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Comparative Isolation and Characterization of Bacterial and Fungal Communities Associated with Peels of Selected Cassava Varieties

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ABSTRACT

In Nigeria, Cassava (*Manihot esculenta* Crantz) processing generates large quantities of peels that are often regarded and discarded as waste, contributing to environmental pollution despite their rich organic composition. This has necessitated the need for development of strategies for waste management, bioconversion and microbial resource exploitation. This study aimed to isolate and characterize bacteria and fungi from three cassava varieties (CV) peels. Bacteria and fungi were isolated across three CV peels namely Local Cassava (LCV), Black Cassava (BCV) and White Cassava (WCV) using pour plate method as follows: Freshly generated cassava peels collected under aseptic conditions were gently pounded to increase its surface area, then subjected to serial dilution and culture on selective and differential media. Distinct bacterial and fungal colonies were purified and characterized using standard morphological, cultural, and biochemical techniques, while fungal identification was further supported by microscopic examination of spore and hyphal structures. Results showed that five bacteria; *Bacillus subtilis, Staphylococcus aureus, Bacillus cereus, Pseudomonas aeruginosa and Escherichia coli* and seven fungi; S*accharomyces cerevisiae, Torula herbarum, Schizosaccharomyces pombe, Aspergillus tubingensis, Penicillium chrysogenum, Rhizopus stolonifer* and *Mucor mucedo* were isolated across the three CV peels. LCV had highest percentage prevalence of bacteria and fungi of the three CV peels and highest value of moisture, Carbohydrate, crude protein, and lowest cyanide content of the three CV. The findings from this study underscore the ecological significance of cassava peels as reservoirs of metabolically active microorganisms which can be employed in various beneficial biotechnological processes such as single cell protein production. It was concluded that microorganisms of both bacteria and fungi origin can be isolated from cassava varieties peels with diverse biotechnological potential.

Keywords: Cassava Peels, Bioconversion, Biochemical Techniques, Microscopic Examination.

1.0 Introduction

Cassava (*Manihot esculenta* Crantz) is one of the most important staple crops cultivated in tropical and subtropical regions of the world, particularly in sub-Saharan Africa. Nigeria remains the largest producer globally, contributing significantly to both food security and agro-industrial development (Food and Agriculture Organization [FAO], 2023). During the processing of cassava into products such as gari, fufu, starch e.t.c. large volumes of processing residues e.g. peels and bagasse are generated. These residues represent both a waste management challenge and an opportunity for value-addition (Otekunrin, 2024).

Cassava peels make up the largest proportion of the processing waste produced during the processing of cassava into other forms and can

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make up as much as 10-25% of the weight of the fresh cassava root (Otekunrin *et al*., 2024). Using the estimated weight of the cassava produced in Nigeria, we can safely conclude that million tones of cassava peels are produced in Nigeria as waste every year. The large-scale processing of cassava in factories such as gari and starch factories produces large amounts of cassava peels that prove difficult to dry in the rain and hence often accumulate as heaps of cassava peels (Oyeyinka and Oyeyinka, 2018).

Cassava peels are rich in carbohydrates, fiber, residual starch, and minor amounts of protein and minerals. However, their utilization as feed or industrial substrate is limited by high fiber content, low protein levels, and the presence of anti-nutritional factors such as cyanogenic glycosides (Chandra *et al*., 2020). Interestingly, these same characteristics make cassava peels an excellent substrate for microbial growth.

Microorganisms play a central role in the biodegradation and bioconversion of agro-industrial residues. Through metabolic activities, bacteria and fungi secrete extracellular enzymes such as cellulases, amylases, proteases, and ligninases that break down complex polymers into simpler compounds (Wiebe, 2020). These metabolic capabilities have attracted scientific interest in harnessing indigenous microorganisms for value-added applications, including enzyme production, bioethanol generation, waste detoxification, and single cell protein (SCP) production (Kumar *et al*., 2021). However, the efficiency of such biotechnological processes depends largely on the proper isolation, identification, and characterization of competent microbial strains.

Microbial isolation from cassava peels provide insight into the microbial ecology of this agro-waste. Indigenous microbes are often better adapted to the substrate's physicochemical conditions, including fluctuating moisture content, cyanide levels, and high carbohydrate concentration. Characterizing these isolates morphologically, biochemically, and molecularly allows researchers to determine their taxonomic identity, metabolic potential, safety status, and suitability for industrial application. For instance, species of *Bacillus*, *Aspergillus*, *Saccharomyces*, and *Trichoderma* have been widely reported in cassava waste fermentation systems due to their strong enzymatic profiles and adaptability (Becker, 2007; Okafor, 2018).

Bacterial isolates such as *Bacillus subtilis* are known for their ability to produce thermostable enzymes and degrade complex carbohydrates efficiently. Fungal isolates, particularly filamentous fungi like *Aspergillus* spp., exhibit robust lignocellulolytic activity, enabling the breakdown of fibrous materials in cassava peels (Wiebe, 2020). Proper characterization involves cultural examination, Gram staining (for bacteria), microscopic morphology (for fungi), biochemical profiling, and increasingly, molecular techniques such as 16S rRNA and ITS region sequencing. These approaches ensure accurate identification and reduce the risk of misclassification, which is critical when isolates are intended for feed or food applications.

Understanding the diversity and functional attributes of microorganisms present in cassava varieties' peels also contributes to sustainable waste management strategies. Instead of viewing cassava peels as mere waste, they can be transformed into bioresources through microbial intervention. Isolation and characterization therefore serve as foundational steps in developing environmentally friendly and economically viable bioprocesses. This approach aligns with global sustainability goals promoting circular bioeconomy and waste valorization (FAO, 2023).

Given the large volume of cassava peels generated in Nigeria and other cassava-producing nations, there is a pressing need to systematically study their associated microbial communities. Such research not only supports environmental management but also opens opportunities for protein enrichment, enzyme production, and other biotechnological innovations. Therefore, the present study aim to isolate, characterize and compare microbial diversity across cassava varieties peel.

2.0 Materials and Method

Study Area

This study was carried out in Ilorin, Kwara State, located in the north-central geopolitical $4^0$30'E and $4^0$45'E. Ilorin is located within the transitional zone between the forest region of the south and savannah region of the North which gives it mixed ecological character. The city is bordered to the east by Kogi state, to the north by Niger state, to the South by Osun and Ekiti state and to the West by Oyo state (Amao *et al*., 2024).

Sample Site

Three freshly harvested cassava varieties peels were collected from a farmland in Gambari village located in Surulere area of Oyo state, in the Southwestern region of Nigeria. Geographically Gambari village lies on the between Latitude 8º16'N and Longitude 4º19'E.

Sample Collection

Three varieties of freshly harvested cassava tubers were collected from a farmland at Gambari town in Oyo State of Nigeria. The collected cassava tubers were identified at the herbarium section of Plant Biology Department, University of Ilorin with voucher number UILH/001/1449/2023.The three varieties collected were identified as Local Cassava variety (LCV) popularly known in the southwestern Nigeria as Oko Iyawo, the Black Cassava Variety (BCV) and the White Cassava Variety (WCV).

The identified tubers were peeled with sterile stainless-steel knife and stored in clean and dried polythene bag at ambient temperature and labelled accordingly (Adenike *et al*., 2015).

Isolation of Bacteria and Fungi from Cassava Varieties Peels

Facultative anaerobic bacteria and fungi were isolated from each cassava variety peels using Pour plate method, following serial dilution of the samples. One milliliter of each cassava variety peels sample (already pounded with mortal and pestle to increase its surface area) was added to test tube containing 9mL of sterile distilled water to obtain a dilution factor of 10^{-1}. The test tube was vigorously mixed and 1mL of the solution in the tube was transferred to another test tube containing 9mL of sterile distilled water to obtain a dilution factor of 10^{-2}. The process was repeated up to 10^{-6} dilution and 0.1mL of 10^{-5} and 10^{-6} dilutions was plated on yeast extract medium, potato dextrose agar and nutrient agar plates for yeast, mold and bacteria respectively using pour plate method. Yeast and mold plates were incubated at room temperature for 3-5days while bacterial culture plates were incubated at 37°C for 24-48 hours. Sub-culturing of distinct colonies on Yeast extract medium, PDA plate and nutrient agar plates were done by streak plate method. Pure cultures of bacterial isolates were maintained in glycerol while fungal isolates were maintained on Potato Dextrose Agar slant and stored in refrigerator at 4^0C till further analysis (Akinruli *et al.,* 2022).

Identification of Bacteria Isolated from Cassava Varieties Peel

Identification of bacteria isolates from cassava varieties peel were done using colonial morphology, cellular characteristics and biochemical test.

Colonial Morphology of Bacteria Isolates

Colonial morphological characteristics such as shape of colony, size, margin, elevation, texture and colour of discrete colonies on Nutrient agar were observed visually and recorded accordingly.

Cellular Morphology of Bacteria Isolates

Cellular morphology of each of the isolates was carried out using Gram staining technique. A thin smear of 18-24 hour old culture of each of the isolate was made on clean grease free slide using a sterile inoculating loop. Each smear was spread evenly and heat fixed by passing through a Bunsen burner flame after which the smear was flooded with crystal violet and allowed to stay for 60 seconds. Excess crystal violet stain was washed off with running water and then flooded with Gram's iodine for another 60 seconds to act as mordant. The Gram's iodine was washed off under running tap and 95% ethyl alcohol was then added to the smear for 10 seconds. Subsequently the smears were rinsed immediately using running water to stop decolourization process. A few drops of safranin solution were then used to counter stain the smear and was allowed to stay for another 60 seconds before rinsing off with

water. The slides were then allowed to dry after which they were observed under microscope using oil immersion. The colour, shapes and cell arrangements were observed for each of the isolate (Paray *et al*., 2023).

Physiological and Biochemical Characteristics of Bacteria Isolates

Biochemical tests were carried out in order to further identify and characterize the bacteria isolates and the biochemical test carried out include; Oxidase, Catalase, Citrate, Indole, Methyl Red, Sugar fermentation, Urease, Nitrate reduction and Voges Proskauer test using standard methods.

Characterization of Fungal Isolated from Cassava Varieties Peel

Characterization of fungal isolates from the cassava varieties peels were done microscopically. A drop of lactophenol cotton blue stain was placed aseptically on a clean grease free glass slides and a small portion of the fungal mycelium was carefully transferred from the edge of an actively growing colony onto the stain. The mycelium was gently teased using a sterile needle in order to spread the fungal structures after which a cover slip was placed over it ensuring there was no air bubbles. The slide was then examined under the microscope using ×40 objective of a light microscope and microscopic features such as hyphal septation, arrangement of conidiophores, shape and size of spores and presence of special structure were observed and compared with standard taxonomic keys for fungal identification (Samson *et al*., 2019).

Proximate Analysis of The Cassava Varieties Peel

The moisture content, ash, lipid, protein and total carbohydrate of the samples were determined as follows: The moisture content was determined by drying samples overnight at 105°C until constant weight was achieved. Crude protein content was analyzed by using the Kjeldahl method. Crude fat content was determined using the Soxhlet method the ash content was determined by ashing the samples overnight at 550°C. Carbohydrate content was calculated by difference; i.e. the sum total percentage of moisture, crude protein, crude fat and ash content were subtracted from 100% (Adenike *et al*., 2015). Cyanide content was carried out using Alkaline Picrate reagent prepared using a modification of the method described by Moriasi *et al.* (2017).

Statistical Analysis

All analyses were carried out in triplicate and data generated in this research were subjected to a one-way analysis of variance (ANOVA) and means were compared for significant differences at 5% significance level by the Duncan's multiple range test using the SPSS package version 22.0 software (IBM corp., Armonk, NY).

3.0 Results

Characterization of Bacteria Isolated from Cassava Varieties Peels

A total of twenty-six (26) colonies were obtained from Local Cassava Variety (LCV), White Cassava Variety (WCV) and Black Cassava Variety (BCV) Peels. These were further identified into five (5) distinct isolates following morphological characteristics which include; texture, shape, sizes, elevation, pigmentation and margination of colony. Based on cellular characteristics three (3) of the bacterial isolates were gram positive while the other two (2) isolates were gram negative. The morphological and cellular characteristics of the bacteria isolates are shown in Table 1 while the photomicrographs of cellular morphology of the isolates are presented in Plate 1A-E.

Table 1: Morphological and Cellular Characteristics of Bacteria Isolates

S/N	Sample Code	Colony Size (mm)	Colony Shape	Texture	Margin	Elevation	Pigmentation	Opacity	Edge	Gram Reaction
1	Isolate A	1.2	Circular	Smooth	Entire	Convex	Creamy	Opaque	Flat	+
2	Isolate B	2.6	Circular	Smooth	Irregular	Convex	Off-white	Opaque	Rough	+
3	Isolate C	1.7	Circular	Smooth	Entire	Convex	Yellow/Orange	Opaque	Shining	+
4	Isolate D	1.2	Irregular	Smooth	Entire	Convex	Grey white	Translucent	Rough	–
5	Isolate E	1.2	Irregular	Smooth	Entire	Convex	Grey white	Opaque	Rough	–

Key: + = Positive; – = Negative

Table 2: Biochemical Test of Bacteria Isolates

Shape	Cell arrangement	Pigmentation	Gram reaction	Motility	Endospore	Catalase	Oxidase	Coagulase	Indole	Citrate utilization	M R	V P	Gelatinase	Urease	Glucose	Lactose	Sucrose	Starch	H2S	Gas production	O_2 relationship	Suspected organism
Rods	Chain	-	+	-	-	-	+	-	-	+	+	-	+	+	+	-	-	-	+	+	FA	A
Rods	Pairs	-	+	+	+	+	+	-	-	+	-	+	+	-	+	-	+	+	-	-	FA	B
Cocci	Clustered	+	+	-	-	+	-	+	-	+	+	+	+	+	+	+	+	-	-	-	FA	C
Rods	Single	-	-	+	-	+	-	-	+	-	+	-	-	+	+	+	+	-	-	+	FA	D
Rods	Single	+	-	+	-	+	+	-	-	+	-	-	-	+	-	-	-	-	-	-	OA	E

KEY: A-*Bacillus subtilis;* B- *Bacillus cereus;* C- *Staphylococcus aureus* D- *Escherichia coli* E- *Pseudomonas aeruginosa*

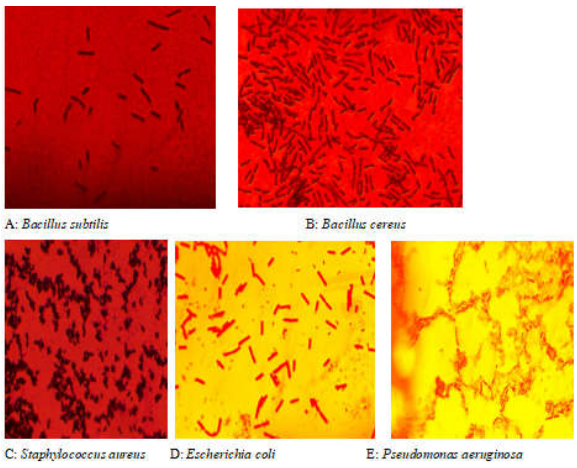

Plate 1 A-E: Photomicrograph of cellular morphology of Isolate A-E

Investigations on cassava peels have shown the presence of a microbial ecosystem consisting of bacteria and fungi with the potential to degrade polysaccharides, ferment, and detoxify. These microorganisms have huge potential in biotechnological applications.

Several studies have documented the prevalence of bacteria such as *Pseudomonas*, *Bacillus*, *Proteus*, and *Sporosarcina*, as well as fungi like *Aspergillus*, *Rhizopus*, *Trichoderma*, and yeasts in cassava varieties peels (Adedayo and Lateef, 2025).

The presence of opportunistic pathogens such as *Staphylococcus aureus, Pseudomonas sp.* aligns with reports that cassava wastes environment harbors mixed microbial population of public health concern. These organisms may not cause disease in healthy hosts but become infectious under favourable conditions. Their presence in cassava peels however, suggest a potential microbiological risk when such peels are utilized as animal feed or in biotechnological applications without adequate treatment (Adeolu and Adewoye, 2019).

Also, from an environmental point of view, the presence of *E. coli* in cassava peels contribute greatly to water and soil contamination which occurs when heaps of cassava peels are openly discharged into nearby lands or water bodies leading to the infiltration of such with leachates containing these pathogenic microorganisms. This not only alters soil microbial composition

but also introduces fecal indicator bacteria into aquatic environment, thereby compromising water quality and posing serious public health concerns (Otitoju *et al*., 2024).

Distribution and Percentage Prevalence of Bacterial Isolates

The Result obtained from this research indicated that Local Cassava Variety Peels (LCV) had the highest number of bacterial isolates and highest percentage prevalence of the isolates with isolates A, B, C, D, and E obtained followed by the Black Cassava Variety (BCV) which had all the isolates present except isolate E but in lower percentage prevalence compared to LCV. The least bacterial isolates and prevalence was recorded in White Cassava Variety (WCV) which had only isolate A present in it. The distribution of the isolate and percentage prevalence is presented in Table 3.

Table 3: Distribution and Percentage Prevalence of Isolated Bacteria from Cassava Varieties Peels

Cassava Variety	Isolate A	Isolate B	Isolate C	Isolate D	Isolate E
LCV (Local)	2 (40%)	2 (66.67%)	1 (50%)	1 (50%)	1 (100%)
WCV (White)	2 (40%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
BCV (Black)	1 (20%)	1 (33.33%)	1 (50%)	1 (50%)	0 (0%)
TOTAL	5 (100%)	3 (100%)	2 (100%)	2 (100%)	1 (100%)

Key: LCV – Local Cassava Variety; **WCV** – White Cassava Variety; **BCV** – Black Cassava Variety

Characteristics of Fungi Isolated from Cassava Varieties Peels

A total of seven (7) species of fungi were isolated from the three cassava varieties peels namely; LCV, BCV and WCV. The isolates were presumptively identified following microscopically and cultural characterization as *Saccharomyces cerevisiae, Torula herbarum, Schizosaccharomyces pombe, Aspergillus tubingensis, Penicillium chrysogenum, Rhizopus stolonifer* and *Mucor mucedo*.

Aspergillus tubingensis, though not commonly reported in literature plays an important role in primary degradation of cassava peels biomass through the secretion of extracellular enzymes such as cellulases, amylases and pectinases leading to conversion of cassava wastes into fermentable sugars and organic acids thereby enhancing substrate utilization and value addition. However, their presence may pose safety concerns due to their ability to secrete mycotoxins under favourable conditions especially in case of improper or poor storage (Abdulmumini *et al*., 2025).

Also, studies had revealed that *Rhizopus sp.* were frequently isolated alongside *Aspergillus*, further underscoring the role of saprophytic molds in degrading organic residues in cassava peels (Akinruli *et al*., 2022). This is in agreement with the result obtained in this study where *Rhizopus sp* was isolated alongside *Aspergillus sp*.

Recent studies showed that *Rhizopus stolonifera* possess strong amylolytic and fermentative capabilities, making them suitable for the bioconversion of agro-wastes into valuable metabolites like lactic acid. However, their excessive proliferation may lead to spoilage and reduce the nutritional value of cassava-based feed (Akter *et al*., 2025).

Saccharomyces cerevisiae plays a central role in the fermentative conversion of cassava peel substrate through metabolism of simple sugars into ethanol and other metabolites and it also contributes to microbial safety by competitively inhibiting and pathogenic organisms through acidification and production of antimicrobial metabolites. Recent studies have reported that Saccharomyces cerevisiae can ferment cassava wastes thereby improves its nutritional value, enhances protein enrichment and reduces antinutritional factors such as cyanogenic compounds (Kaewwongsa *et al*., 2024; Oliveira Junior *et al*., 2026).

In addition, *Schizosaccharomyces pombe* although less frequently reported in cassava peels ecosystems, contribute to fermentation and ecological stability, particularly under unfavourable environmental conditions such as low pH and high metabolites concentration. This organism complements the metabolic activities of *S. cerevisiae* leading to efficient sugar utilization and fermentation performance.

Frequency Distribution and Percentage Prevalence of Fungi Isolates

Results obtained in this study revealed that of the three cassava varieties Local Cassava Variety (LCV) had the highest frequency distribution and

percentage prevalence of all the seven fungi isolates, followed by WCV and then the Black Cassava variety BCV had the least frequency distribution and percentage prevalence of all the fungi isolates. The frequency distribution and percentage prevalence of fungi isolates is presented in Table 4. Also, photomicrograph of the hyphae of the fungal structure is presented in Plate 2 A-G

Table 4: Frequency of Distribution and Percentage Prevalence of Isolated Fungi from Cassava Varieties Peels

Cassava Variety	01	02	03	04	05	06	07
LCV	4 (57.14%)	2 (50%)	1 (50%)	3 (60%)	2 (100%)	2 (100%)	2 (66.6%)
WCV	2 (28.57%)	1 (25%)	0 (0%)	2 (40%)	0 (0%)	0 (0%)	1 (33.33%)
BCV	1 (14.29%)	1 (25%)	1 (50%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
TOTAL	7 (100%)	4 (100%)	2 (100%)	5 (100%)	2 (100%)	2 (100%)	3 (100%)

Key: 01 – *Saccharomyces cerevisae;* 02 – *Torulla herbarum;* 03 – *Schizosaccharomyces pombe;* 04 – *Aspergillus niger;* 05 – *Penicillum chrysogenum;* 06 – *Rhizopus stolonifera;* 07 – *Mucor mucedo.* **LCV** – Local Cassava Variety; **WCV** – White Cassava Variety; **BCV** – Black Cassava Variety

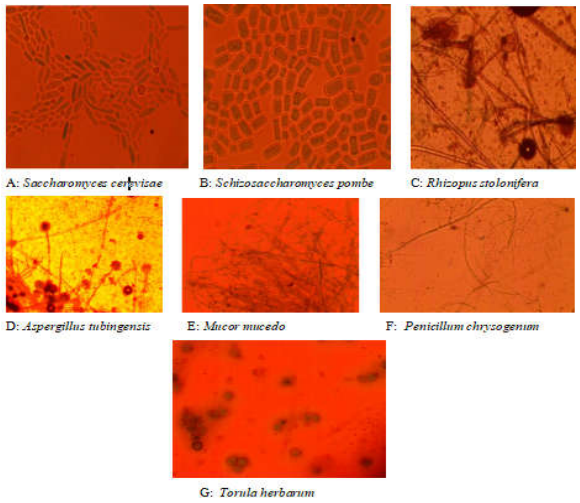

Plate 2 A-G: Photomicrograph of the hyphae structure of fungi isolates A-G.

Proximate Analysis of Cassava Varieties Peel

Proximate analysis of the three varieties of cassava peels showed percentage moisture content, carbohydrate, crude protein, crude fibre and ash content ranged from 10.91±1.30-12.28 ±1.82, 39.26±2.72[a]- 49.97±5.07[c], 0.22 ±o.o1-1.09 ±0.05 , 22.90 ±2.55-29.71 ±3.27 and 2.75 ±0.12-5.16 ±0.32 respectively as shown in Table 2.5 with Local Cassava Variety having highest value of moisture (12.28±1.82[c]), Carbohydrate (49.97±5.07[c]), crude protein(1.09±0.05[b]), and lowest cyanide content(9.45±1.53[b]). This is in agreement with the findings of Punniamorthy *et al*. (2021) which reported that the growth of microorganisms depends mainly on the nutritional composition of substrate. This can be the reason for the highest bacterial and fungal count recorded in LCV.

The proximate composition of cassava peel such as moisture content, carbohydrate content, and protein content has a significant impact on the use and value of cassava peels as substrate for microbial growth. A high moisture content and carbohydrate content in cassava peels will have a significant impact in producing more biomass yield by microbes (Miya *et al*., 2024). Olaniyan *et al*. (2025) noted that in using cassava peels as substrate for the cultivation of mushrooms such as oyster mushrooms, the proximate composition will have a positive impact in producing a high yield and a well-balanced nutritional composition.

Conclusion and Recommendations

This study successfully isolated and characterized bacterial and fungal species associated with peels from selected cassava varieties. The findings confirm that cassava peels, often regarded as agricultural waste, harbor diverse and viable microbial populations.

Table 5: Proximate Analysis of Raw Cassava Varieties Peels

Sample Code	Crude Protein (%)	Crude Lipid (%)	Moisture Content (%)	Total Ash (%)	Crude Fibre (%)	Carbohydrate (%)	Cyanide Level (mg HCN/100 g)
WC	0.97 ± 0.07[c]	11.88 ± 0.51[c]	12.06 ± 1.52[b]	5.12 ± 0.41[c]	29.71 ± 3.27[c]	40.26 ± 2.72[a]	10.00 ± 2.01[c]
BC	0.22 ± 0.01[a]	11.25 ± 0.13[b]	10.91 ± 1.30[a]	5.16 ± 0.32[b]	29.05 ± 3.05[b]	43.41 ± 3.43[b]	9.63 ± 1.20[a]
LVC	1.09 ± 0.05[b]	11.00 ± 0.75[a]	12.28 ± 1.82[c]	2.75 ± 0.12[a]	22.90 ± 2.55[a]	49.97 ± 5.07[c]	9.45 ± 1.53[b]

Means were separated using Duncan's Multiple Range Test. Means followed by the same superscript in the same row are not significantly different (P≤0.05). WC- white cassava variety BC- Black cassava variety LCV- local cassava variety (oko iyawo)

Both bacterial and fungal isolates exhibited distinct morphological, cultural, and cellular characteristics, indicating microbial diversity influenced by cassava variety, handling conditions, and environmental exposure.The predominance of certain Gram-positive bacterial isolates suggests their adaptability to the nutrient composition and ecological conditions of cassava peels. Similarly, the fungal isolates demonstrated characteristic colonial morphology typical of common environmental fungi associated with carbohydrate-rich substrates. The presence of these microorganisms highlights the dual nature of cassava peels: they can serve as valuable substrates for beneficial microbial processes such as fermentation and single cell protein production, yet they may also pose safety concerns if improperly handled or stored.

Based on the findings of this study, the following recommendations are proposed for future studies:

Further studies should incorporate molecular techniques such as 16S rRNA sequencing for bacteria and ITS region sequencing for fungi to achieve precise species-level identification.
Pathogenicity and toxin-producing potential of isolated fungi and bacteria should be evaluated to ensure safety, especially where cassava peels are intended for animal feed or fermentation purpose.
Beneficial isolates with desirable fermentation or enzyme-producing capabilities should be further screened and optimized for applications such as single cell protein production, enzyme production, and bio-conversion processes.

Conflict of interest

The authors declare that they have no conflict of interest.

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