



Microbial Quality Assessment of Some Fruits Obtained from Retail Outlets in Zaria, Kaduna State, Nigeria

Wata, Innocent John^{1*}, Deborah, Ajibade Oyenike² & Umaru, Abdulmalik³

^{1,3} Department of Microbiology, Faculty of Life Sciences, Federal University Dutsin-Ma

²Ifedapo Federal Housing Estates, Ikirun Osun State.

Corresponding Author Email: ^{1}watainnocent@gmail.com ; ORCID: ^{1*}<https://orcid.org/0009-0002-5640-2343>

²deborahajibade5@gmail.com ; ²<https://orcid.org/0009-0009-0516-7073>

³umaruabdulmalik449@gmail.com; ³<https://orcid.org/0000-0001-5153-7201>



ABSTRACT

Although fruits are an essential component of a balanced diet, providing vital nutrients for human health, their high moisture content and frequent exposure to environmental contaminants make them susceptible to microbial contamination, posing serious public health risks. This research work was aimed to assess the microbial quality of selected fruits (pawpaw, watermelon, and pineapple) obtained from retail outlets in Zaria, Kaduna State, Nigeria. Cross-sectional study was conducted in three major retail locations: Samaru, Kwangila, and PZ. A total of 27 fruit samples were randomly purchased and analyzed using standard microbiological techniques to determine the total bacterial and fungal loads. Bacteria isolates were identified through gram staining, biochemical test and morphological characterization, while fungi isolates were identified through morphological characteristics and microscopic view using lactophenol cotton blue stain. The bacteria species identified were gram positive and gram negative to include *Staphylococcus aureus*, *Escherichia coli*, *Bacillus* species and *Pseudomonas*. Antimicrobial susceptibility testing showed that the isolates were highly susceptible and resistant to some antibiotics. Pathogenicity tests were also conducted to assess the spoilage potential of fungal isolates. The results revealed high microbial contamination across all fruit samples, with pawpaw and watermelon exhibiting the highest bacterial and fungal loads. Fungal isolates included *Aspergillus niger*, *Aspergillus flavus*, *Penicillium* spp., *Candida* spp., *Mucor* spp., and *Rhizopus* spp., with *Aspergillus* being the most prevalent. These findings emphasize the need for improved hygiene measures in fruit handling, storage, and retail distribution. Further research is recommended to explore long-term trends in antimicrobial resistance among fruit-associated pathogens.

Keywords:

Microbial contamination, food safety, antimicrobial resistance, fruit spoilage, *Escherichia coli*, *Aspergillus* spp., Zaria, Nigeria.

INTRODUCTION

Fruits are a vital part of the human diet consumed among diverse group. It provides the body with essential nutrients due to its rich array of vitamins, minerals, fibre and antioxidant (Slavin and Lloyd, 2012), which contribute to health and well-being. Regular consumption of fruits has been linked to a reduced risk of chronic diseases such as cardiovascular diseases, type 2 diabetes, and certain types of cancer due to their high antioxidant content (Aune *et al.*, 2017). In addition to their nutritional benefits, fruits are versatile and can be consumed fresh, dried, or processed into juices, jams, and other products (FAO, 2013). Despite these importance, fruits' consumption varies among populace due to several

factors. Research conducted among households in Zaria reveals that income plays a critical role in determining fruit consumption patterns. Many low-income families prioritize staple foods over fruits due to financial constraints, making fruits a less frequent component of their diet (Abdulrahman *et al.*, 2018). Additionally, the seasonal nature of fruit availability significantly affects consumption rates. For instance, fruits like mangoes and guavas are widely consumed during their peak seasons but are scarce and costly at other times (Yakubu *et al.*, 2019). Awareness of the nutritional and health benefits of fruits also varies among the population. A study found that only 28.8% of respondents in Zaria demonstrated a good

understanding of the importance of fruits in preventing chronic diseases such as hypertension and diabetes, while 17% lacked awareness altogether (Olatunji *et al.*, 2021). This knowledge gap further contributes to the low prioritization of fruits in daily diets. Efforts to increase fruit consumption in Zaria should focus on improving affordability, enhancing public awareness campaigns about the health benefits of fruits, and developing strategies to reduce post-harvest losses and stabilize fruit prices throughout the year (Akinyele *et al.*, 2016).

However, despite the benefits derived from fruit consumption, their high water activity and nutrient content make them susceptible to microbial contamination, posing significant public health risks if consumed without proper handling or washing (Ofor *et al.*, 2009). The microbial contamination of fruits can occur at different stages of production, including cultivation, harvesting, transportation, storage, and retailing (Barro *et al.*, 2006). Factors such as the use of untreated water for irrigation, poor handling practices, and inadequate sanitation in retail outlets contribute to the microbial load on fruits (Omemu and Bankole, 2008; Saba and Oranusi, 2020). More so, studies have shown that poor sanitation, lack of refrigeration, inefficient supply chains and handling practices significantly contribute to microbial contamination of fruits in developing regions (Ezekiel *et al.*, 2022).

In Nigeria, fruits sold in retail outlets are often exposed to unhygienic conditions, especially in open markets and by street vendors. These environments create favourable conditions for microbial proliferation, including pathogenic organisms such as *Escherichia coli*, *Salmonella spp.*, and *Listeria monocytogenes* (Nwachukwu *et al.*, 2021). Among bacteria, *E. coli*, *Salmonella spp.*, *Listeria monocytogenes*, and *Staphylococcus aureus* are frequently isolated from fresh fruits, originating from contaminated water, soil, or handling equipment (Harris *et al.*, 2003). For instance, *Listeria monocytogenes* has been linked to outbreaks associated with fruits. Fungal contamination is also prevalent, with species such as *Aspergillus*, *Penicillium*, and *Fusarium* commonly found on fruits. These fungi can produce mycotoxins, such as aflatoxins and fumonisins, which are harmful to human health (Akinmusire, 2011). Additionally, yeasts like *Saccharomyces cerevisiae* and *Candida spp.* are often involved in the spoilage of fruits, causing fermentation and off-flavours. The presence of these microorganisms not only reduces the shelf life and market value of fruits but also raises public health concerns. Effective measures, including proper washing, refrigeration, and the use of safe handling practices, are essential to minimize microbial contamination and ensure the safety of fruits for consumers (Saba and Oranusi, 2020). Microbial contamination of fruits is not only a food safety issue but also a public health concern. The consumption of contaminated fruits has been linked to

foodborne illnesses, such as diarrhoea, typhoid, and listeriosis (Adedeji *et al.*, 2013). Moreover, the emergence of antimicrobial resistance among fruit-associated microorganisms intensifies the risk, as treatment of resulting infections becomes more challenging (Adeshina *et al.*, 2021). Given the increasing demand for fruits in urban areas like Zaria and the associated health risks, assessing the microbial quality of fruits sold in retail outlets is crucial. This study aims to isolate and identify microorganisms associated with vended fruits, evaluate their antimicrobial susceptibility, and assess their pathogenicity to provide evidence-based recommendations for improving fruit consumption safety.

MATERIALS AND METHODS

Study Area

The study was conducted in Sabon Gari local government, Zaria, Kaduna State, Nigeria.

Sample Collection

Twenty-seven (27) different fruit samples comprising of pawpaw, pineapple and water melon were purchased from three key areas in Sabon gari LGA; Samaru market, Kwangila and Sabon gari, from prominent fruit vendors. The samples were then aseptically placed in a sterile polythene bag ensuring no cross-contamination during handling, labelled accordingly and then transported immediately to the Microbiology Laboratory of Federal University, Dutsin-Ma (FUDMA) for further examination and analysis.

Sample Analysis

A total of 25g of each sample was ground using a laboratory mortar and pestle, and the solution was then used for the analysis. A serial dilution method was employed using aseptic technique. 9 mL of media was dispensed into each of ten test tubes with a sterile automatic dispenser or sterile 10 mL pipettes. The tubes were then labelled from 10^{-1} to 10^{-10} , indicating the dilution factor. Then, 1 mL of the enrichment sample was added aseptically to the first tube (10^{-1}) and mixed gently. Subsequently, 1 mL of this dilution was taken and added to the next tube (10^{-2}) and mixed gently. This procedure was repeated for the remaining tubes (10^{-3} to 10^{-10}).

Media Preparation

The media used for microbial analysis included Nutrient agar, EMB, Salmonella and Shigella agar, and Potato Dextrose agar. All media were prepared according to the manufacturer's instructions. Sterilization of the media was achieved using an autoclave at 121°C for 15 minutes.

The sterility of the prepared media was verified by incubating randomly selected plates at 37°C for 24 hours to ensure no contamination (Fawole and Oso, 2001).

Isolation of Microorganisms

Isolation of Bacteria

The pour plate technique was adopted for isolating bacteria. Immediately after the agar had solidified with the sample, it was incubated at 37°C for 24-48 hours. After incubation, the plates were observed for different colony morphologies and recorded as colony-forming units per gram (CFU/g) of fruit (Barro *et al.*, 2006).

Gram Staining

A thin smear of each bacterial isolate was prepared on a clean glass slide, heat-fixed, and stained using the Gram staining procedure. A smear of the bacterial isolates was made on a sterile, grease-free slide and heat-fixed by passing it through a flame. It was then flooded with crystal violet stain for 30-60 seconds, stained, and then rapidly washed off with clean water. The water was tipped from the slide, and then the slide was flooded with Lugol's iodine for 30-60 seconds and washed with clean water. After decolorizing rapidly (a few seconds) with alcohol, it was washed with clean water. Then, the smear was flooded with safranin for 60 seconds and washed off with clean water. The back of the slide was wiped clean using dried cotton wool and placed on a draining rack for the smear to air-dry. The smear was examined microscopically with the oil immersion objective to differentiate between Gram-positive and Gram-negative bacteria (Cheesbrough, 2006).

Identification of Microorganisms

Biochemical tests were performed to identify the isolated microorganisms. These tests included:

Catalase Test

A drop of 3% hydrogen peroxide was placed on a clean glass slide using a sterile loop. A colony of the test organism was picked and mixed with the hydrogen peroxide. The presence of bubbles indicated a positive catalase reaction (Cheesbrough, 2006).

Coagulase Test

A drop of human plasma was placed on a glass slide. A loopful of bacterial suspension was mixed with the plasma. The formation of clumps within a few seconds indicated a positive coagulase reaction (Fawole and Oso, 2001).

Triple Sugar Iron Test

The Triple Sugar Iron (TSI) test is used to differentiate Gram-negative enteric bacteria based on their ability to ferment glucose, lactose, and sucrose, and produce hydrogen sulfide (H₂S) gas. TSI agar is prepared, poured into test tubes, and solidified in a slanted position. A

bacterial colony is picked using a sterile inoculating needle and inoculated by stabbing the butt and streaking the slant. The tube is loosely capped and incubated at 37°C for 18-24 hours. After incubation, results are observed based on color changes: a yellow slant and yellow butt indicate fermentation of glucose, lactose, and/or sucrose, while a red slant and yellow butt suggest glucose fermentation only. A red slant and red butt signify no fermentation. Black precipitate in the butt confirms H₂S production, and gas bubbles or cracks indicate gas production (Cheesbrough, 2006).

Indole Test

3 mL of tryptone broth was inoculated with the bacterial isolate and incubated at 37°C for 24-48 hours. Then, 0.5 mL of Kovac's reagent was added to the broth culture and shaken gently. The formation of a red or pink layer on the surface indicated a positive result (Cheesbrough, 2006).

Citrate Utilization Test

Simon citrate agar was used to test the organism's ability to utilize citrate as a source of energy. The test organism was streaked on Simon citrate agar slant and incubated at 37°C for 24-48 hours. A shift in pH turned the bromothymol blue indicator in the medium from green to blue. If the medium turned blue, the organism was citrate positive; if there was no color change (i.e., it remained green), the organism was citrate negative.

Urease Test

A sterile technique was used. The test organism was inoculated into a bijoux bottle containing 3 mL of sterile Christensen's modified urea broth using an inoculation wire loop. The tubes were then incubated at 37°C for 3 to 12 hours and observed for a colour change from orange to pink, indicating a positive result (Cheesbrough, 2006).

Oxidase Test

A loopful of oxidase reagent (tetramethyl-p-phenylenediamine) was placed on a filter paper in a petri dish. Using a plastic wire loop, a colony of the organism was smeared across the wetted filter paper and observed for a colour change within 30 seconds. A dark purple colouration indicated a positive result.

Motility Test

A semi-solid motility agar was used. The test organism was inoculated into the semi-solid motility agar by stabbing the medium with a needle and incubated at 37°C for 24-48 hours. The medium was observed for diffuse growth radiating from the stab line, which indicated motility.

Methyl Red (MR) Test

An overnight culture of the organisms was inoculated into appropriately labelled tubes of MR-VP broth (containing buffered peptone 7 g, glucose 5 g, and dipotassium phosphate 5 g) using an inoculation loop. A tube without the inoculum was kept as a control. Three drops of MR indicator were added, and the tubes were incubated at 37°C for 24-72 hours. The tubes were then observed, and the appearance of a red colouration indicated a positive result (Cheesbrough, 2006).

Antibiotic Susceptibility Test

Antibiotic susceptibility testing was performed using the disc diffusion method. Each colony of the test isolate was picked with a wire loop and inoculated into nutrient broth and incubated for 3 hours. The turbidity of each broth culture was adjusted to correspond to 0.5 McFarland turbidity standards (corresponding to approximately 10^8 cfu/ml). Each standardized broth culture was used to inoculate the surface of the Mueller-Hinton (MH) agar plates, and the excess fluid was drained into a disinfectant jar. The surface of each inoculated plate was allowed to dry. Using a disc dispenser, the antibiotic discs were aseptically placed on the surface of the inoculated agar plates and then incubated for 24 hours at 37°C. After incubation, the plates were examined for zones of inhibition around each disc. The diameters of the zones of inhibition were measured with a meter rule and recorded.

Isolation of Fungi

A drop of lactophenol cotton blue stain was dropped in the centre of a clean slide. A fragment of the fungus was purchased with the aid of a wire loop and placed in the drop of the stain, teased gently, and covered with a coverslip. The coverslip was not pushed down or tapped to avoid the dislodging of the conidia from the conidiophores. The stained isolate was then viewed under the microscope with $\times 10$ and $\times 40$ objective lenses for its morphological characteristics (Cheesbrough, 2006).

Data Analysis

Data obtained were sorted according to fruit kind and the location from where they were purchased and entered into a Microsoft Excel spreadsheet and presented in tables, piechart and barchats

RESULTS AND DISCUSSION

Mean bacterial load (CFU/ml) of fruit samples purchased from different study areas in Zaria

Table 1 presents the total bacterial load of all isolates from the fruit samples purchased across the study areas. The highest bacterial colony count was observed in PZ (Pawpaw sample PP1, 5.2×10^4 CFU/ml), while the lowest was recorded in Samaru (Pineapple sample PA1, 1.1×10^4 CFU/ml). These results indicate significant bacterial contamination levels across different locations,

with PZ showing the highest microbial presence.

Mean fungal load (pfu/g) of fruit samples purchased from different study areas in Zaria

The results of the total fungal count across the study sites is presented in table 2. The highest fungal colony count was recorded in PZ (Watermelon sample WM1, 9.4×10^4 PFU/g), whereas the lowest was found in Samaru (Pineapple sample PA1, 1.1×10^4 PFU/g). The findings reveal varying levels of fungal contamination, with watermelon samples exhibiting the highest fungal load across locations.

Distribution of bacterial isolates based on media used, colony morphology, gram reaction, and biochemical test

Table 3 shows the bacterial isolates' distribution based on growth media, morphological characteristics, and biochemical test results. A total of 10 bacterial species were identified, with Gram-negative bacteria being predominant. The most frequently occurring isolate was *Escherichia coli*, while *Corynebacterium spp.* was the least identified. The results confirm the presence of both pathogenic and opportunistic bacterial species across fruit samples.

Distribution of all bacterial isolates from the study area

The bar chart illustrates the distribution of bacterial isolates from the sampled fruits. The most frequently isolated bacterium was *Staphylococcus aureus*, followed by *Bacillus spp.*, while *Shigella spp.* had the lowest occurrence. The prevalence of these isolates highlights potential public health risks, as some of these bacteria are known foodborne pathogens.

Distribution of isolates based on each study area

Table 5 presents the distribution of all bacterial isolates from each study site. Samaru had the highest number of isolates, while PZ recorded the least. The variation in isolate distribution across sites may be due to differences in fruit handling, storage, and hygiene practices at each location.

Morphological and microscopic characterization of all fungal isolates from different study areas

The results of the morphological and microscopic characteristics of fungal isolates from fruit samples is presented in table 6. The most frequently occurring fungi were *Aspergillus niger*, *Aspergillus flavus*, and *Candida spp.* Morphological examination showed distinct spore structures, colony colors, and mycelial growth, aiding in fungal identification.

Antifungal susceptibility test profile of fungal isolates

Table 7 shows the susceptibility of fungal isolates to different antifungal concentrations (100 mg/ml, 50

mg/ml, 25 mg/ml). *Candida spp.* exhibited the highest sensitivity with the largest zone of inhibition (32 mm at 100 mg/ml), while *Fusarium spp.* and *Rhizopus spp.* showed lower susceptibility. These results indicate varying antifungal resistance patterns among the isolated fungi.

Antibacterial susceptibility test profile of bacterial isolates

The results of the antibacterial susceptibility profiles of Gram-negative and Gram-positive bacterial isolates are presented in table 8 and Table 8.1 respectively. Among Gram-negative isolates, *Escherichia coli* and *Proteus vulgaris* exhibited multidrug resistance, while *Shigella spp.* was susceptible to most antibiotics. Among Gram-positive isolates, *Staphylococcus aureus* showed intermediate resistance, while *Bacillus spp.* was largely susceptible. These findings shows the emerging antibiotic resistance among bacterial isolates, which may pose

treatment challenges in foodborne infections.

Distribution of all fungal isolates from the study areas

Table 9 shows the occurrence of fungal isolates across the study sites. The most frequently identified fungi were *Aspergillus spp.*, *Penicillium spp.*, and *Candida spp.*, while *Fusarium spp.* had the lowest occurrence. These fungi are commonly associated with fruit spoilage and potential mycotoxin production.

Pathogenicity test results on fresh mature fruits

Table 10 presents the pathogenicity test results, showing fruit spoilage patterns upon inoculation with fungal isolates. The most aggressive spoilage was observed in *Rhizopus spp.* (black fuzzy mold, rapid fruit liquefaction), while *Saccharomyces cerevisiae* showed minimal pathogenic effects. This confirms the ability of certain fungal isolates to cause post-harvest fruit deterioration.

Table 1: Mean bacterial load (cfu/ml) of fruit samples purchased from different study areas in Zaria

S/N	Sample	Mean Total Colony Count (CFU/ml)
1	PP1 Samaru	4.4×10^4
2	PP3 Samaru	3.5×10^4
3	WM1 Samaru	5.0×10^4
4	WM2 Samaru	2.4×10^4
5	WM3 Samaru	3.8×10^4
6	PA1 Samaru	1.1×10^4
7	PA2 Samaru	2.1×10^4
8	PP1 Kwangila	2.8×10^4
9	PP3 Kwangila	1.7×10^4
10	WM1 Kwangila	2.6×10^4
11	WM2 Kwangila	2.1×10^4
12	WM3 Kwangila	3.8×10^4
13	PA1 Kwangila	2.1×10^4
14	PA2 Kwangila	1.9×10^4
15	PA3 Kwangila	2.0×10^4
16	PP1 PZ	5.2×10^4
17	PP2 PZ	3.5×10^4
18	PP3 PZ	2.4×10^4
19	WM1 PZ	3.8×10^4
20	PA1 PZ	2.1×10^4
21	PA2 PZ	3.9×10^4
22	PA3 PZ	2.2×10^4

Key:

PP = Pawpaw
WM = Watermelon
PA = Pineapple

Table 2: Mean fungal load Pfu/ml) of fruit samples purchased from different study areas in Zaria

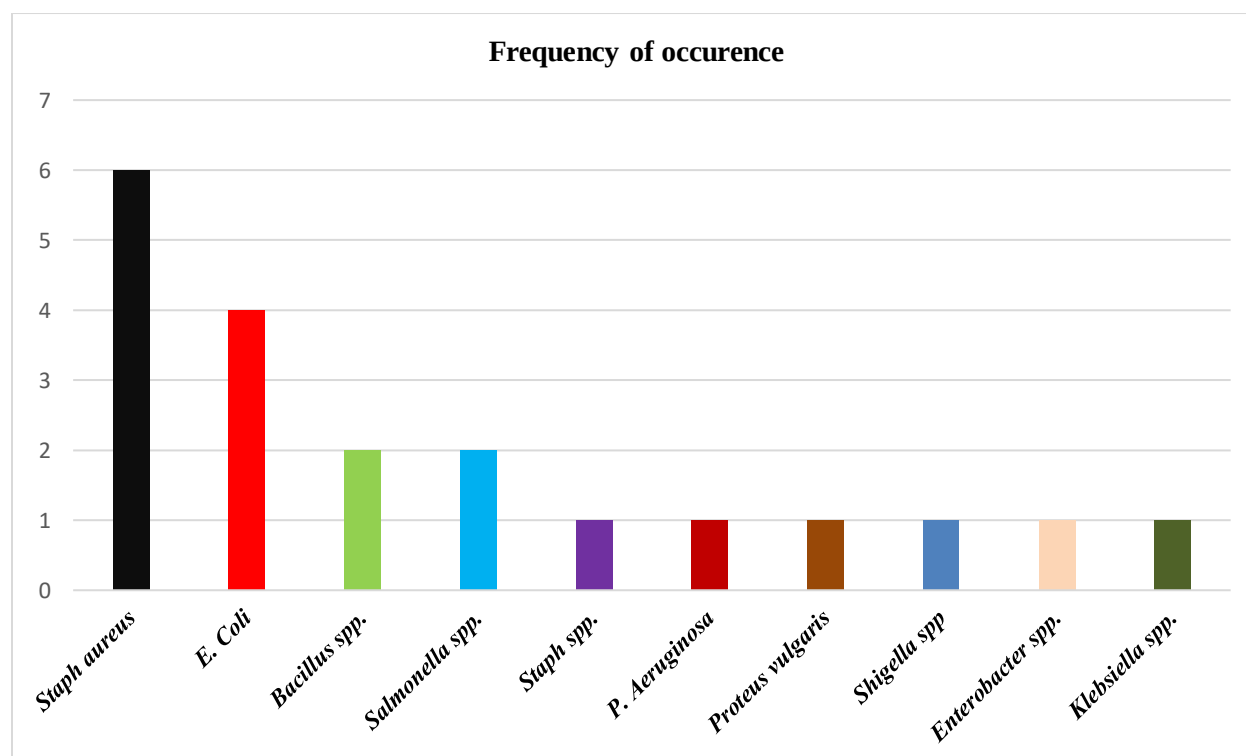
S/N	Sample	Mean total colony count (PFU/ml)
1	PP1 Samaru	2.8×10^4
2	PP2 Samaru	1.9×10^4
3	PP3 Samaru	2.4×10^4
4	WM1 Samaru	2.8×10^4
5	WM2 Samaru	3.7×10^4
6	PA1 Samaru	1.1×10^4
7	PA2 Samaru	1.5×10^4
8	PA3 Samaru	1.8×10^4
9	PP1 Kwangila	2.8×10^4
10	PP3 Kwangila	2.4×10^4
11	WM2 Kwangila	3.8×10^4
12	WM3 Kwangila	3.5×10^4
13	PA1 Kwangila	2.9×10^4
14	PA2 Kwangila	2.9×10^4
15	PA3 Kwangila	3.5×10^4
16	PP1 PZ	6.2×10^4
17	PP2 PZ	4.3×10^4
18	PP3 PZ	2.9×10^4
19	WM1 PZ	9.4×10^4
20	WM2 PZ	2.4×10^4
21	PA1 PZ	1.1×10^4
22	PA2 PZ	1.7×10^4

Table 3: Distribution of bacterial isolates based on media used, colony morphology, gram reaction and biochemical test

S/N	MU	Colony Morphology	GR	CA	CO	TS	IN	MR	VP	CI	MO	UR	Presumptive Organism
1	MSA	White/yellow, circular	+	+	-	Na	Na	Na	N	Na	Na	Na	<i>Staphylococcus</i> spp.
2	MSA	Yellow (mannitol fermentation)	+	+	+	Na	Na	Na	Na	Na	Na	Na	<i>S. aureus</i>
3	N/A	Large, dry, rough	+	+	-	Na	Na	Na	Na	Na	Na	Na	<i>Bacillus</i> spp.
4	N/A	Small, dry, white to yellow	-	+	-	Na	-	+	-	+	+	+	<i>P. aeruginosa</i>
5	EMB	Metallic green sheen	-	+	-	Na	+	+	-	-	+	-	<i>E. coli</i>
6	MaC	Swarming, spreading growth	-	+	-	Na	+	+	-	+	+	+	<i>Proteus vulgaris</i>
7	SSA	Colorless (non-H ₂ S producers)	-	+	-	-	-	+	-	-	-	-	<i>Shigella</i> spp.
8	SSA	Black-centered (H ₂ S production)	-	+	-	+	-	+	-	+	+	-	<i>Salmonella</i> spp.
9	EMB	Mucoid pink	-	+	-	Na	-	-	+	+	+	-	<i>Enterobacter</i> spp.
10	MaC	Large, mucoid pink	-	+	-	Na	-	-	+	+	-	+	<i>Klebsiella pneumoniae</i>

Key:

MU – Media Used, GR – Gram Reaction, CA – Catalase, CO – Coagulase, TS – Triple Sugar, IN – Indole, MR – Methyl Red, VP – Voges-Proskauer, CI – Citrate, MO – Motility, UR – Urease

**Figure 2:** Total distribution of bacterial isolates

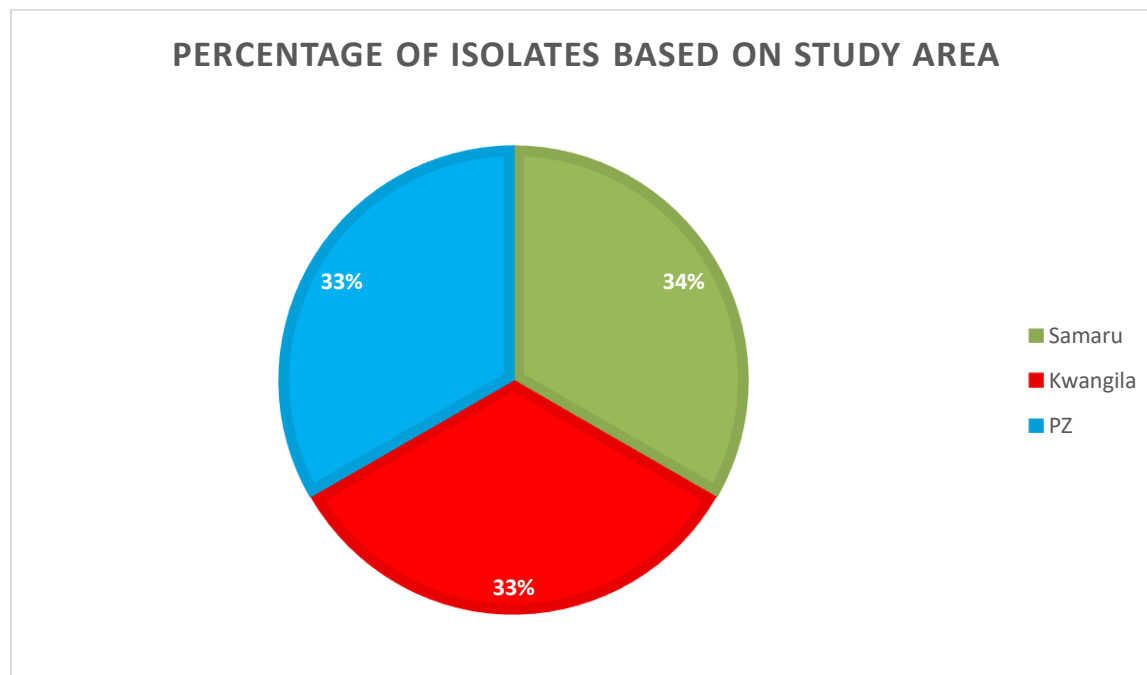


Figure 3: Distribution of Isolates base on each study area

Table 6: Morphological characteristics and microscopic view of all the fungal isolates from the study area.

S/N	Study Area	Sample	Colony Morphology	Microscopic Characteristics	Presumptive Isolate
1	Samaru	PP1	Blue-green, powdery	Septate hyphae, brush-like conidiophores	<i>Penicillium</i> spp.
2	Samaru	PP2	Black, velvety	Septate hyphae, radiating conidial heads	<i>Aspergillus niger</i>
3	Samaru	PP3	White to grey, fluffy	Non-septate hyphae, sporangiophores	<i>Rhizopus</i> spp.
4	Samaru	WM1	Creamy, smooth	Oval budding cells, pseudohyphae	<i>Saccharomyces cerevisiae</i>
5	Samaru	WM2	Black, velvety	Septate hyphae, banana-shaped conidia	<i>Aspergillus niger</i>
6	Samaru	PA1	Yellow-green, powdery	Septate hyphae, conidial heads	<i>Aspergillus flavus</i>
7	Samaru	PA2	Black, velvety	Septate hyphae, conidial heads	<i>Aspergillus niger</i>
8	Samaru	PA3	Creamy, smooth	Oval budding cells, pseudohyphae	<i>Candida</i> spp.
9	Kwangila	PP1	Blue-green, powdery	Septate hyphae, brush-like conidiophores	<i>Penicillium</i> spp.
10	Kwangila	PP3	Black, velvety	Septate hyphae, radiating conidial heads	<i>Aspergillus niger</i>
11	Kwangila	WM2	White, cottony	Non-septate hyphae, sporangiophores	<i>Mucor</i> spp.
12	Kwangila	WM3	Yellow-green, powdery	Septate hyphae, conidial heads	<i>Aspergillus flavus</i>
13	Kwangila	PA1	White to pink, fluffy	Septate hyphae, sickle-shaped conidia	<i>Fusarium</i> spp.
14	Kwangila	PA2	Black, velvety	Septate hyphae, radiating conidial heads	<i>Aspergillus niger</i>
15	Kwangila	PA3	Creamy, smooth	Oval budding cells, pseudohyphae	<i>Candida</i> spp.

16	PZ	PP1	White to pink, fluffy	Septate hyphae, sickle-shaped conidia	<i>Fusarium</i> spp.
17	PZ	PP2	Blue-green, powdery	Septate hyphae, brush-like conidiophores	<i>Rhizopus</i> spp.
18	PZ	PP3	Yellow-green, powdery	Septate hyphae, conidial heads	<i>Aspergillus flavus</i>
19	PZ	WM1	Black, velvety	Septate hyphae, radiating conidial heads	<i>Aspergillus niger</i>
20	PZ	WM2	Yellow-green, powdery	Septate hyphae, conidial heads	<i>Aspergillus niger</i>
21	PZ	PA1	Blue-green, powdery	Septate hyphae, brush-like conidiophores	<i>Penicillium</i> spp.
22	PZ	PA2	Creamy, smooth	Oval budding cells, pseudohyphae	<i>Candida</i> spp.

Table 7: Antifungal susceptibility test profile of fungal isolates to Fluconazole

S/N	Fungal Isolate	Concentration (mm)		
		100mg/ml	50mg/ml	25mg/ml
1	<i>Penicillium</i>	10	6	2
2	<i>Aspergillus Niger</i>	-	-	-
3	<i>Aspergillus flavus</i>	2	-	-
4	<i>Candida</i>	25	12	7
6	<i>Rhizopus</i>	3	1	-
7	<i>Saccharomyces Cereviceae</i>	23	13	6
8	<i>Mucor</i>	-	-	-

Source: CLSI M100, 34th Edition (2024).

Key: R = 0 ≤ 10mm, S = > 10mm

Table 8: Antibacterial susceptibility testing of gram negative bacterial isolates from fruit samples

S/N	Isolate	AM	CPX	SXT	PEF	APX	GN
1	<i>Escherichia coli</i>	10 (R)	22 (S)	12 (R)	24 (S)	13 (R)	19 (S)
2	<i>Salmonella</i> spp.	8 (R)	20 (S)	14 (R)	23 (S)	12 (R)	21 (S)
3	<i>Proteus vulgaris</i>	9 (R)	18 (S)	11 (R)	20 (S)	15 (S)	18 (S)
4	<i>Shigella</i> spp.	11 (R)	19 (S)	13 (R)	22 (S)	14 (R)	16 (S)
5	<i>Klebsiella pneumoniae</i>	7 (R)	21 (S)	10 (R)	25 (S)	16 (S)	20 (S)
6	<i>Enterobacter</i> spp.	12 (R)	23 (S)	15 (S)	21 (S)	18 (S)	22 (S)
9	<i>Pseudomonas</i> spp.	7 (R)	12 (R)	8 (R)	15 (R)	10 (R)	13 (R)

Source: CLSI M100, 34th Edition (2024).

Key:

AM= Ampicillin S ≥ 14 mm, R ≤ 13 mm

CPX= Ciprofloxacin: S ≥ 18 mm, R ≤ 17 mm

SXT = Trimethoprim-Sulfamethoxazole: S ≥ 16 mm, R ≤ 15 mm

PEF = Pefloxacin: S ≥ 19 mm, R ≤ 18 mm

APX= Ampicillin-Sulbactam: S ≥ 15 mm, R ≤ 14 mm

GN= Gentamicin: S ≥ 17 mm, R ≤ 16 mm

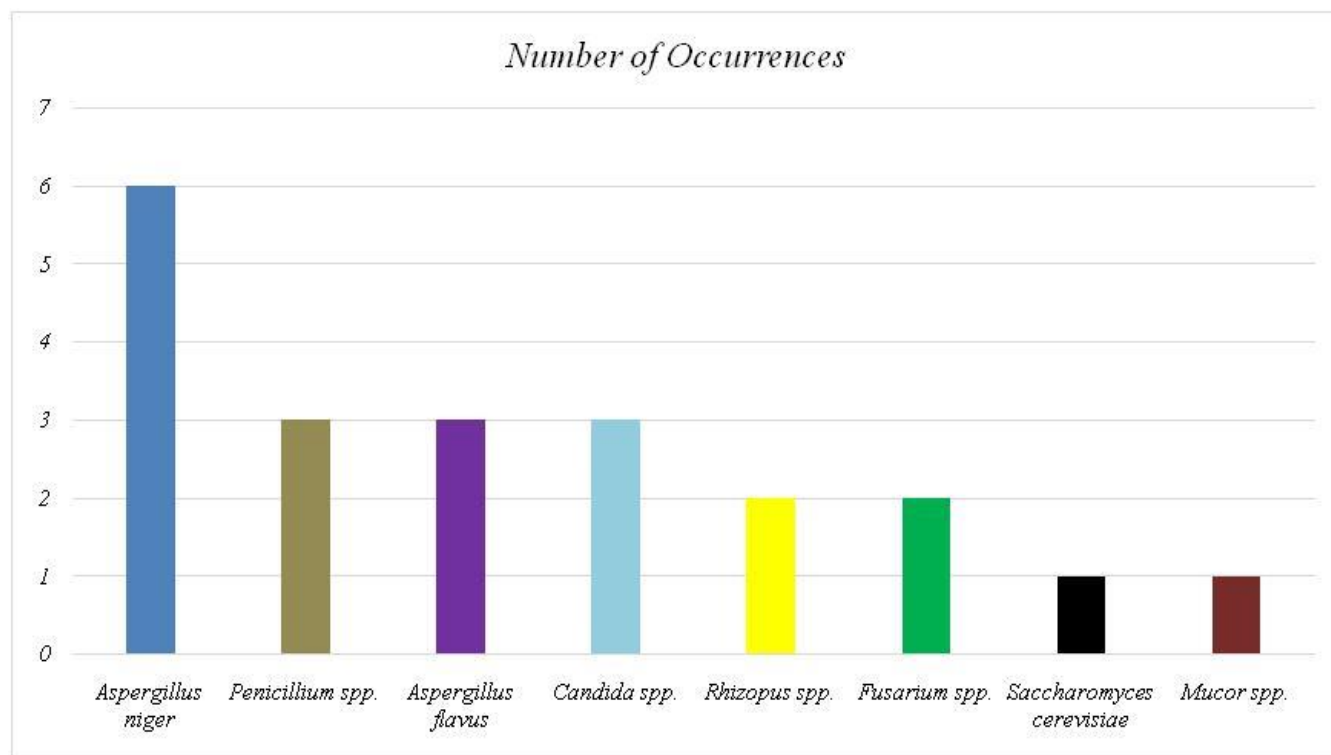
R = Resistance

S = Susceptible

Table 9: Antibacterial susceptibility testing of gram-positive bacterial isolates

S/N	Isolate	AM	SXT	PEF	APX	STREP	R	E	Z
1	<i>Staphylococcus aureus</i>	14 (R)	12 (R)	18 (S)	13 (R)	22 (S)	20 (S)	21 (S)	15 (R)
2	<i>Bacillus spp.</i>	16 (S)	14 (R)	19 (S)	12 (R)	20 (S)	18 (S)	22 (S)	17 (S)
3	<i>Staphylococcus spp.</i>	12 (R)	10 (R)	17 (S)	14 (R)	19 (S)	19 (S)	23 (S)	14 (R)

Source: CLSI M100, 34th Edition (2024).

Key:**AM** = Amoxicillin: S: ≥ 15 mm R: ≤ 14 mm, **SXT** = Sulfamethoxazole-Trimethoprim: S: ≥ 16 mm, R: ≤ 14 mm**PEF** = Pefloxacin: S: ≥ 17 mm, R: ≤ 15 mm, **APX** = Ampicillin: S: ≥ 17 mm R: ≤ 13 mm**STREP** = Streptomycin: S: ≥ 18 mm R: ≤ 14 mm, **R** = Rifampin: S: ≥ 20 mm R: ≤ 16 mm**E** = Erythromycin: S: ≥ 21 mm R: ≤ 15 mm, **Z** = Azithromycin: S: ≥ 16 mm R: ≤ 14 mm**Figure 4:** Distribution of fungal isolate for the study areas**Table 10:** Pathogenicity test results on fresh mature fruits:

S/N	Fruit Type	Colony Morphology	Probable Isolate	Pathogenicity
1	Pawpaw	Greenish-blue mold, soft rot, fruit shriveling	<i>Penicillium spp.</i>	Positive
2	Pawpaw	Whitish slimy patches, softening, fermentation odor	<i>Candida spp.</i>	Positive
3	Pawpaw	White fluffy mycelium, soft rot, rapid spoilage	<i>Mucor spp.</i>	Positive
4	Watermelon	Black mold, rapid fruit decay, wrinkling	<i>Aspergillus niger</i>	Positive
5	Watermelon	Pinkish-white mold, watery rot, internal discoloration	<i>Fusarium spp.</i>	Positive
6	Watermelon	Black fuzzy mold, rapid fruit liquefaction, leaking	<i>Rhizopus spp.</i>	Positive
7	Pineapple	Yellow-green mold, dry rot, potential toxin production	<i>Aspergillus flavus</i>	Positive
8	Pineapple	No visible mold, slight fermentation odor	<i>Saccharomyces cerevisiae</i>	Positive

Discussion

The microbial quality of fruits purchased in retail outlets in Zaria, Kaduna State, Nigeria, was found to have significant bacterial and fungal contaminations, posing potential health risks for consumers. The presence of pathogenic bacteria and fungi across all sampling locations indicates contamination from multiple sources, such as poor handling, environmental exposure, and inadequate hygienic practices during irrigation, handling, transportation, storage and processing into read-to-eat vended fruits. The microbial quality assessment of fruits obtained from retail outlets in Zaria revealed significant contamination levels across different fruit types and locations. The bacterial and fungal loads varied among the sampled fruits, with pawpaw and watermelon exhibiting the highest microbial counts, particularly in the PZ area. This finding aligns with previous studies indicating that fruits with high moisture content, such as watermelon and pawpaw, are more susceptible to microbial growth due to their nutrient-rich environment (Omemu & Bankole, 2008). The presence of pathogenic microorganisms highlights the potential public health risks associated with consuming unwashed or improperly handled fruits. The bacterial isolates identified in this study included *Escherichia coli*, *Staphylococcus aureus*, *Bacillus spp.*, *Salmonella spp.*, *Proteus vulgaris*, and *Shigella spp.* The predominance of Gram-negative bacteria such as *E. coli* and *Salmonella spp.* suggests fecal contamination, likely resulting from inadequate hygiene during handling, transportation, and storage (Nwachukwu et al., 2018). The presence of *E. coli* in fruits is often an indication of fecal contamination, as observed in similar studies where poor sanitation and the use of contaminated water sources contributed to bacterial load (Oladipo & Adeoye, 2014). Additionally, the detection of *Staphylococcus aureus*, a common skin and nasal bacterium, indicates contamination from food handlers, reinforcing the need for proper hand hygiene and handling practices (Harris et al., 2003). Fungal isolates were also prevalent in the sampled fruits, with *Aspergillus niger*, *Aspergillus flavus*, *Penicillium spp.*, *Candida spp.*, *Mucor*

spp., and *Rhizopus spp.* being the most frequently identified. The highest fungal load was recorded in watermelon samples from PZ, with *Aspergillus* species being the most dominant. These findings align with previous research indicating that *Aspergillus* species are commonly associated with fruit spoilage and mycotoxin production (Akinmusire, 2011). The presence of mycotoxin-producing fungi such as *Aspergillus* and *Fusarium* raises concerns regarding potential health risks, including aflatoxin exposure, which has been linked to liver carcinogenicity (Ezekiel et al., 2022). The antimicrobial susceptibility testing of bacterial isolates revealed varying resistance patterns, with multidrug resistance observed in *Escherichia coli* and *Proteus vulgaris*. These findings are consistent with the study of Adeshina et al., 2021, who demonstrate the increasing trend of antimicrobial resistance in foodborne pathogens, posing a significant challenge to infection management. Among Gram-positive isolates, *Staphylococcus aureus* showed intermediate resistance, while *Bacillus spp.* was largely susceptible to tested antibiotics. The high level of resistance observed in some bacterial isolates may be attributed to the misuse of antibiotics in agriculture and food production, emphasizing the need for stricter regulations and monitoring of antimicrobial use (FAO 2013). Fungal susceptibility testing indicated that *Candida spp.* exhibited the highest sensitivity to antifungal agents, whereas *Rhizopus spp.* and *Fusarium spp.* displayed lower susceptibility. The variation in antifungal resistance among isolates suggests the need for targeted antifungal treatments in food preservation and post-harvest management (Erhirhie et al., 2020). Additionally, the pathogenicity test confirmed the spoilage potential of isolated fungi, with *Rhizopus spp.* causing rapid fruit degradation. This reinforces the importance of proper storage and handling practices to prevent fungal-induced spoilage (Saba and Oranus, 2020).

CONCLUSION

The findings of this research have therefore established the presence of different microorganisms associated with selected retail fruits vended within Zaria metropolis. The study revealed presence of both gram-positive and gram-negative bacteria to include *Staphylococcus aureus*, *Escherichia coli*, *Bacillus spp.*, *Pseudomonas aeruginosa*, *Proteus vulgaris*, *Shigella spp.*, *Salmonella spp.*, *Enterobacter spp.*, *Klebsiella spp.*, and *Staphylococcus spp.* The fungi species identified include *Aspergillus niger*, *Penicillium*, *Aspergillus flavus*, *Candida spp.*, *Rhizopus spp.*, *Saccharomyces cerevisiae*, and *Mucor*. The variations in microbial load across different study areas highlight the role of hygiene

practices, storage conditions and environmental factors in fruit contamination. The antimicrobial resistance observed among bacterial isolates raised concerns about drug resistance foodborne pathogens which could pose challenges in treating infections. The presence of *E. coli* and *S. Aureus* suggest a risk of foodborne infections especially when fruits are consumed raw without proper washing. Additionally, fungi contaminants such as *A. Flavus* and *Penicilium spp.* are known producers of mycotoxins which can cause serious health complications, including liver damage and immune suppressions. Some of the microorganisms were resistance and susceptible to various degrees of antibiotics tested. Pathogenicity

tests also link certain fungi and bacteria to fruit spoilage, leading to post-harvest losses and economic impacts. These findings stress the importance of stringent control measures to minimize contamination and improve fruit safety for consumers.

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