Effects of fermentation time on the chemical composition of condiment (*Ogiri-Asala*) from Conophour nut (*Tetracarpidium conophorum*)

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ABSTRACT

Oilseeds are a crucial source for plant-based proteins, especially in developing countries where protein intake is often insufficient. The conophor nut, though frequently overlooked, is rich in high-quality protein and oil. The study investigated the impact of fermentation on the chemical composition of *Ogiri-asala*, a local condiment produced from conophor nut. Conophor nuts were processed and fermented for varying durations (48, 72, and 96 h) to produce *ogiri-asala.* Proximate composition, mineral contents, amino acid profile, antinutritional factors and sensory acceptability of the samples of *ogiri-asala* were evaluated. Results showed that fermentation significantly increased protein content from 26.71% to 30.96%. Fermentation also reduced anti-nutrients which included phytic acid, tannin, saponin, and trypsin inhibitor. There was a notable increase in mineral elements such as calcium (325 to 395 ppm), magnesium (385 to 413 ppm), iron (6.6 to 14.0 ppm), and manganese (15.9 to 21.0 ppm). The sample fermented for 96 h (sample HYF) showed the highest value of protein contents, highest macronutrient levels and lowest antinutritional factors. The overall indispensable amino acids and overall amino acids ratio of all the samples (36.46%, 37.50%, 37.62% and 38.01%) were below 39%, nonetheless, sufficient only for children and adult and not infants. The sample fermented for 96 h (sample HYF) was most accepted by the panelists, in terms of aroma, taste and texture. In conclusion, fermentation enhances the nutritional values of conophor nut and the *Ogiri-asala* produced could serve as an alternative to the conventional ogiri.

KEYWORDS

Conophor nut
Fermentation
Antinutritional factor
Ogiri-asala

1. INTRODUCTION

Fermentation is defined as biological process that produces metabolites through biosynthesis and modification of food nutrients. Fermentation process enhance nutrient composition, flavour, safety, taste, shelf- life, and reduces antinutrient composition of the fermented foods. (Adebiyi *et al.*, 2021). For centuries, alkaline-fermented foods have been a key component of the diet and as a flavorful food condiment in many African indigenous communities, valued for their nutritional benefits, safety, and potential for commercialization (Dabiré *et al.*, 2022). Food condiments are made from fermented seeds of different leguminous and oilseeds plants and are used for flavouring soups and sauces (Okolie et al., 2023).

The conophor plant (*Tetracarpidium conophorum),* which is extensively grown for its healthy fruit-nuts, typically prepared and eaten alongside cooked maize throughout the period of harvest (Oguntoyinbo *et al*., 2023). Tetracarpidium *Conophorum* is an edible nut that is consumed in between meals and is referred to as Kaso or Ngak by local customers in Western Cameroon and it is called Awusa or Asala in the southwest of Nigeria. (Adeyemi *et al*., 2023). It has been discovered that the nuts have significant amounts of macronutrients, such as crude fibre, protein, fat and oil, carbohydrates (Gbadamosi *et al*., 2020). The fruit is known for having a significant amount of oil that contains a lot of polyunsaturated acid particularly linolenic acid making it unstable when use for frying (Oguntoyinbo *et al*., 2023; Oyinloye and Enujiugha, 2017). Several studies have reported the application of the nuts. For instance, Baiyeri *et.al*. (2019) reported that the dried walnut kernels could be processed into flour and used as mixed flour for bread making. Similarly, Nwachukwu and Aluko (2021) reported that the protein isolate of Conophor flour can be incorporated into wheat and corn-based biscuits. The inclusion of the Conophor nuts into the ogi mixture was reported by Adisa and Enuijigha (2020).

Many protein-rich oilseeds, such as melon, and fluted pumpkin and castor, have been cooked and fermented to create a local seasoning known as ogiri. The rural population uses it as an inexpensive soup seasoning. Although used in modest amounts, it is essential for meal and soup preparation. It does not only improve the protein content of soups, but also offers health advantages. Ogiri is traditionally made from melon seeds using an uncontrolled solid state fermentation process, followed by another boiling to soften the seeds for fermentation. Nigeria is home to a variety of fermented seeds, including the following like Dawadawa, made from soybeans, Ogiri, made from melon seeds, Une, and Iru, made from locust beans, (Ejinkeonye *et al.*, 2018)

Concerns over the health effects of ingredients in commercial seasonings are discouraging their use. Conophor nut, a seasonally available oilseed that is high in quality protein, is currently underutilized. Previous studies have shown the effect of fermentation on conophor nut, but no study has been conducted on its fermentation in a similar manner to produce a product analogous to *Ogiri-Egusi*. Producing such a product could improve the economic value of the nuts as a healthier alternative to conventional seasonings. It could also increase the variety of traditionally fermented condiments and offers an affordable, readily available source of protein to many low-income families, especially in developing nations. The objectives of the research were to produce conophor (Ogiri-Asala) condiment exploring the traditional fermentation method, and determining the effects of fermentation period on nutritional profile, antinutrients composition and sensory characteristics of the fermented conophor nut.

2. MATERIALS AND METHODS

2.1 Collection and Preparation of the raw Conophor Nuts for Fermentation

Raw conophor nuts were procured in the nearby town of Modakeke, Osun State, Nigeria. The *Ogiri-asala* was produced by adopting the process operations described by Ejinkonye et al., (2018) with minimal ammendments. The nuts were washed with protable water and cooked at 12psi and 121 °C for 30 min in an autoclave. The cooked nuts were cooled to room temperature, carefully shelled to extract the nuts, and then cut into small pieces manually with a knife to about 1 mm thickness. The sliced nuts were divided into four (4) sample parts, one part remained unfermented which served as the control sample for the study, and was dried using a hot air oven (Uniscope Laboratory Oven, England)

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at 60 °C for 8 h. The fermented nuts were dried with a hot air oven (Uniscope Laboratory Oven, England) at 60 °C for 8 h. The samples were milled using maxcella mixer grinder (SR-14733, Marlex, Duncan), sieved (350μm) and then stored in polythene bags at 5 °C until use

2.2 Proximate Composition of Fermented Conophor Flours

The proximate composition (moisture, crude fiber, crude protein, ash, crude fat, and carbohydrate contents) of the fermented and unfermented conophor flour samples was determined using the method of AOAC (2010).

2.3 Moisture content determination

Each sample (5 g) was measured into a porcelain dish of a known weight before being baked dried at 105 °C in a gallenkamp hot air oven to achieve a constant weight. The material was cooled in the desiccator and then weighed. The percentage (%) of moisture content was calculated as follows:

$$\text{Moisture Content}\left(\%\right) = \frac{D_1 - D_2}{D_1} \times 100$$

D_1 corresponds to initial weight of sample measured before the drying process (g)
D_2 corresponds to initial weight of sample measured after the drying process (g)

2.4 Determination of crude protein

The crude protein content determination employed the modified micro-kjeldahl technique. Weighing 0.2g each sample into a digestion flask. Each flask received an addition of 0.8 g of the Kjeltec catalyst and 15 ml concentrated sulfuric acid. A preheated digester that had been set to 420 °C was used to heat each flask and, was digested until a clear solution was obtained. Following digestion, the flasks were taken out, allowed to cool, then the resulting content was thinned out with a 50 ml of distilled water. Thereafter, 50 ml of 40% NaOH was spontaneously poured into each flask after being put inside the micro-kjedahl analyzer distillation unit. The solution was then reheated to produce ammonia, which was then distilled for about 4 minutes into a conical flask which contains 25 ml of 2% boric acid. During the distillation process, ammonia and boric acid were mixed to create an ammonium borate solution. The distilled solution was then titrated with 0.1 M hydrochloric acid until the solution turned to purple-gray, which indicate end point. The percentage (%) nitrogen was calculated as follow:

$$\text{Nitrogen} = \frac{1.40 \times \text{m(ml titrant} - \text{ml blank)}}{\text{original weight of sample}}$$

Where M refers to the Molarity of the acid, ml titrant indicates the volume (mL) of 0.1 M HCl used in the titration, and ml blank represents the volume (mL) of acid employed for the blank determination.

The protein content was calculated by multiplying the measured nitrogen percentage (%) by a factor of 6.25.

2.5 Crude fat content determination

The Soxhlet extraction apparatus was used to calculate crude fat. Sample (2g) was weighed into a non-fat extraction thimble, closed the thimble with cotton wool, and set it inside the appropriate extractor chamber. About two-thirds of the capacity of previously weighed distillation flask was filled with n-hexane, then heated continuously for four hours on a heating mantle. After that, n-hexane was extracted from the distillation flask and placed in a sterile container until nearly all of it was eventually extracted. The residual solvent was separated by evaporating it in an oven set and regulated at 70 °C. The distillation flask was then permitted to cool inside a dessicator, after which its weight was taken. The change in the weight of distillation flask before and after the process was used to estimate the oil yield of the sample.

$$\text{Crude fat} = \frac{\text{final weight of flask} - \text{initial weight of flask}}{\text{weight of sample}} \times 100$$

2.6 Crude fibre determination

Using what was left from the sample that was used for the extraction process to determine the crude fat, the crude fiber was determined. The sample (2g) was weighed into 500ml beakers and mixed with 200 ml of boiled 1.25% H_2SO_4 and to prevent solids sticking to the edges of the beaker it was subjected to heating for thirty minutes while being turned periodically. After filtering each solution, 50 to 70 ml of distilled water was used to rinse it. The entire residue was taken out, put inside a 500 ml beaker containing 200 ml of 1.25% NaOH was added, and the resulting solution was heated for 30 minutes. Thereafter, the content in each beaker was taken out, filtered and rinsed with a small volume of distilled water sufficient to ensure thorough washing. Thereafter, the residue was put into an ash dish and heated to 130 °C for drying, allowed to cool in a desiccator before being weighed again. After charred for 30 minutes at 600 °C, the residue was re-cooled in the desiccator before being weighed. The proportional percentage of crude fibre in each sample was estimated as:

$$\text{Crude fibre } \% = \frac{\text{weight after drying} - \text{weight after ashing}}{\text{original weight}} \times 100$$

2.7 Total ash determination

In a porcelain crucible dish, each sample around 5 g was weighed, put over a hot plate and the sample was scorched. After that, the dish was positioned for approximately 550 °C in a muffle furnace 2 hours to produce a white-gray ash. The crucible was allowed to cool in a desiccator before it was weighed. The percentage of ash was determined based on the formula below:

$$\text{Ash } \% = \frac{\text{weight of ash}}{\text{original weight of sample}} \times 100$$

2.8 Carbohydrate determination

Difference was used to determine the percentage of carbohydrates; Carbohydrate content (%) = 100% - (percentage addition of moisture content, crude proteins, crude fat, crude fiber, and ash content)

2.9 Determination of Mineral Composition of Fermented Samples

Mineral composition was determined using the method of AOAC (2005). A 5 ml portion of the digestion mixture was mixed with 0.5 g of each sample in separate 75 ml tubes. The tubes were then swirled and placed inside a fume cabinet, where the digestion process was carried out for 2 hours at 150 °C. Thereafter, the samples were taken out of the digester, allowed to cool for ten minutes, 3 mls of 6M HCl were added and mixed. The mixture was allowed to continue the digestion process for an additional one and a half hours. It was then taken out of the digestor, allowed to cool. Afterwards, the samples volume in each digestion tube was made up to 50 ml with distilled water. The concentrations of mineral composition of each sample were determined using a Perkin Elmer 403 type atomic absorption spectrophotometer.

2.9.1 Amino acid Determination

The S433 Amino Acid Analyzer (SYKEM, Germany) was used to determine the amino acid composition. Before hydrolyzing for 24 hours with 6N HCI at 110 °C, samples were freeze-dried (Blackburn, 1978). After hydrolysis, samples were put in a sodium citrate buffer with a pH of 2.2 and kept refrigerated. 50ul of the hydrolysate was taken for analysis. By hydrolyzing the material with sodium hydroxide, tryptophan was independently determined. Cysteine was quantified as cysteic acid and methionine was as well measured as methionine sulphone, after performic acid was oxidized before being hydrolyzed in 6 N HCI (Blackburn,1978).

2.10 Determination of antinutritional factors

2.10.1 Phytate determination

The amount of phytate was assessed using the method outlined by Maga (1983). Each sample of approximately 2g and about 100ml of 2% hydrochloric acid was measured out into a conical flask with a volume of 250 ml. The mixture was allowed to soak for 3 hours and was filtered through double sheets of toughened filter paper. To achieve the required acidity, dilution of 50 ml of each filtrate with 107 ml of distilled water in a 500 ml conical flask was carried out. For each solution, 10 ml of a 0.3% ammonium thiocyanate (NH_4SCM) solution was added as specified. A standard iron (III) chloride solution that contained 0.00195 g of iron per ml was used to titrate the solution. The final point had a five-minute duration of a light brownish-yellow tone. The percentage of phytic acid was determined based on the formula below:

$$\text{Phytic acid } \% = \frac{\text{Titre value} \times 1.19 \times 100 \times 3.55}{\text{weight of sample}}$$

2.10.2 Tannin determination

Tannin content was calculated using the modified method of analysis of AOAC (1984). A 50 ml beaker containing 20 ml of 50% methanol was filled with about 0.20 g of the sample. The beaker was wrapped in para-film and heated for an hour in a bath of water calibrated at 77°C to 80 °C. It was shaken vigorously to achieve consistent mixture. Into a 100 ml volumetric flask, the extract was filtered with double-sheets Whatman No. 41 filter paper. Following the addition of 20 millilitres of water, together with 2.5 ml of the Folin-Denis reagent, and 10 millilitres of 17% sodium carbonate solutionNa_2CO_3 were added and thoroughly mixed. After proper mixing, distilled water was added to the mixture to reach appropriate level, and allowed to stand for 20 minutes for bluish-green colour to develop. The standard tannin solutions 0–10 ppm range were prepared and treated similarly using the procedure as the 1 ml sample described earlier. Absorbance of the reference solutions and tannic acid samples was recorded at 760 nm with a Spectronic 21D spectrophotometer, following the development of colour. The following formula was used to determine the amount of Tannin:

$$\%\text{Tannin} = \frac{\text{absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{weight of sample} \times 10000}$$

2.10.3 Saponin determination

The saponin analysis was conducted using the spectrophotometric technique of Brunner's (1984). About 1 g of the finely powdered flour sample was added to a 250 ml beaker filled with 100ml of isobutyl alcohol. The mixture was agitated for 5 hours on a UDY shaker to achieve even mixture. After the mixture was filtered through Grade 1 Whatman filter paper, 20 millilitres of magnesium carbonate solution at 40% saturation were added into a 100 ml beaker, thereafter the mixture was transferred to a 200 ml volumetric flask and diluted to the appropriate level. The mix containing saturated $MgCO_3$ was subsequently passed through Grade 1 Whatman filter paper, resulting in an uncoloured solution. A 1 ml portion of the uncoloured solution and 2 millilitres of ferric chloride solution at 5% concentration were placed into a volumetric flask of 50 ml capacity, followed by the addition of distilled water to bring the volume to the calibration mark. The mixture was allowed to stand for 30 minutes to allow the hue to turn blood crimson. From a stock solution of saponin, standard saponin solutions (0–10 ppm) were made. As was done for the 1.0 ml sample above, 2 ml of 5% $FeCl_3$ was used to treat the standard solutions. After color evolution, Jenway Spectrophotometer was employed to take the measurements of the absorbance of the blank and the standard saponin solutions at 380 nm wavelength. A plot of absorbance against concentration of standard (ppm) gave a straight line curve, from which the sample concentrations were extrapolated.

2.10.4 Determination of trypsin inhibition activity (TIA)

The procedures described by Arntfield *et al*. (1985) was employed to estimate trypsin inhibitor. In order to extract the samples, 1 g of each sample was weighed and dissolved in 50 ml of a 0.5 M NaCl solution. The liquid was subjected to agitation for 30 minutes at ambient temperature, followed by centrifugation. The obtained supernatant underwent clarification using Grade 1 Whatman filter paper. The determination was done using the filtrate (extract). In a test tube or beaker, 20 ml of 0.1% trypsin solution were mixed to 10 ml of the filtrate. A 10 ml blank of distilled water was prepared. The mixture contents were given a minimum of five minutes to stand before its absorbance of that material at a wavelength of 410 nm was determined. Trypsin inhibition activity was calculated as the quantity of trypsin units inhibited (TIU) per unit weight (g) of the sample analyzed

2.10.5 Sensory analysis

Sensory evaluation using 15 trained panelists was carried out on unfermented, fermented samples as well as commercial egusi ogiri used to prepare egusi soup using preference test. A 7-point hedonic scale was employed, where 1 denoted dislike extremely and 7 denoted like extremely. The panelists were instructed to conduct sensory evaluations of the samples, assessing them for aroma, texture, taste, colour, and overall acceptability.

2.10.6 Statistical analysis

Each experiment was performed in triplicate. Data collected were analysed using Analysis of Variance (ANOVA) using SPSS software (version 20). Mean separation was performed using Duncan's new multiple range test procedure to find out any statistical differences between them.

3 RESULTS AND DISCUSSIONS

3.1 Proximate composition

The proximate composition of the fermented conophor flour samples is displayed in Table 1. There were increments in crude protein concentrations, fat, fiber, ash and moisture content with fermentation time. The protein content of the samples increased from 26.71% to 29.96%. The significant increase ($p < 0.05$) of protein content with fermentation time appears to stem from a complex interplay of biochemical processes. Enzyme synthesis during these processes facilitates the degradation of complex molecules, thereby augmenting protein availability. The reduction of anti-nutritional factors, which can impede protein bioavailability, also contributes to the observed increase in protein content. Furthermore, the generation of novel proteins during germination and fermentation adds to the overall protein yield.

Table 1: Proximate composition (%) of condiment (Ogiri-Asala) from conophor nut

Sample	Moisture	Fat	Ash	Crude protein	Crude fiber	Carbohydrate
BUF	4.360.4[a]	48.220.08[a]	3.160.10[a]	26.710.04[a]	6.090.06[a]	11.460.02[d]
DTF	4.450.05[b]	50.910.07[b]	3.270.15[b]	27.050.01[b]	6.350.02[b]	7.970..05[c]
CJF	4.510.08[b]	53.600.12[c]	3.540.09[c]	27.780.03[c]	6.770.06[c]	6.800.08[b]
HYF	4.680.03	54.900.07[d]	4.030.12[d]	29.960.01[d]	7.130.08[d]	5.670.04[d]

Values reported are means s.d of triplicate determinations
Means values within each column bearing a different superscript roman letter are significantly (p0.05, LSD) different. BUF- Sample at fermentation time 0 h; DTF- Sample at fermentation time 48 h; CJF- Sample at fermentation time 72 h; HYF- Sample at fermentation time 96 h

Ultimately, the proteolytic conversion of insoluble storage proteins into soluble forms enhances protein accessibility (Chinma *et al*., 2020). The obtained value for protein level of fermented conophor nuts was greater than the value reported by Forsido *et al.* (2020), for fermented oat 11.1%-14.4%, fermented barley (8.8%-12.4%) and teff (8.1%-11.8%), respectively, but lower than the value (44.66%-58.40%) for the hazelnut kernel meal as reported by Altop *et al*. (2019) after fermentation for 7 days (168 hours). The higher fat content (48.22%-54.90%) during fermentation could result from the disintegration of complex compounds, causing more lipids to be released and caused microbes like mould and yeast to produce lipids at the same time (Anaemene and Fadupin, 2022). The increment in the fat content as fermentation time progressed in this study aligned with the result of Adegbehingbe and Daramola (2021) on fermented locust beans and fermented lima beans from 21.16%-26.22% and 3.68%-4.74% respectively. The fermentation method of processing significantly increased ($p < 0.05$) the total ash percentage of the fermented samples from 3.16% to 4.43% during fermentation period. The rise in the amount of ash during fermentation resulted from microbial activities breaking down substances and releasing minerals, as well as dismantling complex compounds that previously bound minerals (Adebo *et al.*, 2022). This pattern is consistent with the study by Adebo *et a*l., (2022) which showed that after undergoing various fermentation periods, ash concentration increased in fermented soymilk, mung bean flour, and tamarind seeds. This was, however, contrary to the decrease in values of African locust beans reported by Falade and Akinrinde, (2021) and Oloyede and Akintunde (2019). The crude fibre content increased significantly ($p < 0.05$) as the fermentation time increased (6.09% to 7.13%), and this rise was attributed to the accumulation of both acidic and alkali compounds, which resulted in increased fibre values (Olukomaiya, *et al*., 2020). This aligned with increase in fiber content findings on the nutritional status of fermented *C. cujete* seeds and their potential as aqua-feed (Suleiman, 2019). The moisture content was found to increase with an increase in the fermentation time value from 4.36% to 4.68%. The measured moisture contents were below the 10% benchmark generally cited as necessary to preserve the storage stability of flour. The level of moisture content is an indication of storage stability. (Chinma *et al*., 2020). The low moisture level found in this research indicated the flour samples would be resistant to microbial deterioration. During fermentation, a reduction in carbohydrate content from 11.46% at 0 hours to 5.76% at 96 hours was recorded. This reduction was attributed to the declining carbohydrate ratio of overall mass, leading to the percentages of nutrients to be redistributed (Olagunju *et al.*, 2018)

3.2 Influence of fermentation on mineral composition of conophor nuts

Impact of fermentation period on the mineral content of fermented conophor nut is illustrated in Table 2. Fermentation time significantly ($p < 0.05$) improved the values of magnesium, calcium, iron, manganese and chromium, but resulted in decreased sodium, potassium and zinc. The breakdown of antinutritional compounds and the release of soluble form of bonded minerals during the fermentation process are presum-

Table 2: Mineral composition (ppm) of condiment (Ogiri-Asala) from conophor nut

	Samples			
	BUF	DTF	CJF	HYF
Ca	325±0.02[a]	3430.01[b]	3480.03[b]	3950.01[c]
Mg	3850.01[a]	3950.04[ab]	4030.02[bc]	4130.02[c]
Na	4150.06[c]	3750.03[b]	3700.04[b]	2730.03[a]
K	10080.03[d]	8730.04[c]	8230.1[b]	7630.2[a]
Fe	6. 60.01[a]	9.10.04[b]	9.80.2[a]	14.00.05[c]
Zn	250.01[b]	240.01[b]	210.01[a]	200.01[a]
Mn	15.90.12[a]	18.10.05[b]	18.40.03[b]	21.00.07[c]
Cr	0.20.02[a]	0.50.01[a]	0.60.04[a]	1.00.06[b]
Pb	-	-	-	-
Cu	-	-	-	-

Values reported are means s.d of triplicate determinations
Mean values within each column bearing a different superscript roman letter are significantly (P0.05, LSD)
BUF- Sample at fermentation time 0 h; DTF- Sample at fermentation time 48 h; CJF- Sample at fermentation time 72 h; HYF- Sample at fermentation time 96 h

ably the causes of this increase in mineral content during the fermentation (Chinma *et al*., 2022; Azeez *et al.,* 2022). With increasing fermentation time, minerals level such as Mg, Ca, Mn and Fe also increased (Olagunju *et al*., 2018). Nevertheless, the decrease in minerals during fermentation is probably caused by the minerals being used as co-factors for certain enzymes required in the catalytic processes of macromolecules, as well as some minerals leaching out when the nut seeds are cooked (Bhinder *et al.*, 2021). Upon conclusion of fermentation, potassium was the most abundant macromolecules, followed by magnesium and calcium, and sodium was the least abundant. Manganese has the highest levels of microminerals, followed by zinc and iron, with chromium having the lowest levels. Lead and copper could not be detected in all samples. Lead and chromium concentrations might have been below regulatory or practical detection limits.

3.3 Effect of fermentation time on amino acids profile of conophor nut

The amino acids profile of conophor seeds as influenced by fermentation is shown in Table 3. Fermentation was found to affect the indispensable and dispensable amino acids of conophor seeds differently. Fermentation brought an increase ($p < 0.05$) to all the indispensable amino acids of the conophor nuts except for cysteine, tyrosine, threonine, and valine. This is likely due to the fermentation microbes, particularly lactic acid-producing bacteria, producing essential amino acids (Mohapatra *et al.,* 2019). Conversely, it decreased the amounts of dispensable amino acids except arginine, proline glutamic acid and glycine with fermentation period. The decrease in indispensable amino acids could be linked to microbial metabolism of amino acids during fermentation process (Wu, *et al*., 2022). The overall indispensable amino acids of conophor also improved from 33.850 to 35.403mg/100g upon conclusion of fermentation with 4.59% increase. The overall dispensable amino acids showed a distinct trend, initially decreasing from 59 mg/100g to 55.692 mg/100g after 48 hours of fermentation. This was followed by a slight increase to 56.980 mg/100g at 72 hours, and then another decrease to 57.738 mg/100g after 96 hours with 2.14 % percent increase during the whole fermentation period. The unique pattern in overall dispensable amino acids impacted the overall amino acid contents. Specifically, the overall amino acid content decreased from 92.850 mg/100g to 89.114 mg/100g after 48 hours of fermentation, then rose to 91.348 mg/100g at 72 hours, and finally reached 93.141 mg/100g after 96 hours. The indispensable amino acid to overall amino acid ratio (IAA/TAA) in all samples was below 39%, which falls short of the WHO's recommended level for infants. However, according to WHO standards, which require IAA/TAA ratios above 39%, 26% and 12% for infants, children and adults respectively (Mohammed *et a*l., 2023) the ratios in the cooked and fermented samples were

Table 3: Total amino acid composition (mg/100g) of condiment (Ogiri-Asala) from conophor nut

Amino acids	Samples			
	BUF	DTF	CJF	HYF
Isoleucine	4.226	4.821	4.124	4.348
Threonine	4.182	3.923	4.052	4.001
Leucine	7.014	6.623	7.009	7.000
Valine	3.952	3.358	3.460	3.891
Lysine	1.366	3.006	3.141	3.605
Phenylalanine	4.059	3.920	4.570	4.530
Cysteine	1.492	1.059	1.366	1.366
Methionine	1.632	2.098	2.117	2.103
Total Sulphur amino acids	3.124	3.157	3.483	3.469
Tyrosine	2.600	2.205	1.903	2.160
Histidine	2.240	2.409	2.626	2.399
Overall indispensable amino acids (TIAA)	33.850	33.422	34.368	35.403
Arginine	12.058	12.018	12.827	12.320
Aspartic acid	9.471	8.293	7.502	7.446
Glutamic acid	17.823	17.625	17.492	18.462
Serine	6.028	4.769	5.033	5.137
Proline	3.925	3.589	4.584	4.856
Glycine	5.288	5.151	5.562	5.306
Alanine	4.407	4.250	3.980	4.211
Overall dispensable amino acids	59.000	55.692	56.980	57.738
Overall amino acids (TAA)	92.850	89.114	91.340	93.141
IAA/TAA (%)	36.46	37.50	37.62	38.01

BUF- Sample at fermentation time 0 h; DTF- Sample at fermentation time 48 h; CJF- Sample at fermentation time 72 h; HYF- Sample at fermentation time 96 h

nonetheless sufficient for children and adults.

3.4 Influence of fermentation time on the antinutritional compounds of conophor nut-seeds

Fermentation processing significantly ($p < 0.05$) lowered the phytate, tannin, saponin and trypsin inhibitor contents as shown in Figure 1-4. The highest concentrations of tannin, saponin, phytic acid and trypsin inhibitor were witnessed in the cooked unfermented sample. This means that all antinutritional compounds were reduced during fermentation. The figures showed that the cooked (unfermented) seeds had phytic acid, tannin and saponin and trypsin inhibitor concentrations of 0.058%, 0.037% and 0.029% and 31.42TIU/mg respectively which was drastically reduced and significantly different at the end of 96 h fermentation time with values of 0.008%, 0.011%, 0.013% 19.68TIU/mg for phytic acid, tannin, saponin and trypsin inhibitor respectively.

Specifically, phytic acid levels dropped by 86.21%, tannin by 70.27%, saponin by 55.17%, and trypsin inhibitor by 37.36%.

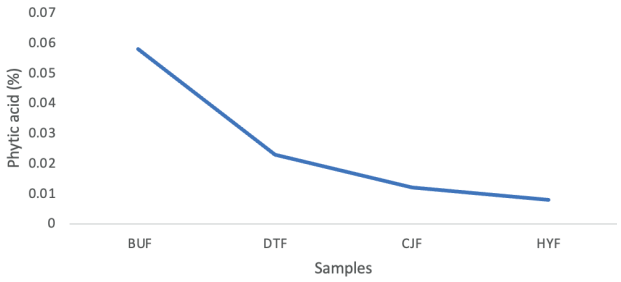

Figure 1 Influence of fermentation on Phytic acid concentration

BUF- Sample at fermentation time 0 h; DTF- Sample at fermentation time 48 h; CJF- Sample at fermentation time 72 h; HYF- Sample at fermentation time 96 h

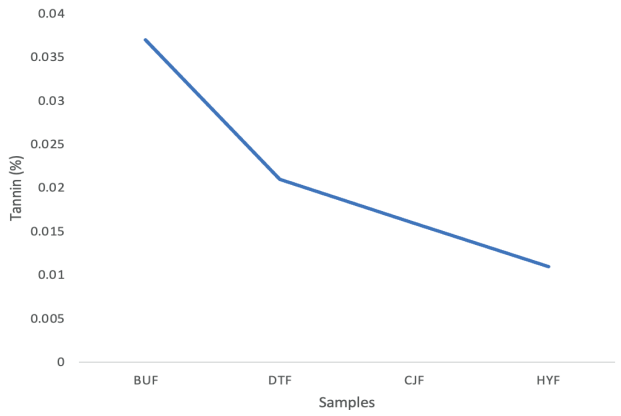

Figure 2 Influence of fermentation on Tannin concetration

BUF- Sample at fermentation time 0 h; DTF- Sample at fermentation time 48 h; CJF- Sample at fermentation time 72 h; HYF- Sample at fermentation time 96 h

The decline in tannin and phytic acid concentrations in the fermented samples was ascribed to the enhanced biochemical action of phytate-degrading and tannin enzymes, including tannin acyl hydrolases, phosphatases, phytases, and polyphenol oxidases, during fermentation. (Sharma *et al*., 2022). Additionally, fermenting bacteria contributed to the decline in phytic acid through their enzymes, which break down phytic acid and its associated complexes (Adebiyi *et al*., 2019). Tannin level was reduced also due to the possibility of the increase in its water solubility (Hawashi *et al.*, 2019). The reduction in trypsin inhibitor was ascribed to enzymatic hydrolysis, which leads to a diminution in trypsin inhibitor activity (Oluseyi and Temitayo, 2015; Adebo *et a*l., 2022). Furthermore reduction in saponin could be ascribed to the enzymatic activity that break down saponins by releasing sugar side chains and reducing their water solubility and content during fermentation (Rui, 2017; Jeyakumar and Lawrence, 2022)

3.5 Sensory acceptability of the conophor ogiri

The sensory evaluation result in Table 4 shows the responses of the panelists to the different sensory attributes of the fermented conophor nut analogue "ogiri". The ogiri from melon sample used as control sample showed no significant differences in sensory attribute evaluated when compared with sample HYF. The statistical mean value for test soup obtained for aroma ranged from 4.05 to 6.60 and the samples were significantly different ($p < 0.05$). The soup prepared with 96 h fermented sample was preferred most for aroma scoring 6.60 while sample BUF was rated low because the distinct ammonia smell was not perceived at all. The preferred sample for colour was BUF

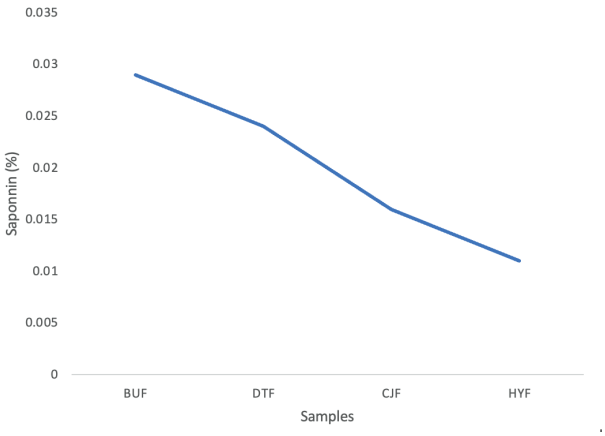

Figure 3 Influence of fermentation time on Saponnin concentration

BUF- Sample at fermentation time 0 h; DTF- Sample at fermentation time 48 h; CJF- Sample at fermentation time 72 h; HYF- Sample at fermentation time 96 h

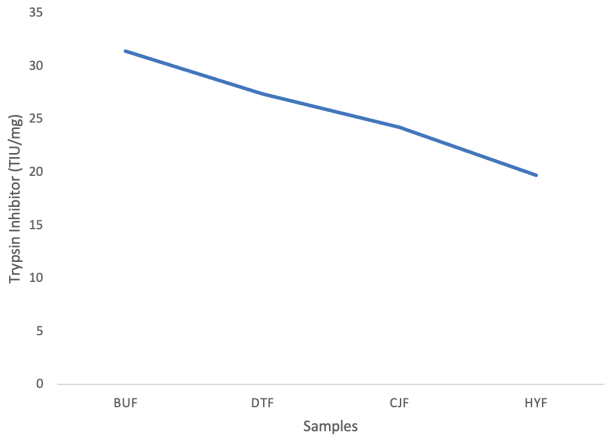

Figure 4 Influence of fermentation time on Trypsin Inhibitor concentration

BUF- Sample at fermentation time 0 h; DTF- Sample at fermentation time 48 h; CJF- Sample at fermentation time 72 h; HYF- Sample at fermentation time 96 h

with a score of 6.73. This was due to the maillard reaction occurring fermentation, potentially making other samples less appealing. There was no significant difference between SMF and HYF sample for taste attribute but SMF was assigned with a score of 6.54. Sample HYF was rated best with a score of 6.63 among the studied samples. The overall acceptability level of the fermented sample increased with increase in fermentation time from 3.62 to 6.63. The HYF sample had the highest mean

Table 4: Sensory analysis scores of control, non-fermented and fermented samples

Sample	Colour	Aroma	Taste	Texture	Overall acceptability
SMF	6.201.38[b]	6.601.09[d]	6.540.87[c]	6.200.53[c]	6.801.21[d]
BUF	6.731.051[c]	4.050.78[a]	4.031.25[a]	3.670.76[a]	3.620.64[a]
DTF	5.800.45[b]	4.670.32[b]	5.331.06[b]	5.401.28[b]	4.870.98[b]
CJF	5.070.76[a]	5.401.15[c]	5.120.54[b]	5.601.09[b]	5.800.43[c]
HYF	4.871.25[a]	6.600.54[d]	6.330.93[c]	6.670.38[d]	6.631.04[d]

Values reported are means s.d of triplicate determinations
Mean values within each column bearing a different superscript roman letter are significantly (P0.05, LSD). SMF- Ogiri from melon seed (Control sample); BUF- Sample at fermentation time 0 h; DTF- Sample at fermentation time 48 h; CJF- Sample at fermentation time 72 h; HYF- Sample at fermentation time 96 h

sensory score and rated best among the studied samples with no significant difference with SMF sample which was ogiri from melon seeds. Its high ratings for flavour and texture contributed to its acceptance. Notably, the overall acceptability rating increased as fermentation time went up (from 3.62 to 6.63), implying that longer fermentation time might be beneficial for producing high quality conophor nut ogiri.

4. CONCLUSION

The study examined the influence of fermentation period on the conophor (*Tetracarpidium conophorum)* nuts. It was discovered that the nutritional value of *Tetracarpidium conophorum* nuts changed during fermentation. Fermentation significantly reduced the anti-nutrient content of the conophor nut. The conophor nuts fermented for 96 h had the highest amount of nutrients, particularly protein content, and this fermentation time was chosen as the optimal. In comparison, the fermented conophor nut condiment contained more nutrients, and its soup was as consumer-friendly as commercial ogiri soup. Therefore, fermentation of conophor nut for ogiri should be encouraged because it could be used as a food ingredient to enhance nutrition for both children and adults

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