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Effects of fermentation and wrapping materials on nutritional and anti-nutritional properties of watermelon (*Citrillus lanatus*) seeds.

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ABSTRACT

Purpose: This study aimed at evaluating the effects of fermentation on minerals, anti-nutrients, and proximate properties of seeds of watermelon subjected to fermentation.

Methodology: Three containers were used for the fermentation process viz: calabash, plastic, and foil paper. The dehulled, sorted and washed watermelon seed were boiled until soft and divided equally into the different containers and fermented for nine days. The mineral, proximate and anti-nutritional properties of the fermented samples were compared with ogiri samples purchased from the market.

Findings: There was increase in protein values of the fermented samples as follows calabash (37.55%), foil paper (35.51%) and plastic (35.51%) compared to the sample bought from the market (35.30%) and with significant difference across samples at 95% confidence level. Samples fermented in plastic had the highest moisture content (13.89%) and there was a significant difference among other samples with the sample fermented in calabash had the least value (11.24%). The sample in foil had the highest value (26.85%) of fat while the sample bought from market have the least (25.72%). Potassium was highest in value ranging from 41.65mg/100g to 27.45mg/100g while cupper with values ranging from 0.30mg/100g to 0.08mg/100g was least across the samples. For anti-nutrients, flavonoid has the highest (2.20mg/100g) while alkaloid was the least in value (0.49mg/100g). Samples fermented had better results in terms of mineral, proximate and anti-nutritional properties as compared to sample bought from the market. Results from this study revealed that fermentation significantly increase the nutrient and reduce the anti-nutrient contents of water melon seeds. Results from this study revealed that this unconventional substrate may serve as alternative to melon and ugba seeds for the production of ogiri, an indiginous condiment.

Recommendation: Watermelon seed as an unconventional substrate could be used as an alternative to oily seeds in the production of ogiri condiment, as well as a healthy and cheap source of protein. Conversion of watermelon seed to ogiri is a form of turning waste to wealth, thus creating job opportunities thereby reducing poverty.

Keywords: *Fermentation, epicarp, watermelon, nutritional, anti-nutrient, ogiri*

INTRODUCTION

Fermented food condiments form an integral part of African diets (Ejinkeonye *et al*., 2018). Foods are usually fermented to produce varieties of a better quality in terms of nutritional value, digestibility, and palatability for consumers, improving food safety through inhibition of pathogens and removal of toxic compounds (Odunfa, 1981). Ogiri is a fermented food condiment of wide acceptance (Jacob *et al*., 2015). Ogiri is a product of indigenous fermentation of oily seeds such as melon seed (*Citrullus vulgaris*), castor seed (*Ricinus communis*) and fluted pumpkin (*Telferia ocidentalis*) (Peter-Ikechukwu *et al*., 2015).

Wrapping materials are food packaging materials such as polypropylene, polystyrene, calabash, aluminum foil, plant leaves etc (Rabash *et al*.,2011). Reasons such materials are used to wrap are; infusion of product with beneficial phytonutrients, imparts on product distinctive aroma and taste, environmental friendliness, low cost, lack of toxicity and renewability (Peter-Ikechukwu *et al*., 2014). It has been reported severally that wrapping material provide the right environmental conditions for a food material relevant to processing and has been observed to improve the nutritional qualities (Peter-Ikechukwu *et al*., 2015).

Water melon is a tropical fruit which grows in almost all part of Africa and South East Asia. It is large, oval, round or oblong in shape. It is an economically important fruit with pleasant taste, rich water content and very rich in mineral and vitamins (Collins *et al*., 2007). They are an excellent source of protein and many beneficial minerals (Abiodun and Adeleke, 2010). However as highly beneficial as these seeds are most of the watermelon seeds are discarded (Dane & Liu, 2007). Based on the nutritional composition of watermelon seed, this unconventional substrate may serve as an alternative to known oily seeds used in producing ogiri condiment. The present study is aimed at assessing the effect of wrapping materials on the nutritional and anti-nutritional properties of ogiri produced by fermentation of an underutilized and non-conventional substrate, watermelon seed.

MATERIALS AND METHODS

Sample collection and preparation

Matured, fresh, healthy watermelon fruits were obtained from Sasa market in Akure, Ondo State, Nigeria. The fruits were cut open with a sterile knife, the water melon seeds were collected then washed in distilled water until clean, and they were sun dried then dehulled and kept in airtight container until used. Whole healthy seeds (600g) were boiled in 1200mL of distilled water in a clean aluminium pot for four hours to soften the seeds, during boiling water was intermittently added to avoid burning. On completion of the softening of the seeds they were mashed, divided into three portions, each portion were placed into a transparent plastic container, foil paper and calabash respectively to ferment.

Fermentation of sample

Fermentation was done at room temperature (28 ± 2^{o}C) for nine days. During fermentation microbial count of the Total viable bacteria and fungi were determined using nutrient agar and Potato dextrose agar respectively. The Physio-chemical parameters pH (pH 211 microprocessor, Hanna Instrument, England), total titratable acidity (TTA), and temperature were observed. The proximate, mineral and anti-nutrient composition of the fermented product was determined according to (Jacob *et al*., 2015)

Mineral analysis

Potassium, sodium, phosphorus and other minerals were determined using the method of AOAC (2010) and Jacob *et al.* (2015).

Determination of anti-nutrients

The anti-nutrients (saponin, phenol, alkaloid, flavonoids, tannin, phytate, oxalate and cyanide) were determined using standard methods as reported by Day and Underwood (1986), Bradbury *et al*. (1991), Makkar *et al*. (1993) and Jacob *et al*. (2015), respectively.

RESULTS AND DISCUSSION

The viable microbial count increased exponentially from day 1 to day six then started decreasing gradually. This could be attributed to the pH becoming alkaline and increase in temperature (Table 1). The sample wrapped in calabash had the highest microbial counts of $1.88x10^{10}$ cfu/mL for bacteria on 6^{th} day and $2.01x10^{10}$ sfu/mL for fungi growth on 7^{th} day, followed by plastic with $1.88x10^{10}$ cfu/mL bacteria on the 7^{th} day and $1.21x10^{10}$sfu/mL fungi respectively on the 7^{th} day, while foil paper had the least of $1.72x10^{10}$cfu/mL for bacteria on the 7^{th} day and $1.52x10^{10}$ sfu/mL for fungi on the 7^{th} day which could be because aluminium foil paper and plastic have non-porous nature which can prevent the entrance of external growth factors like air, moisture and oxygen needed for microbial growth. It could also trap heat which condenses to form water droplet that falls back on the fermenting sample. As observed in the result, there is no significant difference ($p \geq 0.05$) in the microbial count from day 1 to day 9 which may be because all the wrapping materials used had very small pour size (Ojokoh & Babatunde, 2014)

Table 1: Microbial load during fermentation of watermelon for 9 days

Days	Bacteria (calabash)	Fungi (calabash)	Bacteria (plastic)	Fungi (plastic)	Bacteria (foil paper)	Fungi (foil paper)
1	1.8×10^2	1.6×10^2	1.4×10^2	1.3×10^2	1.7×10^2	1.4×10^2
2	5.2×10^3	4.7×10^3	3.5×10^3	2.4×10^3	3.6×10^3	3.7×10^3
3	7.0×10^7	7.4×10^6	6.8×10^6	6.9×10^6	7.3×10^6	7.1×10^6
4	1.48×10^8	1.42×10^7	1.68×10^7	1.36×10^7	1.83×10^7	1.40×10^7
5	1.82×10^8	1.78×10^7	1.78×10^7	1.57×10^7	1.80×10^7	1.68×10^7
6	1.88×10^{10}	2.11×10^9	1.92×10^9	1.30×10^9	1.97×10^9	1.82×10^7
7	1.82×10^{10}	2.01×10^{10}	1.88×10^{10}	1.21×10^{10}	1.72×10^{10}	1.52×10^{10}
8	1.66×10^{10}	1.89×10^{10}	1.74×10^{10}	1.09×10^{10}	1.69×10^{10}	1.36×10^{10}
9	1.52×10^{10}	1.76×10^{10}	1.68×10^{10}	1.01×10^{10}	1.64×10^{10}	1.21×10^{10}

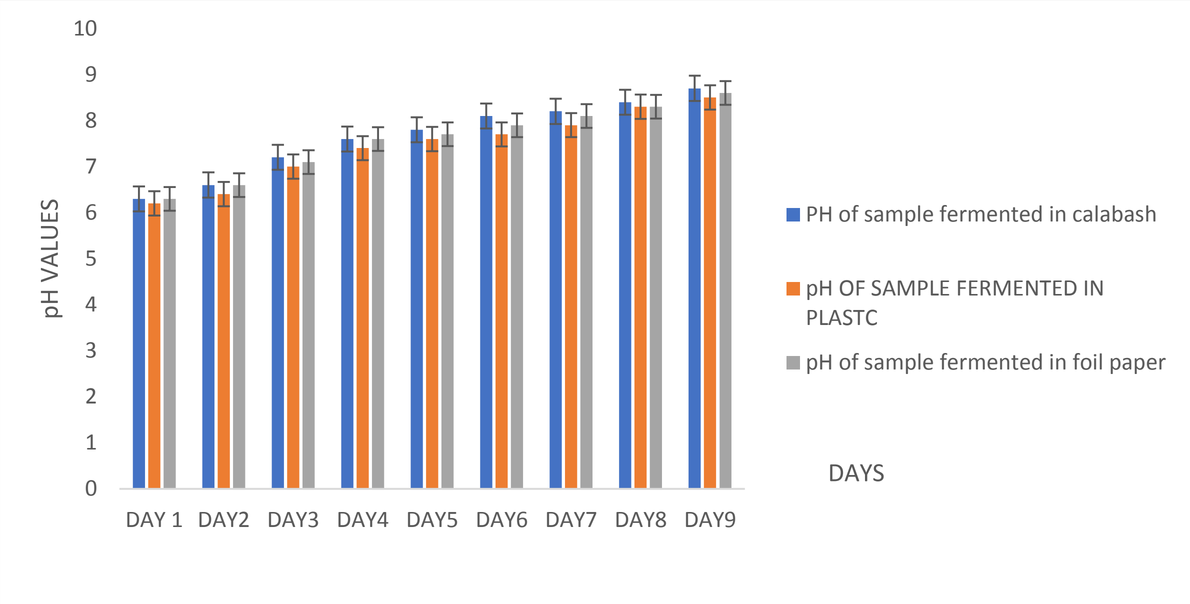

Figure 1: Change in pH values during fermentation of watermelon seed for 9 days

The result of the changes in pH as observed during fermentation of watermelon seeds is shown in fig 2. The pH across the samples increased from slightly acidic to alkaline. The increase in pH from slightly acidic to alkaline is in consonance with the observation of Odunfa (1981) who reported the ability of dominant microorganisms to carry out metabolic and enzymatic hydrolytic activities that break down protein into amino acids and ammonia.

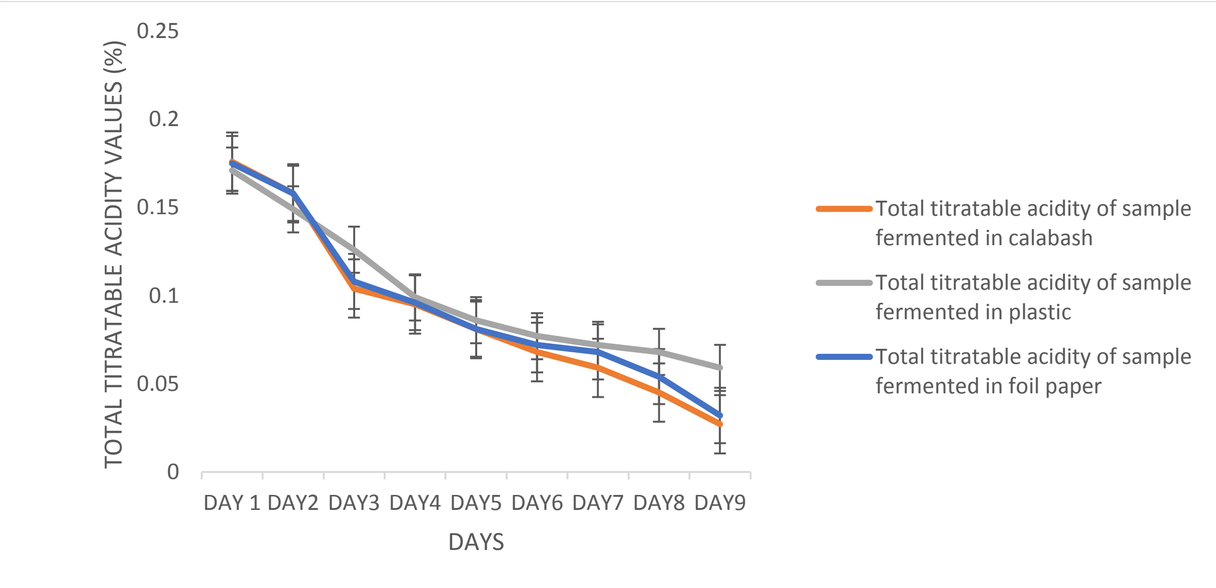

Figure 2: changes in Total titratable acidity values during fermentation of watermelon seed for 9 days

The titratable acidity (Fig. 2) decreased from 0.170% to 0.069% for calabash, 0.173% to 0.069% for foil paper and 0.176% to 0.055% for plastic. This could be due to ability of dominant microorganisms to carry out enzymatic activities that break down protein into ammonia across the samples which is in consonance with the report of Ojokoh and Babatunde, (2014).

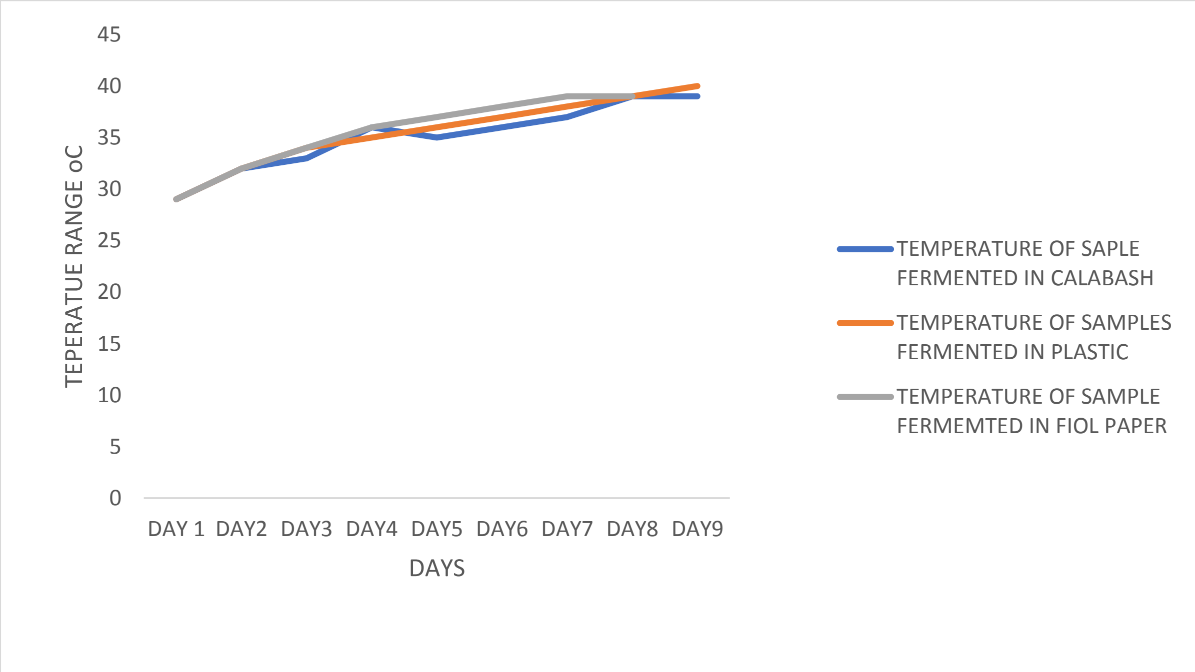

Figure 3: Changes in temperature values during fermentation of watermelon seed for 9 days

The temperature increased from 29^{o}C to 41^{o}C across the samples. (Fig.3) Plastic and aluminium foil had final temperature of 41^{o}C which may be due to non-porous nature of both which will prevent the leaking of heat energy generated during growth and metabolic process (Ojokoh & Babatunde, 2014).

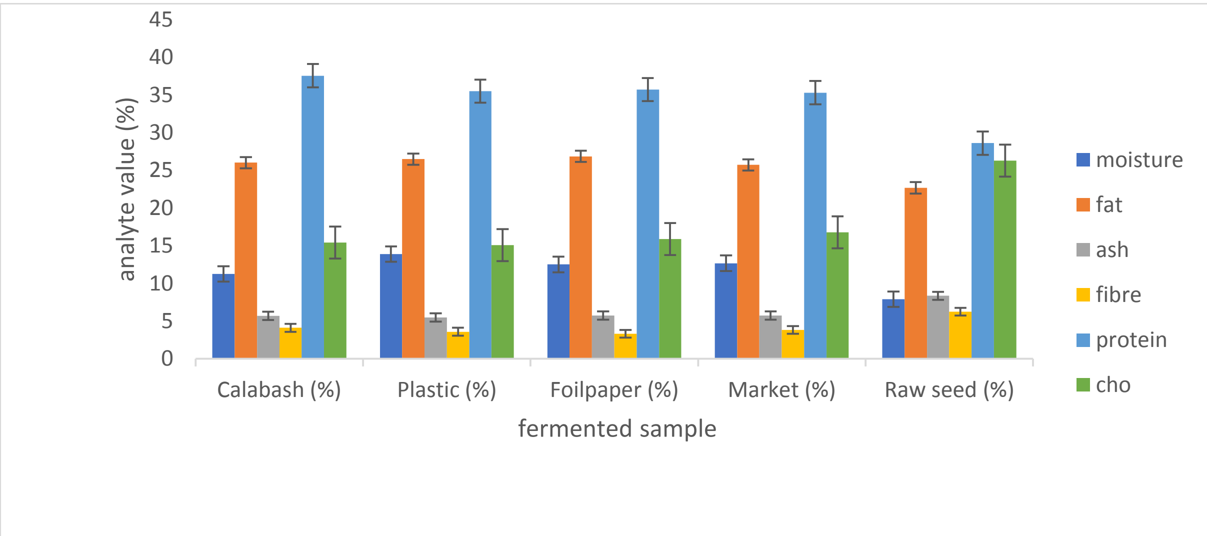

Figure 4: Proximate composition during fermentation of watermelon seed for 9 days

The proximate result (fig. 4) of the fermented watermelon samples revealed that sample fermented in plastic had the highest value for moisture (13.89 ±0.03%), there is a significant difference among other samples with the raw having the least value at $p \leq 0.05$. This could be attributed to the non-porous nature of plastic. The sample in foil had the highest value for fat there is significant difference among the fermented samples at $p \leq 0.05$, with the raw having

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the least value (26.85 ±0.10%). The raw sample had the highest value for crude fibre, ash and carbohydrate content with significant difference across the samples while for protein the fermented samples had higher values compared to the raw and with significant difference across samples.

Table 2: Mineral composition (mg/100g) of fermented and raw watermelon seeds

CODE	Calabash	Foilpaper	Plastic	Market
Na	19.30 ± 0.10^{f}	17.65 ± 0.15^{f}	15.45 ± 0.05^{f}	19.60 ± 0.20^{g}
K	41.65 ± 0.45^{g}	36.05 ± 0.25^{g}	30.65 ± 0.15^{g}	27.45 ± 0.15^{h}
Mg	2.65 ± 0.20^{c}	7.06 ± 0.02^{c}	3.23 ± 0.03^{c}	5.45 ± 0.35^{e}
Ca	8.61 ± 0.29^{d}	7.80 ± 0.10^{e}	4.41 ± 0.24^{d}	3.55 ± 0.05^{b}
Fe	1.07 ± 0.01^{a}	0.74 ± 0.00^{b}	0.84 ± 0.12^{b}	0.56 ± 0.03^{ab}
Mn	0.06 ± 0.00^{a}	0.032 ± 0.02^{a}	0.02 ± 0.00^{a}	1.04 ± 0.02^{bc}
Zn	0.53 ± 0.00^{ab}	0.53 ± 0.00^{b}	0.84 ± 0.00^{b}	1.30 ± 0.10^{d}
Cu	0.06 ± 0.00^{a}	0.02 ± 0.00^{a}	0.30± 0.00^{a}	0.08 ± 0.00^{a}
P	9.72± 0.00^{e}	7.67 ± 0.20^{d}	9.05 ± 0.00^{e}	6.60± 0.10^{f}

The results for the mineral composition are shown in Table 2. Potassium had the highest value ranging from 41.65mg/100g to 27.45mg/100g. High concentration of potassium in the body, increase iron utilization, helps to control hypertension as reported by Jacob *et al*. (2015). Sodium had a value of 19.30mg/100g to 15.45mg/100g. Its availability is vital because it regulates fluid balance in the body and helps in the proper functioning of muscles and nerves as reported by Jacob *et al*. (2015). Sodium is part of the resting membrane potential of a cell. The value of phosphorus ranges from 9.72mg/100g to 6.60mg/100g the recommended daily value is 1000mg, whole grains, seeds and vegetable are low in phosphorus while dairy products, meats, fish are rich in phosphorus. The deficiency is rare only in starvation because of its prevalence in food.

There is a high mineral content in watermelon seed (table 2) which may be as a result of the high ash content reported in Figure 4. Potassium had the highest value of all the trace metals across the samples thereby making watermelon a cheap source of this important mineral. Calcium is the next in value in the fermented sample, it is a constituent of bones, helps the body to combat correctly, blood to clot and the nerves to convey messages (Jacob *et al*. 2015). When calcium is deficient the body withdraws the needed calcium from the bones which if not replaced makes the bones weak and break easily. Calcium is essential for disease prevention and control.

From table 2, the values of Fe, Mn, Zn, Cu and Mg shows the mineral content of fermented watermelon seed far richer than those of melon, mango seeds, gmelina fruit and selected Nigerian oil seeds as reported by Jacob *et al*. (2015). The high value of some of the minerals may satisfy the nutritional needs of consumers.

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Table 3: Antinutritional composition (mg/100g) of fermented and raw watermelon seeds

Anti-nutrients	Market	Foil paper	Plastic	Calabash	Raw seed
Saponin	$0.06{\pm}0.0^{e}$	$0.04{\pm}0.0^{c}$	$0.05{\pm}0.0^{d}$	$0.05{\pm}0.0^{d}$	$2.12{\pm}0.0^{g}$
Phenol	$0.03{\pm}0.0^{bc}$	$0.03{\pm}0.0^{b}$	$0.03{\pm}0.0^{b}$	$0.03{\pm}0.0^{b}$	$1.07{\pm}0.0^{e}$
Alkaloid	$0.18{\pm}0.0^{h}$	$0.15{\pm}0.0^{f}$	$0.16{\pm}0.0^{g}$	$0.17{\pm}0.0^{g}$	$0.49{\pm}0.0^{a}$
Flavonoids	$0.04{\pm}0.0^{d}$	$0.03{\pm}0.0^{b}$	$0.04{\pm}0.0^{c}$	$0.04{\pm}0.0^{c}$	$2.20{\pm}0.0^{h}$
Tannin	$0.09{\pm}0.0^{f}$	$0.09{\pm}0.0^{d}$	$0.09{\pm}0.0^{e}$	$0.10{\pm}0.0^{e}$	$0.59{\pm}0.0^{c}$
Phytate	$0.11{\pm}0.0^{g}$	$0.12{\pm}0.0^{e}$	$0.12{\pm}0.0^{f}$	$0.12{\pm}0.0^{f}$	$0.52{\pm}0.0^{b}$
Oxalate	$0.02{\pm}0.0^{ab}$	$0.02{\pm}0.0^{a}$	$0.02{\pm}0.0^{a}$	$0.02{\pm}0.0^{a}$	$1.12{\pm}0.0^{f}$
Cyanide	$0.01{\pm}0.0^{a}$	$0.02{\pm}0.0^{a}$	$0.02{\pm}0.0^{a}$	$0.02{\pm}0.0^{a}$	$0.72{\pm}0.0^{d}$

The anti-nutrients in the raw and fermented watermelon seed (table 3) revealed flavonoid to be the highest (2.20mg/100g) while alkaloid had the least value (0.49mg/100g). The values of the anti-nutrient content in the raw was drastically reduced and significantly differed from each other in the fermented watermelon seed. Samples fermented in foil paper had best results, followed by those fermented in calabash and plastic while fermented sample bought from the market had the least values. The reduction could be attributed to the collective effect of fermentation and boiling as reported by Jacob *et al*. (2015) and Ejinkeonye *et al*. (2018). The antinutrient content in the fermented sample revealed a safe level that should not cause any health risk in animals.

CONCLUSION

From the materials used in fermenting the watermelon seed, samples fermented in the calabash was best followed by those fermented in foil paper, then plastic while samples bought from the market was least as adjudged from nutritional value and reduced level of antinutrient present in each fermented sample. The fermented samples were rich in protein and fat. Moreover, they are also rich in potassium, sodium, phosphorous and other essential minerals for good growth, disease prevention and control.

RECOMMENDATION

Therefore, watermelon seed as an unconventional substrate could be used as an alternative to oily seeds in the production of ogiri condiment, as well as a healthy and cheap source of protein. Conversion of watermelon seed to ogiri is a form of turning waste to wealth, thus creating job opportunities thereby reducing poverty.

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