



## Bacteriological Profile of Roasted Peeled Cashew Nuts Sold in Selected Markets in Keffi, Nasarawa State, Nigeria

By

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### Abstract

*This study evaluated the bacteriological profile of roasted peeled cashew nuts sold in selected markets within Keffi. Fifty wrapped samples of roasted peeled cashew nuts were randomly purchased from five major markets, namely Sambisa, Angwan Lambu, Tsohon, Neighbourhood, and Kara markets. Standard microbiological procedures involving serial dilution, spread plate technique, Gram staining, and biochemical characterization were employed for the isolation and identification of bacterial contaminants. The total viable count, total coliform count, total faecal coliform count, and Staphylococcus aureus count were determined using selective culture media. Results revealed varying degrees of bacterial contamination across all sampled markets. Sambisa Market recorded the highest microbial load with total viable count of  $8.3 \times 10^5$  cfu/ml, total coliform count of  $6.1 \times 10^5$  cfu/ml, total faecal coliform count of  $3.7 \times 10^5$  cfu/ml, and Staphylococcus aureus count of  $2.2 \times 10^5$  cfu/ml. The bacterial isolates identified included Pseudomonas spp., Klebsiella spp., Bacillus spp., Escherichia coli, Enterobacter spp., and Staphylococcus aureus. The findings demonstrated that roasted peeled cashew nuts sold within Keffi metropolis may constitute a potential public health risk due to microbial contamination.*

**Keywords:** Bacterial contamination; Food safety; Microbial load; Ready-to-eat foods; Roasted cashew nuts

## INTRODUCTION

Cashew nut from the Cashew tree is an economically important tropical crop native to north-eastern Brazil. The Portuguese introduced the crop to other parts of the world in 1578, after which it became widely cultivated across tropical regions under diverse environmental conditions (Wahauwuele et al., 2022). Today, cashew production represents a strategic agricultural enterprise in many African countries because of its contribution to income generation, food security, and international trade. Côte d'Ivoire, for example, is currently recognized as one of the world's leading producers and exporters of cashew kernels, exporting 30,022 tonnes of kernels in 2021, which was twice the quantity exported in 2020 and four times that of 2019 (Mukuddempetersen et al., 2020). The increasing demand for cashew products has consequently stimulated their processing and commercialization in local and international markets.

Among processed cashew products, roasted peeled cashew nuts are widely consumed as ready-to-eat snacks because of their desirable taste, nutritional value, and convenience. However, concerns regarding their microbiological safety have continued to increase due to the possibility of contamination during processing, handling, storage, transportation, and vending. Several studies have shown that roasted nuts sold in markets may harbor harmful microorganisms resulting from poor hygienic practices and inadequate storage conditions. In Nigeria, significant bacterial and fungal contamination has been reported in both raw and roasted nuts, with microbial loads exceeding acceptable standards and the presence of organisms such as Enterococcus, Bacillus, and Staphylococcus species, indicating poor microbiological quality of market-sold products (Omorodion and Oyedele, 2024). Similarly, contamination of roasted snacks with toxigenic fungi and mycotoxins, including aflatoxins, has been documented and

remains a major food safety concern (Nnam and Eruteya, 2023).

The public health implications of contaminated ready-to-eat foods are substantial. Aflatoxin exposure has been associated with both acute and chronic health conditions, including immune suppression and increased risk of liver cancer following prolonged consumption of contaminated foods (Salisu et al., 2020). In addition to fungal toxins, bacterial pathogens commonly associated with ready-to-eat foods, such as *Staphylococcus aureus* and coliform bacteria, may cause foodborne illnesses when consumed in contaminated products. Studies on ready-to-eat foods in Nigeria have consistently reported high microbial loads above recommended limits, emphasizing the need for improved food safety surveillance and control measures (Izah et al., 2022). Furthermore, advanced microbial profiling of ready-to-eat foods has revealed diverse bacterial contaminants and biotoxins, highlighting the limited surveillance data available for specific products such as roasted cashew nuts (Makinde et al., 2023).

Although food safety interventions including improved hygienic practices, vendor education, enhanced processing standards, and regulatory monitoring have been recommended to reduce contamination in ready-to-eat foods, implementation of these measures remains inconsistent in many local markets (Nnam and Eruteya, 2023; Omorodion and Oyedele, 2024). In addition, the effectiveness of these interventions in ensuring the microbiological safety of roasted peeled cashew nuts sold in Nigerian markets is still unclear (Solomon et al., 2025). More specifically, there is limited information on the bacteriological profile of roasted peeled cashew nuts sold in markets within Keffi, Nasarawa State, thereby creating an important regional data gap.

This study was therefore undertaken to evaluate the bacteriological profile of roasted peeled cashew nuts purchased from selected markets in Keffi, Nasarawa State. The study specifically aimed to determine the bacterial load of roasted peeled cashew nuts and to isolate, identify, and characterize the bacterial contaminants associated with the samples.

## Materials and methods

### Study Area

This study was conducted at the Microbiology Laboratory of Bingham University using roasted peeled cashew nut samples obtained from markets within Keffi. Keffi is located in Nasarawa State in the North-Central region of Nigeria and lies approximately 53 km from the Abuja and about 137 km from Lafia, the state capital. Geographically, Keffi is situated between latitude 8°50'N and longitude 7°52'E (Akwa et al., 2007). The area is characterized by intense commercial activities and a high level of human interaction, factors that may contribute to microbial contamination of ready-to-eat food products sold within the metropolis.

### Sample Collection and Preparation

A total of fifty wrapped samples of roasted peeled cashew nuts were purchased from five major markets within Keffi metropolis, namely Sambisa Market, Tsohon Market, Angwan Lambu Market, Neighbourhood Market, and Kara Market. Ten samples were collected from each market using aseptic procedures to minimize external contamination during handling and transportation. Immediately after collection, the samples were transported to the Microbiology Laboratory of Bingham University for microbiological analysis. For sample preparation, 20 g of each roasted peeled cashew nut sample were aseptically crushed using a sterile porcelain mortar and pestle. Subsequently, 1 g of the homogenized sample was transferred into a test tube containing 10 ml of sterile peptone water and thoroughly mixed to obtain a homogeneous suspension. The resulting mixture served as the stock solution for serial dilution procedures.

Ten-fold serial dilution of each stock solution was carried out using sterile normal saline. Ten sterile test tubes containing 9 ml of normal saline were arranged in a test tube rack. One milliliter of the stock suspension was aseptically transferred into the first tube to obtain a dilution factor of  $10^{-1}$  and mixed thoroughly. Thereafter, 1 ml from the first dilution was transferred into the second tube to obtain a dilution factor of  $10^{-2}$ . This procedure was repeated sequentially until a final dilution of  $10^{-5}$  was achieved. Microbial load determination of the roasted peeled cashew nut samples was carried out using the spread plate technique. Aliquots of 0.1 ml from the  $10^{-5}$  dilution were inoculated onto sterile culture media, including Nutrient Agar for total viable bacterial count, MacConkey Agar for total coliform count, Eosin Methylene Blue Agar for faecal coliform count, and Mannitol Salt Agar for the isolation of *Staphylococcus aureus*. The inoculum was evenly spread across the surface of the agar plates using a sterile spreader. The inoculated plates were incubated at 37°C for 24–48 h. Following incubation, discrete colonies that developed on the culture media were counted using a colony counter (BCLC-101), and the results were expressed as colony-forming units per milliliter (CFU/ml). All analysis were carried out in triplicate as described by Ugwu (2024). Representative colonies were repeatedly subcultured on freshly prepared Nutrient Agar plates to obtain pure cultures for further characterization and identification.

### Isolation and Identification of Bacterial Isolates

Distinct bacterial colonies obtained from the culture media were aseptically picked using a sterile wire loop and streaked onto sterile Nutrient Agar plates for purification. The plates were incubated at 37°C for 24 h. Pure isolates obtained after incubation were characterized based on their cultural, morphological, and biochemical properties. Representative bacterial isolates were selected and identified according to the methods described by Adeoye and Lateef (2021).

Gram staining was performed to differentiate bacterial isolates into Gram-positive and Gram-negative groups following the method of Makut et al. (2022). A thin smear of each bacterial isolate was prepared on a clean grease-free glass slide, air-dried, and heat-fixed by passing the slide through a flame several times. The smear was flooded with crystal violet stain

for 60 s and rinsed gently with running water. Iodine solution was then added as a mordant for another 60 s before rinsing with water. The smear was subsequently decolorized with 95% acetone and immediately rinsed with clean water. Counterstaining was performed using safranin for 30 s, after which the slide was rinsed and air-dried. The stained slides were examined microscopically under  $\times 100$  oil immersion objective to determine the shape, arrangement, and Gram reaction of the isolates. Gram-positive bacteria appeared purple or violet, whereas Gram-negative bacteria appeared pink or red (Makinde et al., 2023).

**Biochemical Characterization of Isolates**

Biochemical tests were conducted to identify bacterial isolates based on their metabolic activities and enzymatic reactions. The tests performed included catalase, indole, oxidase, coagulase, citrate utilization, sugar fermentation, and starch hydrolysis tests. The catalase test was carried out by placing a drop of hydrogen peroxide on a clean slide and emulsifying a portion of the bacterial colony into the reagent. Immediate bubble formation indicated a positive reaction (Ugwu, 2024). For the indole test, bacterial isolates were inoculated into freshly prepared peptone water and incubated at 37°C for 24 h. After incubation, a few drops of Kovac’s reagent were added. Formation of a red ring at the surface of the medium indicated a positive result, while absence of colour change indicated a negative result (Ugwu, 2024).

The oxidase test was performed using freshly prepared oxidase reagent (1% phenylenediamine). A portion of the bacterial isolate was smeared onto filter paper impregnated with the reagent. Development of a deep purple colour within 5–30 s indicated a positive reaction. For the coagulase test, bacterial colonies were emulsified in physiological saline on a clean slide, after which citrated human plasma was added and mixed gently. Visible clumping of cells within one minute was recorded as a positive result. Citrate utilization ability was determined using Simmons citrate agar. Test organisms were inoculated onto the agar slants and incubated at 37°C for 24 h. A colour change of the medium from green to blue indicated citrate utilization (Ugwu, 2024). Sugar fermentation tests were conducted using phenol red broth supplemented with 1% glucose or lactose. Durham tubes were inserted into the broth medium to detect gas production. Following inoculation and incubation at 37°C, a change in colour from red to yellow, with or without gas bubbles in the Durham tubes, indicated positive fermentation reactions (Makinde et al., 2023). Starch hydrolysis was determined by streaking bacterial isolates onto starch agar plates followed by incubation at 35°C for 48 h. After incubation, iodine solution was added to the plates. The appearance of a clear zone around bacterial growth indicated starch hydrolysis, whereas a

blue-black coloration signified the absence of starch degradation (Makinde et al., 2023).

**Determination of Bacterial Occurrence**

The occurrence of bacterial contaminants in roasted peeled cashew nuts sold within Keffi metropolis was determined based on the frequency of bacterial isolates recovered from the samples collected across the five selected markets. The prevalence of bacterial contamination was assessed by evaluating the presence and distribution of bacterial species isolated from the roasted peeled cashew nut samples.

**Results and Discussion**

**Bioloal of Peeled Roasted Cashew Nuts**

The microbiological analysis of roasted peeled cashew nuts sold in selected markets within Keffi revealed varying degrees of bacterial contamination across all sampled locations. Sambisa Market recorded the highest microbial load, with total viable count (TVC), total coliform count (TCC), total faecal coliform count (TFCC), and *Staphylococcus aureus* count of  $8.3 \times 10^5$  cfu/ml,  $6.1 \times 10^5$  cfu/ml,  $3.7 \times 10^5$  cfu/ml, and  $2.2 \times 10^5$  cfu/ml, respectively. This was followed by Angwan Lambu Market, which showed TVC of  $5.3 \times 10^5$  cfu/ml, TCC of  $4.1 \times 10^5$  cfu/ml, TFCC of  $2.4 \times 10^5$  cfu/ml, and *S. aureus* count of  $1.3 \times 10^5$  cfu/ml. Tsohon Market, Neighbourhood Market, and Kara Market also exhibited considerable bacterial loads, although comparatively lower than those recorded in Sambisa Market (Table 1). The high microbial counts observed across the markets suggest substantial microbial contamination of the roasted peeled cashew nuts, possibly resulting from poor sanitary conditions, improper handling, exposure to contaminated environments, and inadequate storage practices after roasting (Kumari et al., 2025). The elevated total viable and coliform counts obtained in this study are indicative of unsanitary handling and environmental contamination during processing and marketing. Coliform organisms are commonly used as indicators of hygiene quality in food products, while faecal coliforms specifically indicate possible contamination from faecal sources (Martin et al., 2016). The presence of faecal coliforms in the roasted cashew nut samples therefore raises serious food safety concerns, especially because roasted cashew nuts are consumed as ready-to-eat snacks without further heat treatment. Similar findings were reported by Omorodion and Oyedele (2024), who documented high microbial contamination in processed cashew nuts and groundnuts sold in Nigeria, including the occurrence of *Bacillus*, *Klebsiella*, and *Staphylococcus* species associated with poor postharvest handling and unhygienic storage conditions (Lawal & Fagbohun, 2014; Adetunji et al., 2018)

**Table 1: Bioloal of roasted pilled cashew nuts purchased in some selected markets in Keffi ( $10^5$ )**

Sample	Total viable	Total coliform	Total fecal coliform	<i>Staphylococcus aureus</i>
Location	count (cfu/ml)	count (cfu/ml)	count (cfu/ml)	count (cfu/ml)

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Sambisa market	8.3	6.1	3.7	2.2
Angwan Lambu	4.1	3.0	2.2	1.6
Tsohon market	3.8	2.6	2.0	1.1
Neighbourhood market	5.3	4.1	2.4	1.3
Kara market	4.0	2.8	1.4	1.0

The bacterial isolates identified in this study included *Pseudomonas* spp., *Klebsiella* spp., *Bacillus* spp., *Escherichia coli*, *Enterobacter* spp., and *Staphylococcus aureus* (Table 2). The occurrence of *Escherichia coli* is particularly significant because it is widely recognized as an indicator organism for faecal contamination (Nowicki et al., 2021). Its presence suggests contamination from handlers, contaminated water, utensils, or market environments. Likewise, the detection of *Staphylococcus aureus* indicates contamination through direct human handling since the organism is commonly associated with the skin, nose, and hands of food vendors (Kadariya et al, 2014). The isolation of *Bacillus* species may also be attributed to environmental exposure because these organisms are spore-forming bacteria capable of surviving harsh environmental conditions and thermal processing (Bahaddad et al., 2023).

The findings of this study agree with recent reports demonstrating that nuts and plant-based food products may harbour potentially pathogenic microorganisms despite undergoing thermal processing. Kyrylenko et al. (2023) and Correia et al. (2026) reported the occurrence of microbial contaminants, including *Bacillus* species and other spoilage organisms, in plant-based ingredients used for dairy alternatives, including cashew-derived products. Similarly, Louvau and Harris (2023) detected *Salmonella*, coliforms, and *Escherichia coli* in naturally contaminated cashews associated with foodborne outbreaks, emphasizing the persistence of microbial contaminants in nut products. These studies support the present findings and demonstrate that roasted or processed nuts can still become contaminated during post-processing handling, packaging, transportation, and retail display.

**Table 2: Cultural, Morphological and Biochemical Characterization of Bacterial isolates from Roasted pilled cashew nuts in Keffi markets**

Cultural characteristics	Morphological characteristics		Biochemical characteristics						Sugar utilization				Inference	
	Gram Stain	Shape	CA	IN	OX	Coa	CIT	H <sub>2</sub> S	Ma l	Gl u	La c	Suc		
Smooth flat colonies and greenish on NA	-	Rod	-	+	-	-	-	-	-	-	-	-	-	<i>Pseudomonas</i> sp
Bulk colonies and Milkish on NA	+	Rod	+	+	+	-	+	-	+	+	+	+	+	<i>Bacillus</i> sp
pinkish (MCA)	-	Rod	+	-	-	+	+	+	+	+	-	-	-	<i>Klebsiella</i> sp
Greenish metallic sheen on EMB and Pink on MCA	-	Rod	+	+	-	+	+	-	+	+	+	-	-	<i>Escherichia coli</i>
Purple on EMB and light pink (MCA)	-	Cocci	-	-	-	-	+	-	+	+	+	+	+	<i>Enterobacter</i> sp
Colonies with golden yellow on MSA	-	Rod	+	-	-	+	+	+	+	+	-	+	+	<i>Staph aureus</i>

Note: - = Negative; + = positive; CA = Catalase; CT = Citrate; VP = Voges-Proskauer; In = Indole; Nit = Nitrate; Man = Mannose; Mal = Maltose; Glu = Glucose; Lac = Lactose; Suc = Sucrose; MSA = De Man, Rogosa and Sharpe Agar; NA = Nutrient Agar; MCA = MacConkey Agar, MSA= Mannitol salt agar.

The public health implication of these findings is considerable because consumption of contaminated ready-to-eat cashew

nuts may predispose consumers to foodborne illnesses such as gastroenteritis, diarrhoea, vomiting, and staphylococcal food poisoning. The high microbial load observed also suggests poor compliance with food hygiene standards among vendors within the sampled markets. Therefore, there is a need for improved sanitary practices during processing, handling, packaging, and storage of roasted cashew nuts. Regular monitoring of microbial quality by relevant food safety agencies, coupled with public health education for food

vendors, is essential to minimize contamination risks and ensure consumer safety. In addition, proper packaging and hygienic retail practices should be encouraged to reduce exposure of roasted cashew nuts to environmental contaminants during marketing and distribution.

## Conclusion

The present study revealed significant bacterial contamination in roasted peeled cashew nuts sold in selected markets within Keffi. High microbial loads, including total viable bacteria, coliforms, faecal coliforms, and *Staphylococcus aureus*, were detected across all sampled markets, indicating poor sanitary practices during handling, storage, and retailing. The isolation of potentially pathogenic organisms such as *Escherichia coli*, *Klebsiella* spp., *Bacillus* spp., and *Staphylococcus aureus* further highlights the potential health risks associated with the consumption of contaminated ready-to-eat cashew nuts. These findings emphasize the need for improved food hygiene practices, proper packaging, routine microbial quality assessment, and effective regulatory monitoring to minimize contamination and enhance the microbiological safety of roasted peeled cashew nuts sold in local markets.

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