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Research Article
Hydrolysis of Cassava Peels with Concentrated Cellulase from *Bacillus subtilis* Improved Its Nutritional Contents

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Abstract

Background and Objective: Unabated increase in the price of animal feed ingredients has necessitated search for readily available and cheaper cassava peels. This study was designed to hydrolyze cassava peels with concentrated cellulase from *Bacillus subtilis* with a view to improve its nutritional contents. **Materials and Methods:** Bacteria associated with cassava waste water were identified using standard microbiological techniques. The isolates were screened for cellulase production in submerged state fermentation using biochemical assay. The cassava peels were degraded with partially purified cellulase from the best-producing bacterium. The nutritional composition of cellulase-hydrolyzed samples was determined using standard chemical methods. **Results:** The bacterial isolates identified were: *Bacillus cereus*, *Pseudomonas aeruginosa*, *Micrococcus* sp. and *Lactobacillus acidophilus*. *B. subtilis* CWW2 the had the highest specific enzyme activity, it was therefore selected for further studies. There was an appreciable increase in the protein content of the sample hydrolyzed with concentrated enzyme from 1.4% to 5.6%, while there was reduction in the fiber and contents by approximately 22 and 40%, respectively. The lignin, cellulose and hemicelluloses contents decreased from 6.10, 31 and 27% in untreated samples to 3.7, 26 and 16% in the samples hydrolyzed with enzyme preparation. There was also a reduction in the anti-nutritional compounds of the samples treated with enzyme when compared with untreated samples. The treatment of the sample with concentration enzyme resulted in increase of vitamins. **Conclusion:** In conclusion, the treatment of cassava peels with concentrated cellulase increased its nutritional contents.

Key words: Cassava peels, concentrated cellulase, chemical composition, anti-nutritional compounds

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The dependency on agricultural wastes to offset the fluctuating global market price of conventional feed ingredients (such as fishmeal) for livestock production have been the focus of developing countries. Due to the search for an alternately cheap cost of raw material for livestock feed formulation, cassava and its by-products has enjoyed widespread patronage[1,2]. Cassava (*Manihot esculenta Crantz*) is a root tuber crop which is a staple diet of a large populace from Africa, Asia and Latin America[3]. Nigeria is one of the major growers of cassava in the world because of favorable biotic and abiotic factors that support its cultivation. The income generated annually from cassava cultivation in Nigeria and the products made from it might compete fairly well with crude oil in the future. Cassava tubers have been fermented into carbohydrate-rich foods such as; 'gari', 'lafun' and 'fufu' etc. It has also been processed into flours as vital ingredient for some commercially packaged foods. Prior to its processing, the cassava peels which are wastes are generated by mechanically removing the outer coverings of cassava and could serve the purpose of a cost-effective product for the formulation of animal feed. The indiscriminate wasteful handling of this waste has constituted a great environmental challenge due to the obnoxious odour produced when it rots[4]. The limitations to the use of cassava peels in livestock nutrition are their high fibre content, low calorific value and their heavy loads of anti-nutrients like cyanide, tannins and phytates[1].

Due to the toxic nature of cassava, its natural edibility becomes almost impossible. Cassava toxicity is as a result of the presence of chemical constituent like cyanogenic glycosides, linamarin and lotaustralin[5]. Overtime consumption of foodstuff and products with these residual chemical constituents in this crop is capable of inducing chronic diseases such as; goiter, cretinism, tropical atoxic neuropathy and tropical diabetics[6]. It is against this backdrop that cassava processing becomes essential so as to reduce the toxicity and increase its nutritive value thus enhancing edibility.

Bioconversion is an effective and productive alternative for the utilization of waste products. This process is carried out by the fermentation of the agricultural wastes such as maize cobs with fungi thus increasing the world food and feed supplies especially those high in protein and most of the nutritionally valuable diets with small amount of anti-nutrient content[7]. Adeleke *et al.*[8] stated that biodegradation which is a similar process with bioconversion by microbial fermentation of cassava peels improves the food nutritive qualities like shelf life, texture, aroma, nutritional quality, digestibility and reduction in anti-nutrient content under different conditions. Hydrolase enzymes such as mannanase and cellulases of microbial origins have been shown to improve feed bioconversion and performance of broilers, fish, turkeys and swine in recent years[9,10]. For instance, cellulases play an important role in the breakdown of glycosidic bonds in the main chain of cellulose into glucose[9,10]. The treatment of Palm (*Elaeis guineenis*) Kernel Expeller (PKE) rich in cellulose with cellulase had been reported to increase the metabolizable energy and improve the nutrient digestibility[11]. In a similar study, the treatment of palm kernel meal by mannanase from *Penicillium italicum* led to an increase in its nutritive values[10]. Therefore, in an effort to understand the financial constrains attached to the purchase of animal feed, we treated cassava peels, an under-utilized agricultural waste with concentrated cellulase from *Bacillus subtilis* with a view to improve its nutritive values.

MATERIALS AND METHODS

Study area: This research was carried out at the Department of Microbiology, Federal University of Technology, Akure, Nigeria between March and June, 2017.

Sample collection: Cassava waste water and cassava peels were obtained from a cassava processing factory at Oke-Odo, Akure, Ondo State, Nigeria. The cassava waste water was refrigerated at 4 centigrade (°C) and the cassava peels were sundried for 5 days and milled for microbiological analyses.

Isolation and identification of bacteria associated with cassava waste water: Appropriate dilution factor from serially diluted cassava waste water was pour plated with already sterilized molten nutrient agar and incubated at 37±2°C for 18 h. The resultant bacterial colonies were counted and expressed as colony forming unit per millimeters (CFU mL^{-1}). The emerged colonies from the agar plates were sub-cultured to obtain pure culture. Pure bacterial isolates were presumptively identified by means of morphological features and some biochemical tests according to taxonomic indices described in Bergey's Manual of Determinative Bacteriology[12].

Primary screening of bacterial isolates for cellulase production: The bacterial isolates were screened for cellulase production on carboxymethylcellulose (CMC)-agar medium as described by Arotupin and Olaniyi[13]. The pure bacterial cultures were cultivated individually in Minimal Salt Medium (MSM) supplemented with CMC as the sole carbon source. The bacterial pellets obtained after centrifugation of the broth was

serially diluted with sterile distilled water and introduced into the agar well on the plates and then incubated at 37°C for 24 h. Congo red was used to wash used to wash the plates for a clearer or more visible zone of hydrolysis. The best bacterial isolate with maximum zone of hydrolysis on CMC-agar was maintained on nutrient agar slants and kept in the refrigerator at 4°C.

Secondary screening of bacterial isolates for cellulase production: The bacterial isolates were screened for cellulase production in MSM with the composition as follows: (g L^{-1}) 0.2 g peptone water, 0.2 g yeast extract, 0.2 g $NaNO_3$, 0.1 g K_2HPO_4, 0.05 g $MgSO_4.7H_2O$, 0.05 g KCl, 0.01 g $FeSO_4.7H_2O$, 10 g CMC and 1000 mL distilled water. Afterward, the sterile MSM was inoculated with 1000 µL of pure bacterial culture and incubated at 37±2°C for 22 h. After a period of incubation, the enzyme production medium was centrifuged at 6000 revolutions per minute (rpm), 4°C for 20 min. The supernatant was collected and used for enzyme assay.

Cellulase assay: About 500 µL of the enzyme supernatant was pipetted into a test tube containing 500 µL of CMC prepared in 50 mM potassium phosphate buffer pH 6.8. The test tubes were incubated at 45°C for 30 min in a water bath. The reaction of the mixture was terminated with the addition of 2 mL of 3, 5-Dinitrosalicylic acid (DNSA) and then boiled at 100°C for 15 min for colour development. One milliliter distilled water was added to bring up to 3 mL in the test tubes and the absorbance was measured at 540 nm using a UV spectrophotometer. One unit of cellulase activity was defined as amount of enzyme producing 1 µmol of glucose per minute under standard assay conditions[14].

Partial purification of crude cellulase: Crude enzyme from the best cellulase-producing bacterium was purified by ammonium sulphate precipitation to obtain 60% saturation. It was gently stirred on magnetic stirrer for 60 min and allowed to stay overnight at 4°C. The precipitate from the mixture was harvested by centrifugation[9] at 6000 rpm for 30 min at 4°C. The residue of the precipitate was washed into a clean container with 50 mM phosphate buffer at pH 6.7.

Preparation of Cellulase Treated Cassava Peel (CTCP): About 72 g of milled cassava peels was dissolved in 400 mL sterilized distilled water and pasteurized in a water bath at 72°C for 30 min. Afterward, 24 mL of partially purified cellulase was added to pasteurized cassava peels and incubated at room temperature, 28±2°C for 20 h under a sealing system. After treatment, the hydrolyzed product was dried in an oven for 60 h to obtain the CTCP[10].

Chemical constituents of CTCP: The chemical constituents of CTCP were determined following the standard methods of AOAC[15]. Fiber compounds including Acid Detergent Fiber (ADF), Neutral Detergent Fiber (NDF) and Acid Detergent Lignin (ADL) contents were measured sequentially with a fiber automatic analyzer (Fibertec System, M, Tecator, Hoganas, Sweden)[16]. Hemicellulose was calculated as NDF-ADF, cellulose as ADF-ADL, while lignin content is obtained by the subtraction of residue after extraction from ash. Phytate was determined through the extraction of the samples with hydrochloric acid and sodium sulphate and absorbance measured at 660 nm[17]. Tannin was determined using the method of vanillin hydrochloric acid and absorbance was measured at 500 nm[18]. Oxalate determination was done according to the standard method of Day and Underwood[19], while cyanide content was evaluated by the method of Obadoni and Ochuko[20]. The vitamin contents in CTCP were determined in line with the standard methods of AOAC[15].

Statistical analysis: Data obtained are analyzed by analysis of variance and significant differences between means are compared using Duncan multiple range test.

RESULTS

Total bacterial counts and identification: Table 1 shows the total bacterial counts obtained from cassava waste water. The bacterial population after 18 h of incubation was 19.0×10^5 and 8.0×10^8CFU mL^{-1}, respectively. In Table 2, morphological and biochemical characterization of the bacterial isolates revealed the identities of *Bacillus cereus*, *B. subtilis*, *Pseudomonas aeruginosa*, *Micrococcus* sp. and *Lactobacillus acidophilus*.

Primary and secondary screening of bacterial isolates for cellulase production: The diameter of the zone of CMC hydrolysis from each of the bacterial isolates is presented in Table 3. The *B. subtilis* showed the highest diameter of zone of CMC hydrolysis of 28.0 mm, followed by *B. cereus* with a value

Table 1: Total bacterial counts

Time (h)	Nutrient agar $\times10^5$ (CFU mL^{-1})	Nutrient agar $\times10^8$ (CFU mL^{-1})
24	19.0±0.41[a]	8.0±0.33[a]
48	25.0±0.21[b]	11.0±0.23[b]
72	41.0±0.11[c]	24.0±0.31[c]

Means with the same superscript letters along the same column are not significantly different ($p<0.05$)

Table 2: Morphological and biochemical characteristics of the bacterial isolates from cassava waste water

IC	Cell shape	Cell appearance	Colour	Gram reaction	Motility	Indole	Coagulase	Citrate	Oxidase	Catalase	Spore	Starch hydrolysis	Sugar fermentation			PO
													Glucose	Lactose	Sucrose	
CWB1	Rd	Ch	W	+	+	-	-	+	-	+	+	+	A	A	+	*Bacillus cereus*
CWB2	Rd	Ch	W	+	+	-	-	+	+	+	+	+	AG	AG	+	*Bacillus subtilis*
CWB3	Rd		G	-	+	-	-	+	+	+	-	-	AG	-	-	*Pseudomonas aeruginosa*
CWB4	Co	Ch	Y	+	+	-	-	-	+	+	-	-	A	A	-	*Micrococcus* sp.
CWB5	Rd	Ch	CW	+	-	-	-	-	-	-	-		A	AG	+	*Lactobacillus acidophilus*

Rd: Rod, Co: Cocci, Ch: Chains, W: White, G: Green, Y: Yellow, CW: Creamy white, +: Positive, -: Negative, A: acid production, AG: Acid-gas production, IC: Isolate code, PM: Probable organisms

Table 3: Primary screening of bacterial isolates for cellulase production

Isolates	Diameter (mm)
Bacillus cereus	23
Bacillus subtilis	28
Pseudomonas aeruginosa	20
Micrococcus sp.	17
Lactobacillus acidophilus	11

Table 4: Secondary screening of *Bacillus subtilis* for cellulase production

Bacillus strains	Protein content (μmol mL^{-1})	Enzyme activity (mg mL^{-1})	Specific enzyme activity (μmol/min/mg)
Bacillus subtilis CWW1	0.0364 ± 0.12^{b}	0.16 ± 0.14^{a}	$4.52 \pm 0.11 \pm 0.11^{a}$
Bacillus subtilis CWW2	0.0272 ± 0.22^{a}	0.56 ± 0.11^{b}	$20.40 \pm 0.11 \pm 0.13^{b}$

Means with the same superscript letters along the same column are not significantly different ($p<0.05$)

Table 5: Proximate composition of fermented and unfermented cassava peels (%)

Sample	Fiber	Protein	Fat	Ash	CHO	Moisture
Unfermented cassava peels	21.62 ± 0.04^{b}	1.20 ± 0.13^{a}	1.81 ± 0.01^{a}	2.53 ± 0.03^{b}	26.62 ± 0.16^{b}	11.34 ± 0.13^{a}
Fermented cassava peels	16.91 ± 0.03^{a}	5.60 ± 0.11^{b}	1.72 ± 0.20^{a}	1.52 ± 0.00^{a}	22.33 ± 0.04^{a}	49.04 ± 0.10^{b}

Means with the same superscript letters along the same column are not significantly different ($p<0.05$)

of 20.0 mm and the least value of 11.00 mm was shown by *L. acidophilus*. Table 4 shows the specific enzyme activity exhibited by two strains of *B. subtilis*. The isolate designated *B. subtilis* CWW2 had higher specific enzyme activity of 20.40 μmol/min/mg, while the second strain of *B. subtilis* with a code CWW1 had lesser value.

Proximate composition of CTCP: Table 5 shows the proximate composition of CTCP. The crude protein increased from 1.20% in the unhydrolyzed sample to 5.60% in the sample hydrolyzed with concentrated enzyme. However, there was decrease in the crude fiber, fat and carbohydrate (CHO) of CTCP when compared with untreated samples. Crude fiber and CHO decreased from 21.62 and 26.62% in untreated samples to 16.91 and 22.33% in CTCP, respectively.

Fiber composition of CTCP: Fiber constituents of CTCP are shown in Table 6. There was decrease in all the fiber constituents of CTCP evaluated. Lignin, cellulose and hemicelluloses decreased from 6.10, 31.00 and 27.00% in unhydrolyzed samples to 3.70, 26.00 and 16.00% in CTCP, respectively.

Table 6: Fiber composition of fermented and unfermented cassava peels (%)

	Fiber composition (%)		
Samples	Lignin	Cellulose	Hemicellulose
Unfermented cassava peels	6.10 ± 0.44	31.00 ± 0.14	27.00 ± 0.23
Fermented cassava peels	3.70 ± 0.23	26.00 ± 0.22	16.00 ± 0.14

Means with the same superscript letters along the same column are not significantly different ($p<0.05$)

Table 7: Anti-nutrients composition of fermented and unfermented cassava peels (mg kg^{-1})

	Anti-nutrient composition		
Samples	Cyanide	Oxalate	Phytate
Unfermented cassava peels	7.35 ± 0.17^{b}	0.54 ± 0.31^{b}	474.40 ± 0.22^{b}
Fermented cassava peels	0.10 ± 0.55^{a}	0.21 ± 0.12^{a}	172.00 ± 0.07^{a}

Means with the same superscript letters along the same column are not significantly different ($p<0.05$)

Table 8: Mineral composition of fermented and unfermented cassava peels (cmol kg^{-1})

	Mineral composition					
Sample	Na	K	Ca	Mg	P	Fe
Unfermented cassava peels	412.00±0.01[a]	375.00±0.00[a]	112.00±0.01[a]	321.40±0.11[a]	171.25±0.00[a]	112.21±0.02[b]
Fermented cassava peels	517.00±0.01[b]	498.00±0.02[b]	212.50±0.02[b]	323.80±0.02[b]	175.12±0.00[b]	101.23±0.01[a]

Means with the same superscript letters along the same column are not significantly different ($p<0.05$)

Table 9: Vitamins composition of fermented and unfermented cassava peels (mg kg^{-1})

Sample	Vitamin A	Vitamin C
Unfermented cassava peels	0.02.0±0.01[a]	0.27.0±0.03[a]
Fermented cassava peels	0.02.0±0.00[a]	0.43.0±0.01[b]

Means with the same superscript letters along the same column are not significantly different ($p<0.05$)

Anti-nutritional composition of CTCP: In Table 7, there was remarkable decrease in the cyanide, oxalate and phytate contents of CTCP. Cyanide, oxalate and phytate declined in CTCP by approximately 99, 61 and 64%, respectively when compared with unhydrolyzed.

Mineral composition of CTCP: Table 8 reveals the mineral composition of CTCP. The treatment of cassava peels with concentrated enzyme led to an increase in all the minerals except for Fe^{2+} where a slight decrease was observed.

Vitamin composition of CTCP: The vitamin composition of CTCP is presented in Table 9. The vitamin C content of enzyme-treated sample was higher than the value obtained from untreated sample.

DISCUSSION

In this study, concentrated cellulase from a bacterium isolated from cassava waste water improved the nutritional contents of cassava peels. Cassava peels have been said to be susceptible to biodegradation and the susceptibility of cassava peels to biodegradation depends on the cassava varieties[21]. The use of low-cost agricultural wastes such as cassava peels in place of expensive ingredient in animal feed formulation is a mean by which the cost of livestock production is reduced. Although the use agrowastes as feed ingredients is being discouraged by many animal nutritionists because of high molecular compounds embedded in them that are not susceptible to the digestive enzymes in the gut of livestock. Hence, the animals fed with feeds made with untreated agricultural wastes are denied vital nutrients. Therefore, for the animals to derive maximum dietary benefits from the feeds, the ingredients must be transformed or broken down by microbial enzymes to unlock the available nutrients. The use of mannanase from selected fungi in the bioconversion of cassava peels and palm kernel cake have been documented and reported to improve their nutritional contents[10,11]. However, the use of bacteria in bioconversion of agricultural wastes has not been reported.

There was an increase in the bacterial population with increase in the fermentation time in this study. In contrast to the finding from this study, Olaniyi[22] reported a varied bacterial population when *Delonix regia* seeds an underutilized seed was subjected to submerged fermentation for 72 h; bacterial population increased for the first 48 h and beyond this, a decline was observed in the population. Array of bacteria were isolated from cassava wastes water and identified. Isolation of different bacteria from cassava waste water has been reported[23-25]. Arotupin[23] and Oboh[24] in separate studies isolated arrays of amylase-producing bacteria from cassava waste water. Human activities and lack of practicable control measures in the discharge of the waste water into the environment might account for the presence of these organisms in these samples. The presence of growth promoting factors such as; fermentable sugars, starch, cellulose and essential elements in cassava waste water might enhance its colonization by these organisms. These organisms may probably have emanated from soil and processing materials used in the processing of cassava tubers into other products while the variations of the isolates may be due to the handling process and the prevailing environmental conditions[25,26].

The bacterial isolates from cassava waste water exhibited varied enzyme activities both in the solid and liquid media supplemented with CMC as an inducer. The cellulase production on CMC agar media and liquid cellulase production media had been documented for *Bacillus pumilus*[27], *Pseudomonas fluorescens*[28]; *Cellulomonas* sp.[29], *Paenibacillus* sp.[13] and many more. The variations of enzyme activities shown by the bacterial might depend on the slight variation in the DNA sequence of the organisms[14,30].

There was an increase in the protein content of the cassava peels subjected to concentrated cellulase treatment from *B. subtilis*. The increase in protein contents of some enzyme hydrolyzed agricultural wastes has been reported. Khanongnuch *et al.*[9] partially purified cellulase and

mannanase and treated copra meal and it's led to the improvement of its nutritive values while corn-soybean meal was treated by Jackson *et al.*[31] with β-mannanase and evaluates its effects on laying hen performance. In separate studies conducted by Olaniyi[10] and Saenphoom *et al.*[11], palm kernel cake and cassava peels were hydrolyzed with partially purified cellulase and mannanase respectively and resulted into increase in their protein contents. The increase in protein contents of fermented cassava peels had been reported by different investigators[8,24]. One of the factors that could account for apparent increase in the protein content of the peels might be due involvement of cellulase which is proteinous in nature coupled with the fact that the concentrated enzyme might have increased the bioavailability of the protein hitherto lockup by the plant cell walls[10,11]. There was significant decrease in the fiber content of enzyme-treated sample when compared with raw sample.

There was significant decrease in the fibre content of enzyme-treated cassava peels when compared with untreated sample. The reports of Olaniyi *et al.*[2] and Olaniyi[10] are in agreement with the finding from this study. According to Olaniyi[10], approximately 64% in the crude fiber of enzyme-treated palm kernel meal was observed. Similarly, Olaniyi *et al.*[2] reported 13.75 and 29.70% reduction in crude fiber of cassava peels and corn cob respectively subjected to enzyme treatment. The reduction in the crude fiber in treated sample might be due to the presence of crude-fiber degrading enzymes that was produced and partially purified alongside with enzyme of interest. It can also be deduced that the concentrated cellulase was effective in degrading cellulose which is the major constituent of cassava peels[10,11,32]. The fiber compounds namely cellulose, hemicelluloses and lignin contents declined in the sample treated with concentrated cellulase. The finding from this study agrees with the observations of Albores *et al.*[33] who reported reduction in the lignin, hemicellulose and cellulose of fungal enzyme-treated palm kernel cake. The presence of hydrolyzing and oxidizing enzymes in the enzyme solution may accounts for the reduction in these compounds[10,34].

The treatment of cassava peels with concentrated cellulase led to the reduction of all anti-nutrient compounds in the sample. The reports of Olaniyi *et al.*[2] and Olaniyi[10] confirmed the findings from this current investigation. Olaniyi[10] subjected cassava peels and corn cob to enzyme treatment and phytate, tannin and cyanide contents reduced by approximately 29, 7 and 58%, respectively. The reduction in these anti-nutritional compounds might be attributed to the action of certain hydrolases secreted with cellulolytic enzyme[10,34]. Ojokoh *et al.*[35] reported reductions in anti-nutrient compounds of fermented groundnut and popcorn, while Aro[1] reported similar reduction in these compounds for fermented cassava tuber. Similarly, Wedad *et al.*[36] and Omid *et al.*[37] reported reductions in fermented sorghum cultivars and fermented canola meal, respectively in their separate investigations. The mineral contents of the sample subjected to enzyme treatment were higher than the value obtained from the control (untreated sample). An observation similar to this was reported by Olaniyi *et al.*[2] for cassava peels and corn cob subjected to enzyme treatment. Ojokoh *et al.*[35] and Akinyele *et al.*[38] reported for some fermented products. The increase might be attributed to the presence of some micro-elements that form part of the biological macromolecules which were released from the cellular structure into enzyme solution.

The vitamin C content of enzyme treated sample was higher that the untreated sample. The findings from this study are not consistent with those reported by Osman *et al.*[39], who found that fermentation, resulted in a marked decrease in vitamin contents of some fermented products. According to Makun *et al.*[40], cereals are rich sources of minerals, vitamins, carbohydrates, oils and proteins, but when processed through wet-milling and fermentation majority of the nutrients especially water-soluble vitamins are lost leaving mostly carbohydrates and are therefore grown mainly for energy.

CONCLUSION

This research revealed that bacteria associated with cassava waste water were good producer of cellulase. The treatment of cassava peels with concentrated cellulase resulted in the improvement of its nutritional and vitamin contents. There was also appreciable decrease in the anti-nutritional and fiber compounds. It is therefore recommended that enzyme-treated cassava peels regarded as having no economic value could be integrated into animal nutrition.

SIGNIFICANCE STATEMENT

This present study aimed at evaluating the hydrolytic effects of partially purified cellulase from *Bacillus subtilis* on the nutritional quality of cassava peels. This study revealed the presence of cellulase-producing bacteria from cassava waste water and improvement in the nutritional contents of cassava peels hydrolyzed with concentrated cellulase.

REFERENCES

1. Aro, S.O., 2008. Improvement in the nutritive quality of cassava and its by-products through microbial fermentation. Afr. J. Biotechnol., 7: 4789-4797.
2. Olaniyi, O.O., E.O. Bankefa, I.O. Folasade and T.V. Familoni, 2015. Nutrient enrichment of mannanase-treated cassava peels and corn cob. Res. J. Microbiol., 10: 533-541.
3. Murugan, K., Yashotha, K. Sekar and S. Al-Sohaibani, 2012. Detoxification of cyanides in cassava flour by linamarase of *Bacillus subtilis* KM05 isolated from cassava peel. Afr. J. Biotechnol., 11: 7232-7237.
4. Ojo, A. and E.A. Akande, 2013. Quality evaluation of 'gari' produced from cassava and sweet potato tuber mixes. Afr. J. Biotechnol., 12: 4920-4924.
5. Aro, S.O., V.A. Aletor O.O. Tewe A.N. Fajemisin B. Usifo and J.A. Adesida, 2008. Studies on the nutritional potentials of Cassava Tuber Wastes (CTW) collected from a factory. Proceedings of the 4th Annual Conference of SAAT, May 21, 2008, Akure, Nigeria, pp: 86-92.
6. Nwafor, O.E. and F.E. Ejukonemu, 2004. Bioconversion of cassava wastes for protein enrichment using amylolytic fungi-A preliminary report. Global J. Pure Applied Sci., 10: 505-507.
7. Busari, M.B., E.O. Ogbadoyi, O.A.Y. Daudu, I.M. Animashahun, L. Yusuf and A.A. Lateef, 2013. Effect of natural and combined fungal fermentation on phytate, tannin and some mineral contents of corn cobs. Int. J. Applied Biol. Res., 5: 82-89.
8. Adeleke, B.S., B.J. Akinyele, O.O. Olaniyi and Y.A. Jeff-Agboola, 2017. Effect of fermentation on chemical composition of cassava peels. Asian J. Plant Sci. Res., 7: 31-38.
9. Khanongnuch, C., C. Sa-nguansook and S. Lumyong, 2006. Nutritive quality of β-mannanase treated copra meal in broiler diets and effectiveness on some fecal bacteria. Int. J. Poult. Sci., 5: 1087-1091.
10. Olaniyi, O.O., 2014. Effect of beta-mannanase treatment on nutritive quality of palm kernel meal. Afr. J. Microbiol. Res., 8: 2405-2410.
11. Saenphoom, P., J.B. Liang, Y.W. Ho, T.C. Loh and M. Rosfarizan, 2011. Effect of enzyme treatment on chemical composition and production of reducing sugars in palm (*Elaeis guineenis*) kernel expeller. Afr. J. Biotechnol., 10: 15372-15377.
12. Holt, J.G., N.R. Kreig, P.H.A. Sneath, J.T. Staley and S.T. Williams, 1994. Bergey's Manual of Determinative Bacteriology. 9th Edn., Lippincott Williams and Wilkins, Baltimore, MD., USA., ISBN-13: 9780683006032, pp: 136-141.
13. Arotupin, D.J. and O.O. Olaniyi, 2013. Screening and identification of mannanase-producing fungi isolated from selected agricultural wastes. Br. Microbiol. Res. J., 3: 635-644.
14. Miller, G.L., 1959. Use of dinitrosalicylic acid reagent for determination of reducing sugar. Anal. Chem., 31: 426-428.
15. AOAC., 2012. Official Methods of Analysis of AOAC International. 19th Edn., AOAC International, Washington, DC., USA., pp: 426-428.
16. Van Soest, P.J., J.B. Robertson and B.A. Lewis, 1991. Methods for dietary fiber, neutral detergent fiber and nonstarch polysaccharides in relation to animal nutrition. J. Dairy Sci., 74: 3583-3597.
17. De Boland, A.R., G.B. Garner and B.L. O'Dekk, 1975. Identification and properties of phytate in cereal grains and oilseed products. J. Agric. Food Chem., 23: 1186-1189.
18. Price, M.L., S. van Scoyoc and L.G. Butler, 1978. A critical evaluation of the vanillin reaction as an assay for tannin in sorghum grain. J. Agric. Food Chem., 26: 1214-1218.
19. Day, Jr. R.A. and A.L. Underwood Jr., 1986. Quantitative Analysis. 5th Edn., Prentice-Hall Publication, London, UK., ISBN-13: 9780137467280, Pages: 774.
20. Obadoni, B.O. and P.O. Ochuko, 2002. Phytochemical studies and comparative efficacy of the crude extracts of some haemostatic plants in Edo and Delta States of Nigeria. Global J. Pure Applied Sci., 8: 203-208.
21. Aransiola, M.N. and O.E. Fagade, 2015. Studies on the biodegradation of cassava peels (*Manihot esculenta*) and rice straw (*Oryza sativa*) by some selected microorganisms. Int. J. Plant Sci. Ecol., 1: 124-130.
22. Olaniyi, O.O., 2018. Microbiological, nutritional quality and antioxidant activity of fermented *Delonix regia* seeds. J. Food Saf. Hyg., 4: 69-75.
23. Arotupin, D.J., 2007. Evaluation of microorganisms from cassava waste water for production of amylase and cellulase. Res. J. Microbiol., 2: 475-480.
24. Oboh, G., 2006. Nutrient enrichment of cassava peels using a mixed culture of *Saccharomyces cerevisae* and *Lactobacillus* sp. solid media fermentation techniques. Electron. J. Biotechnol., 9: 46-49.
25. Olowoyo, O.O., F.A. Akinyosoye and F.C. Adetuyi, 2001. Micro-organisms associated with some cassava (*Manihot esculenta*, Crantz) products. J. Res. Rev. Sci., 2: 10-14.
26. Kotchoni, O.S., O.O. Shonukan and W.E. Gachomo, 2003. *Bacillus pumilus BpCRI 6*, a promising candidate for cellulase production under conditions of catabolite repression. Afr. J. Biotechnol., 2: 140-146.
27. Bakare, M.K., I.O. Adewale, A.O. Ajayi, A.L. Okoh and O.O. Shonukan, 2005. Regulatory mutations affecting the synthesis of cellulase in *Pseudomonas fluorescens*. Afr. J. Biotechnol., 4: 838-843.
28. Yin, L.J., P.S. Huang and H.H. Lin, 2010. Isolation of cellulase-producing bacteria and characterization of the cellulase from the isolated bacterium *Cellulomonas* sp. YJ5. J. Agric. Food Chem., 58: 9833-9837.
29. Islam, F. and N. Roy, 2018. Screening, purification and characterization of cellulase from cellulase producing bacteria in molasses. BMC Res. Notes, Vol. 11, No. 1. 10.1186/s13104-018-3558-4

30. Olaniyi, O.O. and O. Adebowale, 2017. Production and partial purification of beta-mannanase from *Aspergillus niger* associated with Ilaje Lake, Ondo State, Nigeria. J. Bacteriol. Mycol.: Open Access, 5: 281-285.
31. Jackson, M.E., D.W. Fodge and H.Y. Hsiao, 1999. Effects of β-mannanase in corn-soybean meal diets on laying hen performance. Poult. Sci., 78: 1737-1741.
32. Lawal, T.E., E.A. Iyayi, B.A. Adeniyi and O.A. Adaramoye, 2010. Biodegradation of palm kernel cake with multienzyme complexes from fungi and its feeding value for broilers. Int. J. Poult. Sci., 9: 695-701.
33. Albores, S., M.J. Pianzzola, M. Soubes and M.P. Cerdeiras, 2006. Biodegradation of agroindustrial wastes by *Pleurotus* spp. for its use as ruminant feed. Elect. J. Biotechnol., 9: 215-220.
34. Akinfemi, A., 2012. Chemical composition and nutritive value of fungal treated acha straw. Agric. Sci. Technol., 4: 120-124.
35. Ojokoh, A.O., A.B. Abiola and R.T. Lawal, 2012. Changes in nutrient and antinutrient composition of popcorn and groundnut composite flour subjected to solid substrate fermentation. Afr. J. Agric. Res., 27: 3439-3445.
36. Wedad, W., H. Abdelhaleem, A.H. El Tinay, A.I. Mustafa and E.E. Babiker, 2008. Effect of fermentation, malt-pretreatment and cooking on antinutritional factors and protein digestibility of sorghum cultivars. Pak. J. Nutr., 7: 335-341.
37. Omid, S., F. Mehrdad, C. Chris, Y. Bagher and M.S. Masoomeh, 2012. Study on the effect of solid state fermentation with *Aspergillus niger* on antinutritional factors of canola protein concentrate with aim of using in the diet of rainbow trout (*Oncorhynchus mykiss*). Proceedings of the 11th International and the 4th National Congress on Recycling of Organic Waste in Agriculture, April 26-27, 2012, Isfahan, Iran.
38. Akinyele, B.J., O.O. Olaniyi and D.J. Arotupin, 2011. Bioconversion of selected agricultural wastes and associated enzymes by *Volvariella volvacea*: An edible mushroom. Res. J. Microbiol., 6: 63-70.
39. Osman, M.A., I.E. Abdel Rahman and H.A. Dirar, 2010. Biochemical changes occurring during fermentation of camel milk by selected bacterial starter cultures. Afr. J. Biotechnol., 9: 7331-7336.
40. Makun, H.A., S.T. Anjorin, B. Moronfoye, F.O. Adejo and O.A. Afolabi *et al*., 2010. Fungal and aliatoxin contamination of some human food commodities in Nigeria. Afr. J. Food Sci., 4: 127-135.