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Isolation and Identification of Bacteria Associated with the Spoilage of *Cucumis melo* (Golden Melon) Sold in Zuba and Gwarimpa Market

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Golden melon (*Cucumis melo*) is a popular fruit consumed in Nigeria, prized for its sweet flavour and juicy texture. However, spoilage caused by microbial contamination is a major concern, particularly in markets like Zuba and Gwarinpa, where improper handling and storage practices are common. This study aimed to isolate and identify bacteria associated with the spoilage of golden melon sold in Zuba and Gwarinpa markets in Abuja, Nigeria. Samples were collected and analysed for proximate, bacteria, pathogenicity test and antibiogram susceptibility analysis using standard methods and procedures. The results revealed a diverse bacterial population associated with the spoilage of golden melon, including species of *Enterobacteriaceae*, *Pseudomonas*, *Escherichia coli,*

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Proteus mirabilis, Klebsiella pneumoniaae, Enterobacter aerogenes, Shigella sp, Salmonella sp. All the bacteria species were positive to pathogenicity test except Enterobacter aerogenes, which test negative. To conclude, this study provides valuable insights into the bacterial diversity associated with the spoilage of golden melon in Zuba and Gwarinpa markets. The findings underscore the importance of implementing proper hygiene and storage practices to minimize microbial contamination and extend the shelf life of golden melon.

Keywords: Golden melon; Cucumis melo; bacteria, Zuba, Gwarmpa.

1. INTRODUCTION

The golden melon, also known as *Cucumis melo* is a type of melon that is characterized by its bright golden skin and sweet, juicy flesh. This variety of melon is known for its high sugar content and refreshing flavor, making it a popular choice for desserts, salads, and snacks [1]. Golden melons are typically grown in warm, sunny climates and are harvested in the summer months when they are at their peak ripeness. They are rich in vitamins and minerals, including vitamin C, vitamin A, and potassium, making them a healthy addition to any diet [1]. Golden melon has been reported to be rich in essential vitamins and minerals such as Vitamin C, Vitamin A, potassium, and fiber [2]. These nutrients help boost the immune system, promote healthy skin, and improve overall health [3]**.** Golden melon has a high water content, making it a hydrating fruit and is low in calories and fat, making it a healthy option for weight management. The fiber content in golden melon helps improve digestion and regulate bowel movements. It can prevent constipation and promote a healthy digestive system [2]. Fruits have been reported to be rich in antioxidants such as Vitamin C and beta-carotene, which help reduce inflammation, fight off free radicals, and protect the body against chronic diseases [3]**.** The Vitamin A content in golden melon promotes healthy skin by reducing signs of aging, improving skin elasticity, and maintaining a clear complexion.

Globally, microbial contamination of fruit has been reported as a public health issue as the presence of harmful bacteria, viruses, or fungi has been reported on the surface or inside of fruits [4]. Contamination of fruit can occur at various stages of production, processing, storage, transportation, and handling of fruit. Some common sources of microbial contamination of fruit include soil, water, equipment, and contact with workers or animals. Contaminated fruit can lead to foodborne illnesses such as gastroenteritis, diarrhea, and vomiting [1]. Some common bacteria associated with spoilage of golden melon (*Cucumis melo*) include *Pseudomonas* spp; *Bacillus* spp; *Listeria monocytogenes* and *Enterobacteriacea*e [5,6]. Some of these bacterial are aerobic bacteria that are known for their ability to produce enzymes that break down organic matter, leading to spoilage of fruits and vegetables like golden melon while some are spore-forming bacteria that are commonly found in soil and can contaminate fruits and vegetables during harvesting and processing, leading to spoilage [6] While species such as *Listeria monocytogenes* are pathogenic bacteria that can cause serious foodborne illnesses if consumed and can contaminate golden melons during harvesting, processing, or storage while the *Enterobacteriacea* bacteria are commonly found in the environment and can contaminate golden melons, leading to spoilage and potential foodborne illnesses [4,5]**.**

Zuba Market and Gwarimpa Markets are popular markets located in Nigeria's Federal Capital Territory, Abuja. Zuba Market is known for its wide range of products, including groceries, clothing, electronics, and household goods. It is a bustling market that attracts a large number of shoppers daily. Gwarimpa Market, on the other hand, is also a busy market with a diverse selection of goods, ranging from fresh produce and meat to clothing and household items. It is popular among residents of Gwarimpa and the surrounding areas for its convenient location and affordable prices. Both markets are known for their vibrant atmosphere and are important hubs for trade and commerce in Abuja Federal Capital City of Nigeria. Thus, this study is designed to isolate and identify bacteria species associated with the spoilage of golden melon (*Cucumis melo*) sold in Zuba and Gwarimpa market

2. MATERIALS AND METHODS

Study area: Zuba Market and Gwarimpa Markets are popular markets located in Abuja Federal Capital Territory of Nigeria (Fig. 1). This Market serves the people of Abuja city for their daily needs.

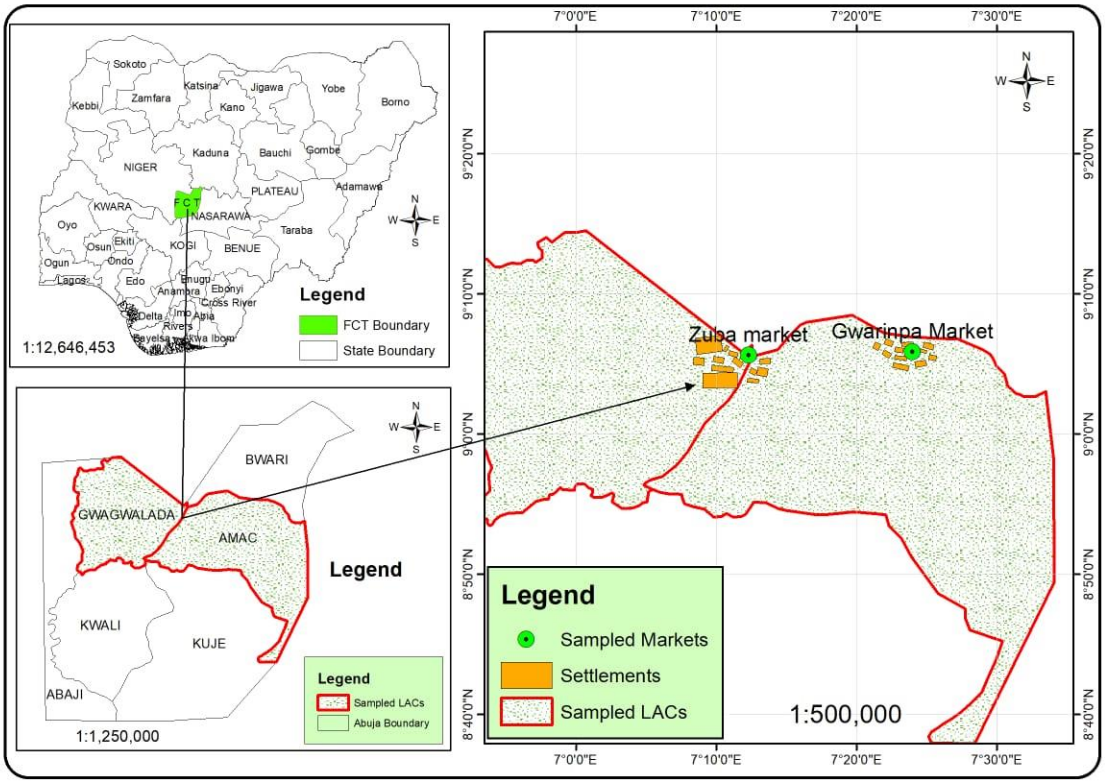

Fig. 1. Map of Nigeria showing federal capital territory with Zuba and Gwarimpa Market Abuja Nigeria

Sample collection: Samples of healthy fruits of *Cucumis melo* were bought from fruit vendors at Zuba Market and Gwarimpa Markets, the fruit samples were transported to to the Department of Biology, Microbiology and Biotechnology, Nile University of Nigeria Abuja Nigeria where it was observed for spoilage.

Determination of Proximate Analysis: The proximate analysis was determined using the standard techniques of Analysis of the Association of Official's Analytical Chemists [7]. The parameters determined include moisture content, ash, fat, protein and carbohydrate.

Isolation of bacteria from *Cucumis melo*: Isolation of bacteria was done using the classical phytopathological method whereby small samples of *Cucumis melo* were cut from points showing visible signs of spoilage to the healthy portions and inoculated onto a sterile distilled water [8]. Thereafter the samples were subjected to serial dilution and 28.0g of nutrient agar was be dissolved in 1 litre of distilled water and the growth media was autoclaved at 121°C temperature for 15 minutes following manufacturer's instruction. The media were allowed to cool down in a sterilized chamber and then pour into each petri dishes containing 1 ml of the diluents. Thereafter, it was allowed to solidify and incubated at a temperature of 37°C for 24 hrs.

Identification and characterization of bacterial isolates: All bacteria isolates were sub-cultured and transferred to a slant media to obtain a pure culture where a gram-staining was conducted to identify the isolates based on the method described by Cheesebrough [9]. Thereafter, various biochemical tests such as catalase test, coagulase test, methyl red test, vogue proskeur test, indole test, citrate utilisation and sugar fermentation test were conducted for further characterization of the isolates [9].

Pathogenicity tests of bacterial isolates: All of the bacterial strains identified by the biochemical test were tested on golden melon. The bacterial suspension of bacteria isolate were prepared in sterile distilled water and infiltrated into the mesocap area of the fruit by using a 2ml syringe. The inoculated fruit were incubated in a completely randomized in laboratory bench for 24–48 h at 20–28 °C. The presence of

inoculation site was recorded within 24–48 h after infiltration. This test was repeated at least three times for each strain. For pathogenicity tests sterilized distilled water (sdH2O) was used as a negative control. The bacterial strains that induced fruit were selected and tested for pathogenicity on young shoots of Golden Delicious apples as described previously Kotan and Sahin [10].

Antibiogram of antibiotics against the isolated bacteria: The disk diffusion agar method tests the effectiveness of antibiotics on a specific microorganism. The isolates were tested against Ceftazidime (30µg), Cefuroxime (30µg), Gentamicin (10µg), Ceftriaxone (30µg), Erythromycin (5µg), Cloxacillin (5µg), Ofloxacin (5µg) and Augmentin (30µg). A nutrient agar plate was first spread with bacteria, and then paper disks of antibiotics were placed atop of it. The bacteria were then allowed to grow on the media at 37ºC for 24 hours and then it was observed for growth and effect of the antibiotic on it. The zone of inhibition produced was measured and recorded. The amount of space around every antibiotic disk indicated the lethality of that antibiotic on the bacteria in question. The zone of inhibition was interpreted as susceptible or resistant or intermediate based on the standard of the Clinical and Laboratory Standard Institute (CLSI).

3. RESULTS

Proximate analysis of *Cucumis melo* sample collected from Zuba and Gwarimpa market Abuja Nigeria. The proximate analysis of *Cucumis melo* sample collected from Zuba and Gwarimpa market Abuja Nigeria is shown in Table 1. Percentage ash and carbohydrate were highest in *Cucumis melo* collected from Zuba Market while moisture content, fat and protein were highest in *Cucumis melo* collected from Gwarimpa market.

Bacteria colony count of *Cucumis melo* sample collected from Zuba and Gwarimpa market Abuja Nigeria. The bacteria colony count of *Cucumis melo* sample collected from Zuba and Gwarimpa market Abuja Nigeria is shown in Table 2. Bacterial colony was highest in *Cucumis melo* collected from Gwarimpa Market and lowest in Zuba Market. The biochemical characteristics of bacteria isolates are shown in Table 3. A total of five bacteria species were isolated from *Cucumis melo* sample collected from Zuba market. The bacteria species are *Escherichia coli, Proteus mirabilis, Klebsiella Pneumoniae, Enterobacter aerogenes* and *Pseudomonas aeruginosa.* While in Gwarimpa market also, a total of five bacteria species were identified which are *Shigella* sp., *Salmonella* sp., *Pseudomonas aeruginosa, Escherichia coli* and *Klebsiella Pneumoniae.*

Pathogenicity test, antibiotic susceptibility and resistant of bacteria isolates on *Cucumis melo* sample collected from Zuba and Gwarimpa market Abuja Nigeria: The pathogenicity test of bacteria isolates on *Cucumis melo* sample collected from Zuba and Gwarimpa market is shown in Table 4. All the bacteria species namely; *Escherichia coli, Proteus mirabilis, Klebsiella Pneumoniae, Shigella* sp. and *Salmonella* sp. were positive to pathogenicity test except *Enterobacter aerogenes,* which shows negative pathogenicity test. The antibiotic susceptibility and resistant of bacteria isolates on *Cucumis melo* sample collected from Zuba and Gwarimpa markets is shown in Table 5. The results shows the different bacterial isolates from golden melon samples have various degrees of antibiotic resistance.

Table 1. Proximate analysis of *Cucumis melo* sample collected from Zuba and Gwarimpa market Abuja Nigeria

Sample location	Moisture (%)	Ash (%)	Fat (%)	Protein (%)	Carbohydrate (%)
Zuba	74.60 ± 1.93	1.02 ± 0.04	0.54 ± 0.02	0.56 ± 0.02	23.24 ± 1.94
Gwarimpa	78.60 ± 1.74	0.92 ± 0.03	0.59 ± 0.06	0.57 ± 0.01	19.10 ± 1.67

Table 2. Bacteria colony count of *Cucumis melo* sample collected from Zuba and Gwarimpa market Abuja Nigeria

Sample location	Colonies forming unit per milliliter (CFU/ml)
Zuba	$1.36 \pm 0.21 \times 10^{-3}$
Gwarimpa	$1.76 \pm 0.02 \times 10^{-3}$

Table 3. Biochemical Characteristic of bacteria the Isolates from golden melon samples collected from Zuba and Gwarimpa Market Abuja

Market	Gram reaction	Shape	Catalase	Indole	Citrate	Methyl red	Vogue Proskeur	Glucose	Maltose	Lactose	Sucrose	Probable Organism
Zuba	-	Rod	+	-	+	-	+	+	-	+	+	*Escherichia coli*
	-	Rod	+	-	+	+	-	+	-	-		*Proteus mirabilis*
	-	Rod	+	-	+	-	+	+	+	+	+	*Klebsiella Pneumoniae*
	-	Rod	+	-	+	-	+	+	-	+	+	*Enterobacter aerogenes*
	-	Rod	+	-	+	-	-	-	-	-	+	*Pseudomonas aeruginosa*
Gwarimpa	-	Rod	+	+	-	+	-	-	-	-	+	*Shigella sp.*
	-	Rod	+	-	-	+	-	+	+	-	-	*Salmonella sp.*
G2	-	Rod	+	-	+	-	+	-	-	-	+	*Pseudomonas aeruginosa*
G4	-	Rod	+	+	-	+	-	+	-	+	+	*Escherichia coli*
G5	-	Rod	+	-	+	-	+	+	+	+	+	*Klebsiella Pneumoniae*

Key: (+) = Positive (-) = Negative

Table 4. Pathogenicity Test on Fresh Golden Melon Samples

Inoculum Bacteria	Test sample after incubation
Escherichia coli	+
Proteus mirabilis	+
Klebsiella Pneumoniae	+
Enterobacter aerogenes	-
Pseudomonas aeruginosa	+
Shigella sp.	+
Salmonella sp.	+

Key: (+) = Positive (-) = Negative

Table 5. Antibiotic Susceptibility and Resistant Pattern of bacteria isolates from golden melon samples collected from Zuba and Gwarimpa Market Abuja

Isolate	SXT (30µ)	CH (30µg)	SP (10µg)	CPX (30µg)	AM (30µg)	AU (10µg)	CN (30µg)	PEF (30µg)	OFX (10µg)	SE (30µg)
Escherichia coli	-	-	+	+	-	-	-	+	+	-
Proteus mirabilis	-	+	+	+	+	-	+	+	+	+
Klebsiella Pneumoniae	-	+	+	+	-	+	+	+	+	-
Enterobacter aerogenes	-	+	+	+	-	-	+	+	+	-
Pseudomonas aeruginosa	-	-	+	+	-	-	-	+	+	-
Shigella sp.	-	+	+	+	+	-	+	+	+	+
Salmonella sp	-	+	+	+	+	-	+	+	+	+

Keys: SXT= Septrin, SP= Sparfloxacin, AU= Augmentin, CN= Gentamycin, PEF= Pefloxacin, OFX= Tarvid, CPX=Ciprofloxacin, CN=Gentamicin, SE=Streptomycin, CH= Chloranphenicol, AM= Amoxicillin

4. DISCUSSION

Fruits and vegetables provide substantial health benefits, along with basic nutrition. Several clinical, preclinical, and in vitro studies have shown that the consumption of vegetables and fruits protects against chronic diseases [11]. The proximate analysis of *Cucumis melo* samples collected from Zuba and Gwarimpa markets in Abuja, Nigeria, reveals interesting differences in their nutrient composition. It is noted that the *Cucumis melo* sample collected from Zuba Market had the highest percentage of ash and carbohydrate. Ash content in food represents the mineral content, and a higher ash content could indicate a higher mineral concentration in the fruit [12]. Carbohydrates are the main source of energy in our diet, and higher carbohydrate content in the Zuba market sample suggests it may be more filling and energizing. On the other hand, the Cucumis melo sample collected from Gwarimpa market had the highest moisture content, fat, and protein levels. Moisture content is important for juiciness and freshness of the fruit. The high moisture content observed in this study could make the *C. melo* highly susceptible to microbial attack [13]. High-fat content may provide a source of energy and essential fatty acids, while protein is important for tissue repair and growth [12]. The differences in the nutrient composition of the two samples could be influenced by various factors such as growing conditions, soil quality, irrigation practices, and post-harvest handling. It is also possible that different varieties of *Cucumis melo* were sold at the two markets, leading to variations in their nutrient profiles.

The presence of *Escherichia coli*, *Proteus mirabilis, Klebsiella Pneumoniae, Enterobacter aerogenes*, *Pseudomonas aeruginosa Shigella* sp. and *Salmonella* sp. in *Cucumis melo* samples collected from both Zuba and Gwarimpa markets raises concerns about the potential contamination of the fruits with pathogenic bacteria as some of this bacteria are of public health importance and has been reported to be associated with spoilage of fruits and vegetables [14]. For instance *Escherichia coli* is a common bacteria found in the intestines of humans and animals [15]. While most strains are harmless, some can cause foodborne illnesses. The presence of *E. coli* in the samples indicates a possible fecal contamination, likely due to poor sanitation practices during handling or storage [16] while *Proteus mirabilis* is a bacterium known to cause urinary tract infections and other infections in humans. The presence of this bacteria in the collected melon fruits is in indication of improper handling or storage conditions that may have led to contamination [14].

Furthermore, the presence of *Klebsiella Pneumoniae* in the melon samples is a matter of public health concern as this species has been reported to cause pneumonia, urinary tract infections, and other healthcare-associated infections [17]. Its presence in the samples is concerning as it indicates a potential risk of infections if the contaminated fruits are consumed. *Enterobacter aerogenes* have been reported to be commonly found in soil and water but can also cause infections in hospital settings. Its presence in the samples may indicate poor hygiene practices during harvesting, transportation, or storage of the fruits [14] while *Pseudomonas aeruginosa* is known to cause infections in wounds, lungs, and other parts of the body. Its presence in both samples suggests a potential contamination at some point in the supply chain.

The presence of *Shigella* sp. and *Salmonella* sp. in the Gwarimpa market samples is of particular concern as these bacteria species are known to cause severe foodborne illnesses such as dysentery and salmonellosis [18]. Proper hygiene practices, including thorough washing and sanitization of fruits, as well as maintaining clean storage and transportation conditions, are essential to prevent the growth and spread of these harmful bacteria.

The positive pathogenicity test results for *Escherichia coli, Proteus mirabilis, Klebsiella pneumoniae, Shigella* sp., and *Salmonella* sp. suggest that these bacterial species can cause disease in humans. This is concerning as contamination of food with pathogenic bacteria can lead to foodborne illnesses in individuals who consume the contaminated food [19]. Also, it is important to determine the antibiotic susceptibility profiles of the bacterial isolates to guide treatment options in case of infections as the susceptibility testing will help healthcare professionals choose the most effective antibiotics to treat infections caused by these bacteria [20]. Different antibiotics may be more or less effective against each bacterial species, so it is important to know the susceptibility profiles of each isolate [20]. The presence of antibiotic resistance in bacterial isolates is of great concern as it limits treatment options and can lead to treatment failure [21].

5. CONCLUSION

In this study, both samples of *Cucumis melo* appear to be nutritious and can be a valuable addition to a balanced diet. Consuming a variety of fruits and vegetables from different sources can help ensure a diverse intake of essential nutrients. The presence of *Escherichia coli*, *Proteus mirabilis, Klebsiella Pneumoniae, Enterobacter aerogenes*, *Pseudomonas aeruginosa Shigella* sp. and *Salmonella* sp. in the *Cucumis melo* samples highlights the importance of food safety and hygiene practices in the food supply chain to prevent the risk of foodborne illnesses. Monitoring antibiotic resistance patterns in bacterial isolates is crucial for public health interventions to prevent the spread of resistant bacteria. The identification of antibiotic-resistant strains in the *Cucumis melo* samples collected from Zuba and Gwarimpa market highlights the importance of proper food safety practices to prevent the spread of these resistant bacteria. Vendors, market operators, and consumers must be aware of potential contamination sources and take necessary precautions to ensure the safety of the food products being consumed.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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