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EFFECT OF FERMENTATION ON THE PROXIMATE AND ANTINUTRIENT COMPOSITION OF BANANA PEELS

Ozabor P.T[1+]
Ojokoh A.O[2]
Wahab A.A[3]
Aramide O.O[4]

[1,3]*Department of Microbiology, Osun State University, Osogbo, Nigeria.*
[1]*Email: praise.ozabor@uniosun.edu.ng Tel: +2347035235241*
[2]*Department of Biotechnology, Federal University of Technology, Akure, Nigeria.*
[4]*Department of Microbiology, Federal University of Technology, Akure, Nigeria.*

(+ Corresponding author)

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ABSTRACT

This study was carried out to investigate the effect of fermentation on the physicochemical and microbiological properties of banana peels with the view of reducing environmental pollution. Bacteria and fungi responsible for the fermentation were isolated and characterized using standard microbiological techniques. A total of 11 microorganisms were isolated and identified as: *Bacillus substilis*, *Lactobacillus fermentum*, *Micrococcus luteus*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Aspergillus flavus*, *Mucor mucedo*, *Rhizopus stonifer*, *Aspergillus niger*, *Saccharomyces cerevisae* and *Penicillium frequentans*. The pH of the samples decreased from 6.88 at 0 hr to 4.80 at 72 hr of fermentation while the titratable acidity increased from 0.21 g/l at 0 hr to 0.36 g/l at 72 hr of fermentation. The results of the proximate analyses revealed that there was a decrease in the ash (15.30 to 5.67%), fat (13.15 to 8.53%) and protein (13.72 to 10.09%) contents after fermentation. The moisture (8.28 to 13.28%) , crude fibre (15.29 to 32.08%) and carbohydrate (29.24 to 35.32%) contents and the energy value also increased from 1087.14 to 1217.75kj/g after fermentation. Results of the anti-nutrient composition shows that there was reduction in Phytate (12.66 mg/g to 9.27mg/g), oxalate (33.31 mg/g to 8.28 mg/g), glycosides (256.19 mg/kg to 149.02 mg/kg), Phenol (8.06% to 4.15 %), Tannin (7.01 mg/g to 3.79 mg/g) and Phytic acid (3.52 mg/g to 2.68 mg/g) contents after fermentation. Therefore, it can be concluded from this study that fermented banana peels are rich in nutrients needed in the formulation animal feeds.

Contribution/Originality: This study contributes to the existing literature that fermented fruit peels such as banana peels can serve as key ingredients in the formulation of animal feeds. Fermentation of these fruit peels enhances the nutritional properties and reduce the anti-nutritional contents, which makes the feeds relatively cheap and easily accessible.

1. INTRODUCTION

Food has been described as one of the basic necessities of life because the need for food begins with life as it provides the essential components of life and growth (Amadi, Ayalogu, & Onyeike, 2011). According to Paul and Wilke (1978) proper management and effective recycling of large volume of food wastes is a major challenge facing the world today. Food waste is any food substance, either raw or cooked that is been discarded or is intended to be discarded. Food wastes are generally generated by fruit/vegetable oil/palm oil, dairy, meat and sea foods industries. Therefore, for every ton of food waste, 4.5 tons of CO_2 is been emitted and one of the food wastes that is been generated largely is banana peels (Hirenpatel, Tejashsurati, & Gaurav, 2012; Maria & Kosseva., 2009).

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Banana is a popular tropical fruit which belongs to the *Musaceae* family. It originated from southeast Asia (Rahman & Kabir, 2003) a non-seasonal crop, grows about 5-9m in height with a tuberous rhizome, hard, long, pseudo stem. Its inflorescence is big with a reddish bract that is eaten as vegetable. Banana fruits are juicy, sweet, full of seeds and has thick peels (Byarugaba-Bazirake, Byarugaba, Tumusiime, & Kimono, 2014). Aurore, Parfait, and Fahrasmane (2009) had earlier documented that banana is the 5th most traded agricultural crop worldwide and is highly rich in carbohydrates, vitamins, minerals and secondary metabolites. Bananas are slightly radioactive than most other fruits because of their potassium content (Amarnath & Balakrishnan, 2007) which makes it an ideal food that can be used to beat down blood pressure (Debabandya, Sabyasachi, & Namrata, 2010). Due to the scarcity and increase in the price of raw materials used in the fermentation processes, scientists have engineered the initiative to move towards the use of organic (food) wastes generated by humans (Patel et al., 2012). Microorganisms plays either a positive or a negative role in food processing. The positive effect includes: preservation of foods, flavour enhancement and the reduction of anti-nutrients. However, the negative effects are: the contamination and spoilage of food products by pathogenic microorganisms (Ojokoh, 2007).

Therefore, fermentation of staple foods has help to provide a major source of nourishment for the rural and urban populations, contributing significantly to food security by increasing the range of raw materials that can be used in the production of edible products including animal feeds (Adewusi, Ojumu, & Falade, 2003). Achinewhu, Barber, and Ijeoma (2002) had earlier reported that fermentation increases the nutrient content of foods through the biosynthesis of vitamins, amino-acids and proteins. Thus, decreasing its anti-nutrient contents which make the food/feed safe to consume. The quantity of banana/plantain peels generated as waste is equivalent to 40% of the total weight of fresh banana in industries producing banana based food products (Tchobanoglous, Theisen, & Vigil, 2001). These peels are been dumped as solid wastes. Therefore, it is of high importance to find significant application of these banana peels as they contribute immensely to environmental pollution (Zhang, Whistler, BeMiller, & Hamaker, 2005).

2. MATERIALS AND METHODS

2.1. Collection of Sample

Banana (*Musa sapientum*) were purchased from Oja-Oba market, Akure,Ondo State, Nigeria. The samples were transported to the laboratory in clean low density polythene bags.

2.2. Preparation and Fermentation Of Samples

The banana fruits were washed in sterile distilled water and 300 grams of the peels were placed into a clean bowl containing 2 litres of distilled water which was covered and allowed to ferment for 72hrs at room temperature (Ojokoh, 2007).

2.3. Physico-Chemical Properties

The method described by AOAC (2012) was used to determine pH, titratable acidity (TTA) and temperature of the fermenting substrates. Samples were taken every 24 hr during the fermentation process and homogenized according to the procedure described by Ojokoh, Fayemi, Ocloo, and Nwokolo (2015).

The pH of fermented banana peel samples were measured using an Orion pH meter (Model 310, Orion Research Inc., Beverly, MA) equipped with glass electrode. pH meter was calibrated with KOH buffer solutions of pH 7.0 and 4.0 before the measurements. The titratable acidity was determined by titrating 20ml of the homogenized sample against 0.1 M NaOH using phenolphthalein as an indicator. Values obtained were expressed as percent lactic acid. All analyses were carried out in triplicate.

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2.4. Microbial Characteristics

The microbial profile of the raw and fermenting banana peel samples were determined at 12 hr interval. The changes in microbial population (cfu/g) of the total aerobic bacteria were determined using nutrient agar (NA) (Merck, Darmstadt, Germany) while De Man, Rogosa and Sharpe (MRS), (Merck) and M17 agar media (Oxoid, Basingstoke, Hampshire, England, UK) was used for the isolation of lactic acid bacteria (LAB) while potato dextrose agar (PDA) was used to isolate fungi and yeasts. Four different distinct colonies were randomly picked following visual assessment from the highest dilution factor of all the media used to determine the dominant microorganisms during the fermentation process. Banana peel samples were analyzed by homogenizing 1g of the fermenting substrate with 9 ml sterile 0.1% buffer peptone water (BWP) (Merck) followed by appropriate dilutions, spread plating and incubated at 37°C for 24 hr (for bacterial isolates) while MRS agar plates were incubated anaerobically using anaerobic jar together with anaerocult system (Merck) at 37°C for 48 hr. Colonies were selected randomly and purified before biochemical identification of the bacterial isolates (Ojokoh, Fayemi, Ocloo, & Alakija, 2014). Fungi isolates were identified using fungi compendium (Alexopolus).

2.5. Proximate Analyses

2.5.1. Moisture

Weight of a previously washed and dried empty evaporating dish was determined using a mettle balance as (W_1). A 10g of fermented banana peel was weighed into an evaporating dish (W_2). The dish and sample were then placed in the oven and dried for 8 hr at 105 °C after which the sample was placed in a dessicator to cool to room temperature (Fawole & Oso, 2001). After cooling, the sample was weighed. This process was continued until a constant weight was obtained, (W_3), (i.e., drying, cooling and weighing were done repeatedly at 30 mins interval until a constant weight was obtained). The weight of the moisture was calculated and expressed as a percentage of weight of the sample analyzed using the formula below:

$$\%\ \text{Moisture content} = \frac{W_2 - W_3}{W_2 - W_1} \times \frac{100\%}{1}$$

Where; W_1 = Weight of empty evaporating dish.

W_2 = Weight of sample + evaporating dish before drying.

W_3 = Weight of sample + evaporating dish after drying at 105 °C (AOAC, 2012)

2.6. Ash

Weight of a previously washed and dried empty crucible was determined using a mettle balance as (W_1). A 5g of fermented banana peel was weighed into the crucible (W_2). The crucible and sample were then placed in muffle furnace set at 550 ^{o}C for 48 hr. After ashing, the crucible and sample were both placed in the desiccator and allowed to cool at room temperature. The sample was then weighed after cooling (W_3). The percentage ash content was calculated thus:

$$\%\ \text{Ash} = \frac{W_3 - W_1}{W_2 - W_1} \times \frac{100}{1}$$

Where; W_1= Weight of empty crucible.

W_2= Weight of sample + crucible.

W_3=Weight of sample + crucible after ashing at 550 °C (AOAC, 2012).

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2.7. Crude Fibre

A 2g of each defatted sample was weighed into a 1L conical flask. 200ml of 1.25% sulphuric acid was added and the content was boiled for 30minutes. The mixture was filtered under vacuum followed by repeated washing with distilled water after which the sample was returned to the flask with the addition of 200ml of 1.25% NaOH solution. The mixture was boiled again for 30minutes and filtered. The sample was thoroughly washed with distilled water followed by 10% HCl solution and further washing was done to free the sample of any adhering acid. This was further treated with 10ml of light boiling petroleum ether and 10ml of ethanol (100%). The sample was scooped back into an empty crucible and placed in a hot-air oven set at 105 ^{0}C to dry for 1hr. After drying, it was placed in a desiccator and allowed to cool to room temperature and weighed. This was then placed in a muffle furnace and ashed for 90 minutes, allowed to cool to room temperature and weighed again . The crude fibre was calculated thus:

$$\% \text{ Crude fibre} = \frac{W_2 - W_3}{W_1} \text{ X } 100$$

Where; W_1=weight of defatted sample.

W_2=weight of sample at 105 ^{0}C.

W_3=weight of sample at 550 ^{0}C (AOAC, 2012).

2.8. Crude Fat

Fat content was determined using soxhlet type of direct solvent extraction method. A previously dried empty extraction thimble was weighed (W_1). A 3g of fermenting substrate was weighed into the thimble and placed in a 500ml capacity soxhlet extractor apparatus (W_2). The extractor apparatus and condenser with the 500ml round bottom flask was then set up and mounted on a heating mantle. Light boiling range petroleum either solvent (40 ^{0}C - 60 ^{0}C) was then added. The extraction was continued until the sample was completely defatted. The thimble was removed and placed in a hot-air oven and dried at 105 ^{0}C for 1 hr after which it was placed in a dessicator and allowed to cool to room temperature before being weighed again (W_3). The crude fat content was calculated thus:

$$\% \text{ Fat} = \frac{W_2 - W_3}{W_2 - W_1} \text{ X } \frac{100\%}{1}$$

Where; W_1= weight of empty extraction thimble.

W_2=weight of sample + extraction thimble.

W_3=dried weight of defatted sample + extraction thimble (AOAC, 2012).

2.9. Crude Protein

A 0.30g of sample was weighed into 50 ml macro-kejeldahl digestion flask. One tablet of copper sulphate catalyst and 5ml of concentrated tetraoxosulphate (VI) acid (H_2SO_4) was added. The flask was placed on a digestion block and digested at low temperature for 30 min, the temperature was allowed to rise until it becomes red hot. The sample was allowed to digest for 2 hr until it became clear. The digests were transferred into a volumetric flask. Each of the transferred digests was diluted with 50ml of distilled water. A 10 ml of each digested sample was measured into the distillation apparatus with gradual introduction of 10ml of 40% NaOH solution. The mixture was distilled by steam- powered heat and distillate collected into 5ml of 2% boric acid solution containing 3drops of mixed indicator. A 50ml of distillate from each duplicate was titrated with 0.01M HCl solution and colour change from blue to pink marked the end point of the reaction. The percentage nitrogen content in each sample calculated was multiplied with a factor 6.25 to get the percentage protein content.

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$$\% \text{ Protein} = \frac{N.F \times M \times V_1 \times T \times PF}{V_2 \times W} \times 100$$

Where; N.F = Nitrogen factor (0.014); M = Molarity of HCl (0.01); V_1 =Final volume of digest (50ml); V_2 = volume of digest used (10ml); T=Titre volume of distillate; W= Weight of sample used; PF= protein multiplication factor (6.25) (AOAC, 2012).

2.10. Carbohydrates Determination

The moisture, crude protein (N×6.25), crude fibre, crude fat and total ash contents of the samples were analyzed before and after 72 hr of fermentation using the method described by AOAC (2012) (Association of Official Analytical Chemists) approved methods. Total carbohydrate content was calculated by using the difference method (subtracting the sum of percent moisture, crude protein, crude fibre, crude fat and ash from 100%).

Total Carbohydrate = 100- (% of crude fibre +% moisture +% protein + %fat + % ash)

2.11. Anti-Nutritional Contents

2.11.1. Tannin

A 0.2g of finely grounded sample was weighed into a 50ml sample bottle, 10ml of 70% aqueous acetone was added to it and mixed thoroughly. The bottles were kept ice bath shaker and shaken for 2 hr at 30 ^{o}C. Each solution was then centrifuged and the supernatant stored in ice. 0.2ml of the solution was pipetted into a test tube and 0.8ml of distilled water was added. Standard tannin acid solutions was prepared from a 0.5mg/ml of the stock and the solution made up to 1ml with distilled water. A 0.5ml of Folin Ciocaeteau reagent was added to both sample bottles and standardized by pipetting 2.5ml of 20% Na_2CO_3. The bottles were vortexed and incubated for 40 min at room temperature after which its absorbance was read at 725nm using AJ- IC03 spectrophotometer against a reagent blank concentration of the same solution from a standard tannic acid curve that was prepared (AOAC, 2012).

$$\text{Tannin acid 1ml extract} = \frac{R \times 100}{\text{Ml of sample used}}$$

Where R = result read from the standard curve.

2.12. Oxalate

A 1g of sample was weighed into 1000ml conical flask. 0.75M H_2SO_4 was added and stirred intermittently with a magnetic stirrer for 1 hr. The mixture was filtered using Whatman No. 1 filter paper. A 25ml of sample filtrate (extract) was collected and titrated hot (80-90°C) against 0.1M$KMnO_4$ solution to the point when pink colour appeared that lasted for at least 30 seconds (AOAC, 2012).

2.13. Phytate

A 4g of sample was soaked in 100ml of 2% HCl for 3 hr and filtered using Whatman No. 1 filter paper. A 25ml of the filtrate was placed in 100ml conical flask and 5cm^3 of 0.03% of ammonium thiocynate solution (NH_4SCN) was added as and indicator. 50ml of distilled water was added to the solution and titrated against 0.00566g per milliter of standard iron (iii) chloride solution which contains 0.00195g of iron per milliliter until a brownish yellow colouration appears and lasted up to 5mins. Phytate content in mg/100g was calculated thus: Iron equivalent = litre value x 1.95

Phytic acid = litre value x 1.95 x 1.19 x 3.55mg/phytic acid (AOAC, 2012).

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2.14. Total phenol

A 0.20ml of plant extract was mixed with 2.5ml of 10% Folinciocalteau's reagent and 2.0ml of 7.50% sodium carbonate solution. The reaction mixture was subsequently incubated at 45^{0}C for 40 min and the absorbance of the coloured mixture was read at 700nm using UV visible spectrophotometer. Garlic acid was used as standard phenol.

2.15. Glycosides

A 4.0g of sample was weighed into a 250ml conical flask and 50ml of distilled water was added to it followed by the addition of 2.0ml of orthophosphoric acid. The sample was then stirred using a magnetic stirrer for 30minutes and covered with aluminum foil. This was left overnight at room temperature in order to liberate all bound hydrocynanic acid. The resulting mixture was then transferred into a 250.0ml distillation flask with a drop of paraffin oil and some anti-bumping chips, distillation water and 1g of NaOH pellet. The distillate was then transferred into a 50ml volumetric flask, made to the mark with distilled water. A 20ml of this solution was measured out into an Erlenmeyer flask with 1.60ml of 5% KI solution. The solution was then titrated against 0.10M solution of $AgNO_3$. Blank was also titrated until the end point was indicated by a faint, but permanent turbidity.

2.16. Statistical Analyses

Analysis of variance (ANOVA) was performed on the data at $p \leq 0.05$ using MINITAB statistical software (Minitab® Release 14.13, minitab Inc., USA). Significant means were separated using the least significant difference (LSD) at $p \leq 0.05$.

3. RESULTS

3.1. Physicochemical Properties

3.1.1. pH, Titratable Acidity and Temperature

The changes in pH, titratable acidity and temperature of banana peels during the fermentation process are presented in Figures 1, 2 and 3 respectively.

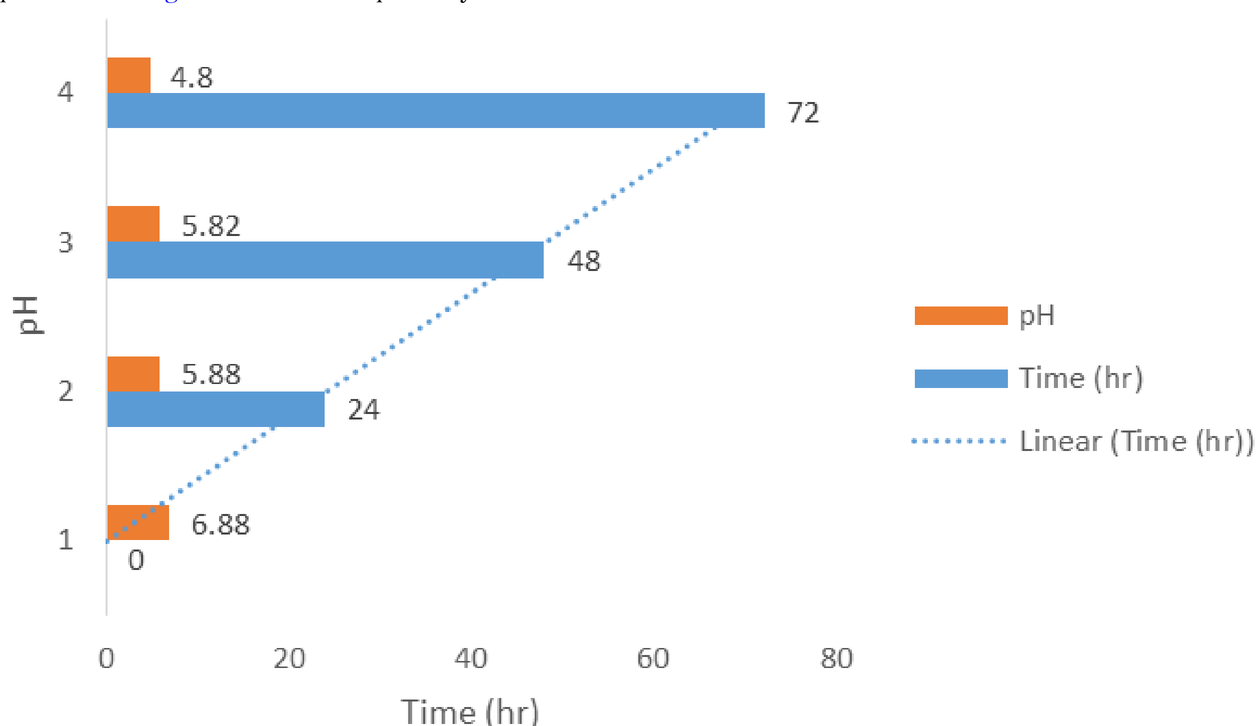

Figure-1. Changes in pH during the fermentation of banana peels. The pH was observed to decrease with increase in fermentation time. The temperature ranged from 6.88 at 0 hr to 4.8 at 72 hr during the fermentation process.

Note: Key: hr= time.

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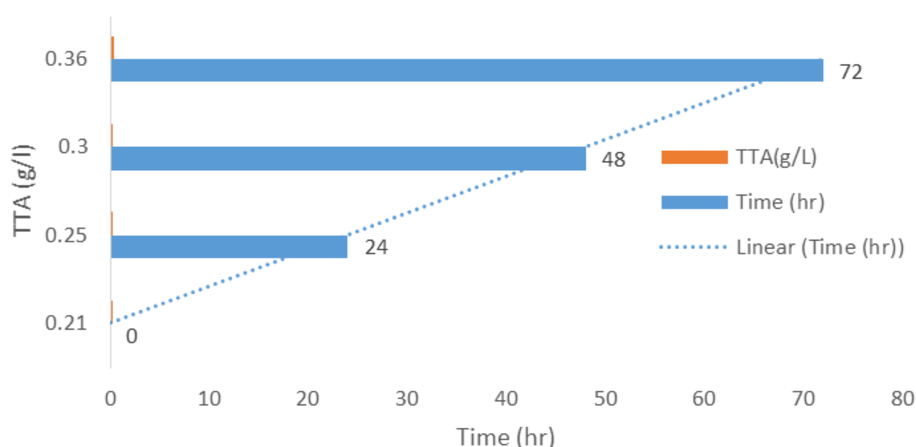

Figure-2. Changes in TTA (g/L) during the fermentation of banana peels. TTA increases with with increase in fermentation time. Therefore, TTA increases with a simultaneous decrease in pH. The TTA ranged from 0.21 at 0 hr to 0.36 at 72 hr during the fermentation of banana peels.

Note: Key: TTA= titratable acidity.

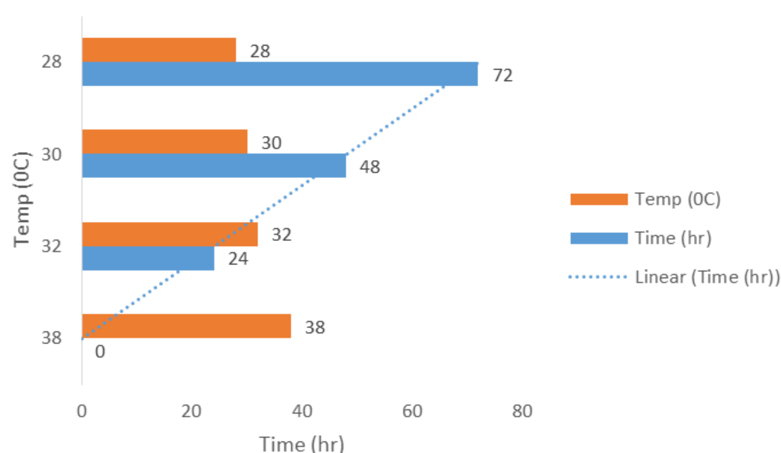

Figure-3. Changes in temperature (⁰C) during the fermentation of banana peels. Temperature was observed to decrease with increase in fermentation time. The temperature ranged from 38 ⁰C at 0 hr to 28 ⁰C at 72 hr during the fermentation process.

Note: Key: ⁰C= temperature.

Table-1. Morphology and biochemical characterization of bacteria isolates during fermentation of banana peel.

Isolate code	Morphological Characteristics	Gram reaction	Spore Test	Motility	Catalase	Coagulase	Oxidase	Glucose Sucrose	Fructose	Galactose	Maltose	Lactose Mannitol	Probable organism
BP1	Creamy white, flat, irregular and rough rod	+	+	+	+	-	-	A A	A	A	A	A A	*Bacillus substilis*
BP2	Creamy , convex, entire and smooth rod	+	-	-	-	-	-	AG AG	AG	AG	AG	AG AG	*Lactobacillus fermentum*
BP3	Yellow, punctiform,entire wrinkled cocci	+	-	-	+	-	+	A A	A	-	-	- A	*Micrococcus luteus*
BP4	White, rough, rhizoid, flat, rod	-	+	+	+	-	+	- -	-	A	A	- A	*Pseudomonas aeruginosa*
BP5	Yellow, regular, raised, entire, smooth diplococcic	+	-	-	+	+	-	AG A	A	A	A	A A	*Staphylococcus aureus*

Note: KEY+= positive ; - = Negative A; AG= Acid production.

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Table 1 presents a total of five (5) bacteria isolated both in the raw unfermented banana peels and also the fermented samples. They are were isolated and identified as: *B. subtilis*, *L.fermentum*, *M. luteus*, *P. aeruginosa* and *S. aureus*.

+= positive -= negative

Table-2. Morphological characteristics of fungi isolated during the fermentation of banana peels.

Cultural characteristics	Colony morphology	Microscopic appearance	Probable organisms
Black mycelia growth	Circular, transparent, rough, raised, lobate, filamentous, black	Hypha is septate. Possess simple upright conodiophores that terminates in a globose or clave swelling, bearing phialides at the apex or radiating from the entire surface. Conidia are one-celled and globose.	*Aspergillus niger*
Glistering cream rice-like mycelia growth	Cream, non-filamentous irregular,raised, ovoid,round in clusters, budding, smooth and glistering	Hypha is absent. Ascospores are globose. Unbranched with swollen apex. conidiophores bears vesicles that produce chains of conidia	*Saccharomyces cerevisiae*
Yellow mycelia growth	Circiular, opaque, smooth, flat,filamentus, yellow-green		*Aspergillus flavus*
Dirty white cotton-like mycelia growth	Colonies are typically coloured white to beige or gey and are fast growing	Hypha is non-septate. Upright conidiophores bear columella and sporangium at the apex that bears the spores which are smooth and regular	*Mucor mucedo*
Greyish fluffy mass, cotton-like mycelia growth	Grey fuzzy colonies. They are round with flattened bases	Hypha is non-septate. Thin sporangiophore arising at nodes bearing lubrella-like sporangium bearing spores which are black in colour	*Rhizopus stonifer*
Rapid growing colonies, gey-green,powdery, striate from the centre to the outside.	Green with yellow border and valvate appearance with darker edges	The conidiosphores are smooth-walled,swollen at the apex.the conidia are globose and smooth walled.	*Penicillium frequentans*

Table 2 shows the total number of fungi isolated during the fermentation of banana peels. Six (6) fungi were isolated and identified as: *A. niger*, *S. cerevisiae*, *A. flavus*, *M. mucedo*, *R.stolonifer* and *P. frequentans* both in the unfermented and fermented banana peel samples.

Table-3. Changes in microbial load at 24 hr interval during fermentation of banana peels.

Time (hr)	Bacteria (cfu/ml)	Fungi (cfu/ml)
0	2×10^2	1×10^2
24	4×10^3	2×10^3
48	6×10^3	4×10^3
72	9×10^5	6×10^5

Note: Key: hr= time cfu= colony forming unit.

Changes in microbial load during the fermentation of banana feels is presented in Table 3. It was recorded that the bacterial and fungal load increased from 2×10^2 at 0 hr to 9×10^5 at 72 hr and 1×10^2 0 hr to 6×10^5 at 72 hr respectively. The bacterial count was significantly higher than the fungal count ($p<0.05$).

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Table-4. Proximate composition of banana peels before and after fermentation.

Proximate Composition (%)	Fermented	Unfermented
Ash content	5.68±0.101^{a}	15.30±0.016^{b}
Moisture content	13.28±0.080^{a}	8. 28±0.010^{b}
Fat content	8.53±0.150^{a}	13.15±0.128^{b}
Crude fibre content	32.08±0.091^{a}	15.29±0.050^{b}
Protein content	10.09±0.117^{a}	13.72±0.149^{b}
Carbohydrate content	35.32±0.231^{a}	29.24±0.071^{b}
Energy value (Kj/g)	1217.75±0.033^{a}	1087.14±0.162^{b}

Note: Values are means± standard deviation of triplicates. Values in the row with different superscripts are significantly different at p<0.05.

Table 4 shows the proximate composition of banana peels before and fter fermetation. It was observed that the moisture , crude fibre and carbohydrate contents as well as the energy value of the banana peels increased after fermentation while the ash, fat and protein contents decreased in value.

Table-5. Anti-nutrients composition of banana peels before and after fermentation.

Anti-Nutrients Composition	Fermented	Unfermented
Phytates (mg/g)	9.27±0.100^{a}	12.66±0.027b
Phytic Acid (mg/g)	2.68±0.022^{a}	3.52±0.011^{a}
Oxalates (mg/g)	8.28±0.018^{a}	33.31±0.080^{b}
Tannins (mg/g)	3.79±0.067^{a}	7.01±0.023^{b}
Phenols (%)	4.15±0.046^{a}	8.06±0.067^{b}
Glycosides(mg/kg)	149.02±0.102^{a}	256.19±0.110^{b}

Note: Values are means± standard deviation of triplicates. Values in the row with different superscripts are significantly different at p<0.05.

From the results of the anti-nutrient contents documented in Table 5, it can be deduced that fermentation had significant effect on the banana peels as all the anti-nutrient contents decreased significantly after fermentation.

4. DISCUSSION AND CONCLUSION

The results obtained from the study provides information on the physico-chemical parameters (such as pH, titratable acidity and temperature), microorganisms involved in the fermentation process, proximate and anti-nutritional contents of fermented banana peels for feed formulation. The pH of the fermented banana peels was recorded to range from 6.88 at 0 hr to 4.80 at 72 hr during the fermentation process which indicates that it decreases as the fermentation time progresses. The TTA of the fermenting substrate was also recorded to range from 0.21 at 0 hr to 0.30 at 72 hr of fermentation. This denotes that as the fermentation time increases, the TTA increases. Therefore, as the pH decreases, there was a simultaneous increase in TTA. In addition the temperature of fermenting substrate was observed to decrease as the fermentation time progresses. Several authors have documented reports on decrease of pH accompanied with increase in TTA during fermentation of food and feed products which aids to inhibit the growth of pathogenic microorganisms. Ojokoh et al. (2015); Ojokoh et al. (2014); Omemu, Oyewole, and Bankole (2007); Omemu and Omeike (2011); Ekwem and Okolo (2017); Ojokoh (2007) documented a similar result during the fermentation of mango peels for feed formulation. Five (5) bacteria (*Bacillus substilis*, *Lactobacillus fermentum*, *Micrococcus luteus*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*) and six (6) fungi (*Aspergillus flavus*, *Mucor mucedo*, *Rhizopus stonifer*, *Aspergillus niger*, *Saccharomyces cerevisae* and *Penicillium frequentans*) were isolated during the fermentation process. *M. luteus*, *P. aeruginosa* and *S. aureus* were isolated from the raw unfermented samples and at the early stage of fermentation. The sources of these organisms could be from the banana peels, indigenous microflora prior to fermentation, utensils used, water and the producers (Ojokoh et al., 2015; Osuntogun & Aboaba, 2004). *B. subtilis* have earlier been reported to be one of the most predominant organisms involved in the fermentation of food and feed products due to the production of metabolites that inhibits the growth of harmful pathogens (Achi, 2005; Afify, Romeilah, Sultan, & Hussein, 2012; Sanni, Onilude, Fadahunsi, Ogunbanwo, & Afolabi, 2002; Soetan, Akinrinade, & Adisa, 2014). However, *L. fermentum* was isolated at the later stage of the fermentation process. Lactic acid bacteria have been documented to be responsible for production of

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inhibitory substances such as organic acids, lactic acid, propionic acid, bacteriocins and hydrogen peroxide which leads to the acidification of the fermenting substrate (Odumodu & Inyang, 2006; Ojokoh, Daramola, & Oluoti, 2013; Oliveira, Zannini, & Arendt, 2014). The fungi isolated from the fermenting substratess had earlier been documented to also produce inhibitory substances against pathogenic microorganisms present in the fermenting substrates. Ojokoh (2007) gave a similar report during the fermentation of mango peels for feed formulation.

The lower ash content obtained from the proximate analysis of the fermented banana peels when compared to the raw unfermented sample is indicative of its low mineral contents. This is in line with the report of Bello (1999) who reported an ash content of 2.49 from corn cobs. Abubakar et al. (2016) documented that an ash content of 1.5-2.5% is recommended for animal feeds. The fermented banana peels had a higher moisture content that the raw unfermented sample. Abubakar et al. (2016); Romelle, Ashwini, and Ragu (2016); Hassan, Hassan, Usher, Ibrahim, and Tabe (2018) had previously documented that the moisture content of a food/feed is an indication of its freshness, shelf-life and the activity of water soluble enzymes and co-enzymes responsible for metabolic activities which can lead to its deterioration. The crude fat content of the fermented banana peels was lower than the unfermented sample peels and this suggests that it can contribute substantially to the energy content of feeds. The low fat content will lead to increase in storage capacity, thereby reducing the development of rancidity (Hassan et al., 2018; Okareh, Adeolu, & Adepoju, 2015). The fermented sample peels had a higher fibre content than the unfermented sample. High fibre content in food/feed helps to remove potential mutagens, steriods and xenobiotics by attaching to dietary fibre components and thereby improves digestion. Hence, this fruit peels boosts the health of live stocks and fish when used as feed (Eleazu, Iroaganachi, & Eleazu, 2013; Ojokoh, 2007; Romelle et al., 2016). The fermented peel sample had a lower protein content than the unfermented sample. The protein content obtained is however higher than that of shea butter fruit pulp (Adepoju & Adeniji, 2008) *Amaranthus* and cocoyam leaves (Adepoju & Adeniji, 2008). The result obtained from this work is however in contrast with the one documented by Ojokoh (2007) who reported a higher protein content from fermented mango peels than the unfermented one. Protein is a major component of food/feed that is required for survival of humans and animals for the supply of substantial amount of amino acids. Fermented samples showed a higher carbohydrate content than the unfermented sample which indicates it can serve as a good source of energy for the animals (Okareh et al., 2015). The energy content of fermented sample peels was higher than the unfermented one. Hassan, Baba, Shibdawa, Mahmoud, and Ishaku (2013) however, documented a much higher energy content from *Moringa oleifera* seeds.

Fermentation greatly decreased the anti-nutritional contents of the fermented banana peel sample. The reduction in phyate and phytic acid could be attributed to the activity of L. fermentum which possess the phytase enzyme that breaks down phytate Ojokoh, Adetuyi, and Akinyosoye (2005). Romelle et al. (2016) also documented that phytic acid in plants have chelating effect on certain essential mineral elements present such as: Ca, Mg, Fe and Zn to form insoluble phytate salts (Agte, Tarwadi, & Cheplonkar, 1999). Low phytate and phytic acid contents have also been reported from fermented watermelon peels, pomegranate peels and citrus peels (Calin-Sanchez et al., 2013). Ladeji, Akin, and Umaru (2004); Johnson et al. (2012) and Romelle et al. (2016) documented that oxalates have the ability to bind to calcium in food, thus, rendering calcium unavailable for the normal physiological and biochemical activities. Fermented pawpaw peels, pomegranate peels and watermelon peels also showed similar amount of oxalates which correlates to the one obtained from this work. According to the report of Ojokoh (2007); Wakil and Kazeem (2012) tannin disrupts the nutritive value of food/feed products by forming complex with protein (both substrate and enzyme), thus inhibiting digestion and absorption. They also bind Fe making it unavailable. Furthermore, condensed tannins can cleave to DNA in the presence of copper ions. Decrease in tannin may be due to the processing that the sample was subjected to coupled with the activities of microbial enzymes involved in the fermentation process.

The phenols in humans and animals have been reported to exhibit wide range of biotherapeutic effects such as anti-bacterial, anti-inflammatory, anti-fungal and antioxidant properties. The total phenolic content obtained from

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this study correlates with the reports of Hans, Shen, and Lou (2007) and Romelle et al. (2016) from the study of anti-nutritional contents of various fruits such as pawpaw, pineapple, mango, apple, banana, orange, pomegranate and watermelon.

Glycosides are carcinogens from which hydrogen cyanide (HCN) may be produced by hydrolysis. HCN is a very poisonous substance formed by the of acids on metal cyanides. Fermentation greatly reduced the level of glycosides present in foods. Akande, Doma, Agu, and Adamu (2010) documented that glycosides can cause dysfunction of the central nervous system, respiratory failure and cardiac arrest. Romelle et al. (2016) documented cyanogenic glycosides of 116.26% from banana peels which is lower than the one obtained from this study. Other fruit peels such as: pawpaw, pineapple, mango, apple, orange, pomegranate and watermelon produced the following glycoside contents: 69.83%, 71.50%, 45.90%, 96.04%, 39.79%, 26.96% and 121.02% respectively.

In conclusion, this study has revealed that fermented banana fruit peels can be used for the production of animal feeds. The effect of fermentation significantly increased the nutritional contents and decreased the anti-nutritional contents which infers that the fermented banana fruit peels is safe for consumption by livestock animals.

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