with *R. oryzae* ranged from 1.98±0.12 - 2.45±0.13. However, the rating showed that the tempe fermented with *R. oligosporus* was more preferred because of the role *R. oligosporus* play in flavour development, substrate acidification and the safety of the tempe. This is in agreement with the reports of organoleptic attributes of tempe reported by Aderibigbe *et al.* (2010), Roubos van de Hill *et al.* (2010) and Hidayah *et al.* (2012). The nutritional assessment of tempe revealed the nutritional information of soybean tempe. However, considering the proximate composition of tempe, tempe inoculated with *R. oligosporus* (Tempe B) had significantly higher protein content (40.8%) than its corresponding tempe inoculated with *R. oryzae* (Tempe A (38.92%)) and soybean without inoculation (Tempe C (38.90%)). This invariably means that fermentation tends to increase the protein content of the product. This is in agreement with the studies of Messina, (1999), Aderibigbe *et al.* (2010) and Hidayah *et al.* (2012). Tempe is a health-promoting food that is rich in nutrient, active substances as the best source of vitamin B12 in vegetarian diets (Steinkraus, 1996). Tempe as a protein-rich food could play a role as a source of high protein food where protein deficiency is a problem most especially in the North-East part of Nigeria (Aderibigbe and Akindele, 2004). Soybean without inoculation (Tempe C) had the highest carbohydrate content

Sample	Colour	Taste	Texture	Aroma	General Acceptability
A	2.18± 0.15 ^b	2.45± 0.13 ^b	1.98± 0.12 ^b	2.40± 0.14 ^b	2.43 ± 0.15 ^b
B	2.85± 0.15 ^a	3.38± 0.11 ^a	2.48±0.13 ^{ab}	2.90± 0.12 ^a	3.05± 0.11 ^a

Columns with different superscripts are significantly different ($P \leq 0.05$)

Sensory scores: Each value is the mean of 10 panelists.

Key: A = Tempe fermented with *Rhizopus oryzae*. B = Tempe fermented with *Rhizopus oligosporus*.

Table 1: Organoleptic attributes of soybean tempe snacks

(36.9%) than the corresponding soybeans inoculated with *R. oligosporus* (Tempe B) (35.7%) and *R. oryzae* (Tempe A) (35.8%). Carbohydrates in soybeans comprise mainly cell wall polysaccharides and the small sugars: fructose, raffinose and stachyose which are reduced during soaking, cooking and fermentation of the soya beans. This is an indication that fermentation affects reducing sugars that are present in soybean very likely that the organisms involved in the fermentation process may have used up the sugars resulting in a decrease in the sugar content. These findings agree with other reports of Mulyowidaro *et al.* (1991); Steinkraus (1996); Egounlety and Aworh, (2003); Aderibigbe *et al.* (2010). The fat contents of the various tempe were $10.4 \pm 0.16\%$ for tempe fermented with *R. oryzae* (Tempe A), $10.7 \pm 0.11\%$ for tempe fermented with *R. oligosporus* (Tempe B) and $10.6 \pm 0.13\%$ for soybean without inoculation (Tempe C). The obtained value for the crude fibre of tempe fermented with *R. oryzae* (Tempe A) was $5.3 \pm 0.14\%$, tempe fermented with *R. oligosporus* (Tempe B) had a value of $5.4 \pm 0.16\%$, soybean without inoculation (Tempe C) had a value of $5.4 \pm 0.14\%$. This is indicative of the fact that fermentation of soybean into tempe by *R. oligosporus* has significantly increased the fibre content of the product which makes absorption and digestion of the product in the gastrointestinal tract easy. This is in agreement with the reports of Kiers *et al.* (2003) and Hidayah *et al.* (2012). The obtained values for the moisture of tempe fermented with *R. oryzae* (Tempe A) was $5.1 \pm 0.17\%$; that of tempe fermented with *R. oligosporus* (Tempe B) had a value of $5.1 \pm 0.17\%$ and soybean without inoculation (Tempe C) had a value of $5.1 \pm 0.15\%$ which shows that there was no significant difference ($p \leq 0.05$) between the tempe concerning moisture content. This was because they were all in dried form and fermentation did not contribute to high moisture content.

Tempe is usually divided into three categories after production based on its quality: good, unfinished and inedible tempe. Good tempe include beans that are bound into a firm, compact cake by a dense, uniform, white mycelium which should permeate the entire cake. Furthermore, the beans should be barely visible. The entire tempe should lift as a single, cohesive cake without crumbling when shaken gently. Although, tempe is not popular among Nigerians, the sensory evaluation was carried out in a locality in Edo State (Auchi). The panelists accepted tempe snacks from soybean. The panelists particularly preferred tempe snacks from soybean fermented with *R. oligosporus* which were described by some of them as a snack with a pleasant, clean, subtly sweet aroma that resembles that of mushrooms. This is consistent with all the attributes of a good tempe as documented by Shurtleff and Aoyagi (2001).

V. CONCLUSION

In this study, *R. oligosporus* and *R. oryzae* were used as starter cultures for tempe productions. The tempe produced with these fungi (*R. oligosporus* and *R. oryzae*) had all the attributes of a good tempe. Tempe produced in this study revealed significant nutritional components that are good for human health and maintenance. There was no much difference in the nutritional composition of the tempe fermented with *R.*

oligosporus and *R. oryzae* apart from the fact that the tempe fermented with *R. oligosporus* was higher in its protein content than the tempe fermented with *R. oryzae*.

VI. RECOMMENDATION

Inclusion of soybean tempe in our diets as a plant protein-rich food which will help to solve the problems of protein-calorie malnutrition especially in internally displaced persons' camps in North East part of Nigeria.

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