

EVALUATION OF THE MICROBIOLOGICAL QUALITY OF INDIGENOUS COMPLEMENTARY FOODS IN KANO STATE

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ABSTRACT

Locally (Indigenous) prepared complementary foods are widely consumed by infants in Nigeria because of their affordability and accessibility; however, their microbiological safety remains a major public health concern. This study evaluated the microbiological quality of three locally prepared complementary food samples (Samples A, B, and C) sold in Tarauni Local Government Area, Kano State, in comparison with a laboratory-prepared counterpart (Sample D). The market samples were unbranded, powdered formula, and sold under open conditions, potentially exposing them to contamination. Standard microbiological methods were employed: Nutrient Agar for aerobic mesophilic bacterial counts, Potato Dextrose Agar for fungal isolation, and the Most Probable Number (MPN) method for coliform estimation, while pathogens were identified through biochemical tests. The aerobic mesophilic bacterial counts of the locally prepared foods ranged from 2.45×10^6 to 1.08×10^7 CFU/mL, exceeding the WHO permissible limit of 1.0×10^3 CFU/mL. Fungal contaminants detected included *Aspergillus niger*, *A. fumigatus*, and *A. flavus*. Coliform counts reached up to 150 MPN/100 mL, and pathogens such as *Escherichia coli*, *Staphylococcus aureus*, and *Shigella* spp were present in the market samples but absent in the laboratory-prepared food, which remained within safety limits. These findings reveal significant microbiological risks associated with locally prepared complementary foods. Strengthening vendor hygiene practices, enforcing regulatory surveillance, and promoting laboratory-standard local formulations are recommended to safeguard infant health.

Keywords: Indigenous complementary foods, Food safety, Microbiological quality, Pathogenic microorganism

INTRODUCTION

Complementary feeding is the introduction of solid and semi-solid foods alongside breast milk and represents a critical phase in the growth and development of infants and young children. In Nigeria, many mothers rely on locally prepared complementary foods such as pap (ogi), millet-based porridges, and cereal–legume blends due to their affordability and cultural acceptance (Omemu and Aderoju, 2020). However, these foods are often produced, handled, and sold under unhygienic conditions, predisposing them to microbial contamination and increasing the risk of foodborne illnesses among infants, whose immune systems are still developing (Igbabul *et al.*, 2022).

Several studies conducted in Nigeria have reported contamination of locally prepared complementary foods with *Escherichia coli*, *Staphylococcus aureus*, *Shigella* spp, and toxigenic fungi such as *Aspergillus* spp, indicating serious food safety concerns (Onweluzo and Nwabugwu, 2009; Akindele *et al.*, 2023; Ezekiel *et al.*, 2019). While these findings highlight the general risks, there remains limited data specific to Kano State, despite its high population density and reliance on open-market complementary foods.

Despite the widespread consumption of locally prepared complementary foods in Nigeria, limited attention has been given to their microbiological safety compared to those produced under hygienic laboratory or industrial conditions. Complementary foods prepared in such controlled environments generally have lower microbial loads due to strict adherence to sanitary standards and processing regulations (Cheesbrough, 2010; WHO/FAO, 2019). Assessing and comparing the microbial quality of market-sold and laboratory-prepared complementary foods is essential to identify potential public health risks and justify the need for improved hygiene practices during local food production and handling.

Therefore, this study was designed to assess the microbiological quality of locally prepared complementary foods sold in Tarauni Local Government Area, Kano State, Nigeria, and to compare them with a laboratory-prepared complementary food formulated from local ingredients under hygienic conditions.

MATERIALS AND METHODS

This study was conducted in Tarauni Local Government Area, Kano State, Nigeria, where locally prepared complementary foods are commonly consumed by infants and young children. The area is one of the densely populated urban centers in Kano metropolis and is characterized by active open markets where food vendors produce and sell various cereal–legume complementary food mixtures.

Sample Collection

A total of four complementary food samples were analyzed, comprising three locally prepared complementary foods purchased from different vendors in the Tarauni markets (coded as Samples A, B, and C) and one laboratory-prepared complementary food (Sample D). The market samples were selected purposively to represent the most commonly consumed complementary foods in the area, ensuring diversity in ingredients and preparation methods.

Sample collection was carried out between February and March 2025, during the dry season when food vending activities are high. Each food type (Samples A–C) was collected in triplicate from different vendors to ensure representativeness and to account for potential variation in handling and preparation. The laboratory-prepared sample was produced under aseptic conditions to serve as a control for comparison.

Although the total number of samples was limited to four, the study design allowed for a focused exploratory assessment, consistent with methodologies adopted in similar

microbiological studies on complementary foods (Adeboye *et al.*, 2024; Awogbenja and Ndife, 2018).

Sample Description

Sample A: A cereal-legume mixture consisting of tiger nut, dates, crayfish, groundnut, soybeans, beans, and millet, obtained from a local vendor in Tarauni market.

Sample B: A blend of wheat, rice, and groundnut, purchased from another vendor in Tarauni market.

Sample C: A mixture of maize, millet, sorghum, soybeans, and groundnut, purchased from a different vendor in Tarauni market.

Sample D (Laboratory-prepared) Formulated using the same composition as Sample B (wheat, rice, and groundnut) but processed under hygienic laboratory conditions. The grains were cleaned, roasted, milled, blended, and packaged aseptically to serve as a hygienically prepared control sample. All the samples obtained from the market (A, B, and C) were in dry powdered form, as they were semi-processed complementary food products meant for further reconstitution with water before feeding. They were collected in the morning hours while displayed for sale under ambient temperature conditions (28–32°C). The samples had been prepared less than 24 hours prior to collection, according to information obtained from the vendors. Each sample was collected in sterile, airtight containers to prevent post-purchase contamination and was transported immediately to the laboratory for microbiological analysis, following standard procedures described by Cheesbrough (2010) and Igbabul *et al.* (2022).

Sample Preparation

Each sample was purchased in sterile, airtight containers. The laboratory-prepared sample (Sample D) was formulated in the microbiology laboratory using the same composition as Sample B (wheat, rice, and groundnut). Ingredients were cleaned, roasted, milled, and aseptically packaged.

For each analysis, triplicate determinations were performed per sample. Ten grams (10 g) of each food sample was homogenized in 90 mL of sterile peptone water to prepare the 10^{-1} dilution, followed by serial dilutions up to 10^{-6} (Fawole and Oso, 2007; Cheesbrough, 2010).

Microbiological Analysis

Aerobic Mesophilic Bacterial Count

The aerobic mesophilic bacterial count of each sample was determined using the pour plate method on Nutrient Agar (NA) as described by Harrigan and McCance (2014) and Cheesbrough (2010). One gram (1 g) of each powdered sample was aseptically homogenized in 9 mL of sterile distilled water to obtain a 10^{-1} dilution, followed by serial tenfold dilutions up to 10^{-6} . From dilutions 10^{-5} and 10^{-6} , 1 mL aliquots were transferred into sterile Petri dishes and overlaid with molten Nutrient Agar cooled to about 45°C. The plates were swirled gently to mix and allowed to solidify. Incubation was carried out at 37°C for 24 hours, which supports the growth of mesophilic bacteria. Plates containing 30–300 colonies were selected and counted using a digital colony counter, and the results were expressed as colony-forming units per milliliter (CFU/g).

Coliform Count (Most Probable Number Method)

Coliform enumeration was carried out using the standard multiple-tube fermentation (MPN) technique as described by Cheesbrough (2010) and Fawole and Oso (2007). Briefly, one gram (1 g) of each powdered sample was aseptically

homogenized in 9 mL of sterile peptone water to obtain the 10^{-1} stock suspension; serial tenfold dilutions were prepared up to 10^{-6} . The MPN test was performed using a three-tube series for each dilution: three tubes containing 10 mL of double-strength lactose broth were inoculated with 10 mL of the appropriate (undiluted) sample suspension for the 10 mL test portion; three tubes containing 10 mL of single-strength lactose broth were inoculated with 1.0 mL aliquots for the 1 mL test portion; and three tubes containing 10 mL of single-strength lactose broth were inoculated with 0.1 mL aliquots for the 0.1 mL test portion. Durham fermentation tubes were used to detect gas production. All inoculated tubes were incubated at 37°C for 24 hours. Tubes showing gas production were scored as positive in the presumptive test.

Presumptive positive tubes were subcultured onto Eosin Methylene Blue (EMB) agar and MacConkey agar for confirmation; characteristic colonies (metallic green sheen on EMB or lactose-fermenting pink colonies on MacConkey) were considered presumptive *Escherichia coli*. Confirmatory identification of *E. coli* was performed by standard biochemical tests (IMViC: indole positive, methyl red positive, Voges–Proskauer negative, citrate negative) as described by Cheesbrough (2010). Positive and negative quality controls (known coliform strain and sterile peptone water, respectively) were included in each run.

The number of positive tubes at each dilution was recorded and the corresponding MPN value was determined from standard MPN tables; results were expressed as MPN per 100 mL (MPN/100 mL) of reconstituted sample equivalent. All assays were performed in triplicate for each sample and the mean MPN value was reported. Procedures, media preparation, and incubation conditions followed Cheesbrough (2010) and Fawole and Oso (2007) to ensure reproducibility.

Isolation and Identification of Pathogens

Isolation of bacterial pathogens was carried out using standard microbiological procedures as described by Cheesbrough (2010) and Fawole and Oso (2007). After incubation of the Nutrient Agar plates used for total viable count, distinct colonies differing in morphology (shape, color, elevation, and edge) were aseptically subcultured by streak plate technique onto selective and differential media, including MacConkey Agar, Mannitol Salt Agar (MSA), and Salmonella–Shigella Agar (SSA), to obtain pure cultures. The plates were incubated at 37°C for 24 hours and examined for characteristic colony appearance.

Typical colonies suggestive of enteric or pathogenic bacteria were further purified by repeated streaking on fresh nutrient agar plates to ensure purity. Pure isolates were then transferred onto nutrient agar slants and stored at 4°C for subsequent biochemical characterization.

Identification of bacterial isolates was based on colony morphology, Gram staining, and a series of biochemical tests, including catalase, coagulase, indole, methyl red, Voges–Proskauer, citrate utilization, and urease tests, following the schemes of Cheesbrough (2010) and Fawole and Oso (2007). The results obtained were compared with standard biochemical profiles for the identification of bacteria such as *Escherichia coli*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Shigella spp.*

Fungal Isolation and Identification

Fungal enumeration, isolation and identification were carried out using the pour plate method on Potato Dextrose Agar (PDA), following the procedures of Pitt and Hocking (2009) and Cheesbrough (2010). One gram (1 g) of each powdered sample was aseptically homogenized in 9 mL of sterile

distilled water to obtain a 10^{-1} dilution, followed by serial tenfold dilutions up to 10^{-6} . From dilutions 10^{-5} and 10^{-6} , 1 mL aliquots were inoculated into sterile Petri dishes and overlaid with molten PDA cooled to approximately 45°C . To inhibit bacterial growth, the medium was supplemented with 0.01 g/L of chloramphenicol before pouring. The plates were gently swirled, allowed to solidify, and incubated inverted at $25 \pm 2^{\circ}\text{C}$ for 5 days, conditions optimal for fungal growth.

After incubation, plates showing discrete colonies (30–300 CFU) were selected, and distinct colonies were aseptically subcultured onto fresh PDA plates to obtain pure fungal cultures. Pure isolates were maintained on PDA slants at 4°C for further identification. Identification of fungi was based on macroscopic features (color, texture, and surface pigmentation) and microscopic characteristics observed after staining with lactophenol cotton blue (LPCB). The observed morphological structures were compared with standard taxonomic descriptions for fungal identification (Pitt and Hocking, 2009).

Data Analysis

Microbial counts obtained from each sample were expressed as mean colony-forming units per gram (CFU/g) for all samples. Prior to microbial enumeration, one gram (1 g) of each powdered complementary food sample was aseptically homogenized in 9 mL of sterile distilled water to create a 1:10 suspension (10^{-1} dilution), from which further serial dilutions were made for analysis.

All data were recorded in triplicates, and results were presented as *mean* $\pm$ standard deviation (SD). Statistical analysis was performed using SPSS version 25.0 (IBM Corp., USA). Differences in microbial load among the samples were evaluated using one-way analysis of variance (ANOVA), and

mean separations were considered significant at $p < 0.05$. The microbiological quality of the complementary foods was interpreted in comparison with the WHO (2008) recommended microbiological safety limits for complementary foods.

RESULTS AND DISCUSSION

Aerobic Mesophilic Bacterial Count

The aerobic mesophilic bacterial counts of the locally prepared complementary foods were presented in Table 1. Sample B recorded the highest bacterial load of 1.08×10^7 CFU/g, followed by Sample A (7.0×10^6 CFU/g), while Sample C had the lowest count (5.3×10^6 CFU/g). The laboratory-prepared sample (Sample D) exhibited a markedly lower count of 1.0×10^3 CFU/g, which falls within the WHO (2008) permissible limit for complementary foods. Statistical analysis revealed no significant difference ($p = 0.359$) among the locally prepared samples (A–C), indicating that bacterial load was not significantly influenced by sample type.

Aerobic Mesophilic Fungal Count

The aerobic mesophilic fungal counts of the locally prepared complementary foods are presented in Table 1. Sample B exhibited the highest fungal count (1.11×10^7 CFU/g), followed by Sample C (3.1×10^6 CFU/g), while Sample A recorded the lowest fungal count among the locally prepared foods (2.45×10^6 CFU/g). The laboratory-prepared sample (Sample D) showed the least contamination (1.0×10^3 CFU/g), which was within the WHO (2008) acceptable microbiological limit for complementary foods. Statistical analysis revealed no significant difference ($p = 0.394$) among the locally prepared samples (A–C), indicating that fungal load was not significantly affected by sample type.

Table 1: Mean Aerobic Mesophilic Bacterial and Fungal Counts (CFU/g) of Locally Prepared and Laboratory-Prepared Complementary Foods

Samples	Bacterial loads (CFU/g)	Fungal loads (CFU/g)
A	7.0×10^6	2.45×10^6
B	1.08×10^7	1.11×10^7
C	5.3×10^6	3.1×10^6
D	1.0×10^3	1.0×10^3

NOTE: Values represent the mean of triplicate determinations ($n = 3$) and are expressed as colony-forming units per gram (CFU/g). WHO (2008) recommends $\leq 1.0 \times 10^3$ CFU/g as the acceptable limit for complementary foods

Coliform Count

The coliform counts of the complementary food samples are presented in Table 2. Sample B recorded the highest coliform level (150 MPN/100 mL), followed by Sample C (20

MPN/100 mL). Sample A showed a count of 3 MPN/100 mL, while the laboratory-prepared sample (Sample D) had the lowest level (<3 MPN/100 mL).

Table 2: Mean Coliform Counts of Locally Prepared and Laboratory Prepared Complementary Food in MPN/100mL

Samples	MPN/100mL
A	3
B	150
C	20
D	<3

Key: MPN = Most Probable Number

Note: Values represent the mean of triplicate determinations ($n = 3$)

Pathogen Identification

The pathogenic bacteria isolated from each complementary food sample were presented in Table 3. *Escherichia coli*, *Staphylococcus aureus*, and *Staphylococcus epidermidis* were detected in all locally prepared samples (A–C), while *Shigella spp.* was identified only in Sample B. No pathogenic bacteria were detected in the laboratory-prepared sample (Sample D).

Fungal species isolated from the complementary food samples included *Aspergillus niger*, *A. fumigatus*, and *A. flavus*. Sample A contained *A. niger* and *A. flavus*; Sample B had *A. niger* only; while Sample C harbored *A. fumigatus* and *A. flavus*. The laboratory-prepared sample (Sample D) showed no detectable fungal growth.

Table 3: Occurrence of Pathogenic Bacteria and Fungi in Locally Prepared and Laboratory-Prepared Complementary Foods

Sample	<i>E. coli</i>	<i>S. aureus</i>	<i>S. epidermis</i>	<i>Shigella spp.</i>	<i>A. niger</i>	<i>A. fumigatus</i>	<i>A. flavus</i>
A	+	+	+	-	+	-	+
B	+	+	+	+	+	-	-
C	+	+	+	-	-	+	+
D	-	-	-	-	-	-	-

Note: (+) = Present; (-) = Absent. Results represent the outcome of triplicate analyses for each food type ($n = 3$), and the values reflect the predominant microorganisms detected

Discussion

The findings of this study reveal a clear microbiological safety gap between locally produced complementary foods and their laboratory-prepared counterpart. Samples A, B, and C exhibited aerobic mesophilic bacterial counts far exceeding the World Health Organization (WHO, 2008) permissible limit of 1.0×10^3 CFU/g, which aligns with earlier findings reported from Nigerian markets (Onuoha *et al.*, 2022). The consistently high bacterial loads observed across the locally produced samples indicate that contamination is widespread, reflecting generally poor hygienic practices during food processing and handling rather than isolated vendor-specific lapses.

Fungal contaminants, including *Aspergillus niger*, *A. fumigatus*, and *A. flavus*, were detected in all locally produced complementary foods, raising concerns about possible aflatoxin contamination. *A. flavus* in particular is known to produce aflatoxins, potent mycotoxins associated with growth retardation, hepatotoxicity, and immunosuppression in infants (Ezekiel *et al.*, 2019). The detection of these fungi in foods commonly consumed by infants underscores the need for improved post-harvest drying, moisture regulation, and storage practices in local markets to minimize fungal proliferation and toxin production.

The detection of coliforms and enteric pathogens, including *Escherichia coli* and *Shigella spp.*, indicates fecal contamination risks that may have originated from the use of contaminated water or poor handling hygiene during processing and sale. Since the samples were obtained in dry powdered form and displayed openly at ambient temperatures in local markets, contamination could have occurred through dust, unclean utensils, or handling by multiple vendors. These bacteria are strongly associated with infant diarrheal diseases, a major cause of morbidity and mortality in low-income settings (Ifeanyi *et al.*, 2017). Similarly, *Staphylococcus aureus* and *S. epidermidis*, both human commensals, suggest contamination through human contact during production and retailing, consistent with findings by Ameh *et al.* (2020).

By contrast, the laboratory-prepared sample consistently met safety standards across all microbial parameters. This reinforces the importance of hygienic processing, controlled environments, and adherence to good manufacturing practices (FAO/WHO, 2003). It also highlights the feasibility of producing safer complementary foods locally, provided proper handling and regulatory oversight are enforced.

The public health implications of these findings are noteworthy, particularly for infants and young children who are highly susceptible to foodborne infections. However, since the locally produced complementary foods analyzed in this study were dry, semi-processed powders intended for reconstitution with hot water before feeding, the immediate risk of infection may be somewhat reduced, as heating can inactivate many vegetative microbial cells. Nonetheless, the detection of enteric pathogens and toxigenic fungi in these products highlights potential hazards arising from poor hygiene and storage conditions.

CONCLUSION

The microbiological assessment of traditionally prepared complementary foods sold in Tarauni Local Government Area revealed unacceptably high levels of aerobic mesophilic bacteria, fungi, coliforms, and pathogenic organisms, all exceeding the recommended safety limits for infant complementary foods as outlined by the World Health Organization (WHO, 2008) and Codex Alimentarius (2007). These findings are consistent with previous reports on microbial contamination of weaning foods in Northern Nigeria (Aliyu and Hassan, 2021), indicating that market-sold complementary foods, although affordable and culturally accepted, may pose potential risks of foodborne illnesses to infants and young children.

In contrast, the laboratory-prepared complementary food sample produced under hygienic laboratory conditions demonstrated microbial loads within acceptable standards, emphasizing the importance of good manufacturing and handling practices in ensuring product safety.

This study was limited by the small number of samples analyzed and the restriction to one geographical area, which may not fully represent all traditionally prepared complementary foods across Kano State. In addition, the microbial analyses did not assess spore-forming bacteria or quantify aflatoxin levels, which could further clarify the safety status of these foods.

Despite these limitations, the findings provided valuable baseline data to inform local interventions aimed at improving food safety practices, promoting vendor hygiene training, and supporting the production of affordable, nutritionally balanced, and microbiologically safe complementary foods for infants in Nigeria.

Based on these findings, the following are the recommendations:

- Policy Enforcement:** Strengthening regulatory monitoring and mandatory microbiological testing of complementary foods before market distribution.
- Capacity Building:** Implementing food safety training programs for local vendors and caregivers, focusing on hygiene, safe storage, and proper handling practices.
- Sustainable Alternatives:** Promoting locally formulated laboratory-standard complementary foods, which can combine cultural acceptability with microbiological safety, thereby reducing dependence on imported commercial formulas.

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