

ORIGINAL ARTICLE

Most Commercial Ready-to-Eat Foods in and Around Universities in Eastern Uganda Contain Pathogenic Drug-Resistant Bacteria: A Call to Action

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ABSTRACT

Uganda faces an escalation of drug-resistant foodborne bacterial diseases. Academic communities such as universities are among the most affected but the burden is under researched. Hence, this study determined the prevalence of drug-resistant bacteria and the associated factors in food, in and around Islamic University in Uganda, to inform and support the way forward. Conventional culture, microscopy, biochemical characterization and disk diffusion methods were used to identify drug resistant, medically important bacterial pathogens from a random sample of 79 ready-to-eat commercial foods. Focus Group Discussions were used to examine the factors enabling the presence of these pathogens in food, among a random sample of 67 food vendors. Data were analyzed using thematic analysis and descriptive statistics with STATA version 15, and World Health Organization guidelines. *Escherichia coli* (30.1%, n=47) and *Staphylococcus aureus* (20.5%, n=32) were the most prevalent, while *Shigella spp.* (1.1%, n=2) was the least. The Total Viable Load of cooked foods (0.91×10^1 CFU/g) and deep-fried fast foods (1.52×10^1 CFU/g) met safety limits, but fresh fruit (3.44×10^4 CFU/g) and vegetable salads (3.58×10^4 CFU/g) exceeded WHO standards. Total Coliform Counts in all samples were unsafe, with fresh fruit (3.30×10^4 CFU/g) and vegetable salads (1.15×10^4 CFU/g) showing the highest contamination. Many isolates were drug-resistant, including *E. coli* to Cefoxitin (48%) and Imipenem (18%), and *S. aureus* to Nalidixic acid (38%) and Gentamycin (26%). Contamination was mainly linked to food handlers' practices (23.5%, n=8) and sociodemographic factors (17.6%, n=6). Stricter hygiene measures and better food safety monitoring are urgently needed.

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1. Introduction

Small scale food serving outlets are frequently utilized in most developing countries because they are affordable, easy to access and as well serve as a vital source of income (Rodrigues Fortes et al., 2020). However, foods in these outlets mostly don't meet proper sanitary standards and can lead to food borne ailments with consequences of morbidity, mortality and hindrance of trade and development (Food and Agriculture Organization/World Health Organization (FAO/WHO), 2016). Food borne ailments are ranked among the greatest causes of human death and suffering (Lee & Yoon, 2021). The risk is more severe in Sub-Saharan Africa, where mortality rate due to diarrheal food borne bacteria is estimated at 700,000, attributed to unsuitable food handling practices especially; poor hygiene and inadequacy of food safety-knowledge among food handlers in commercial food serving units (Christiana Cudjoe et al., 2022). Transmission of enteric bacterial pathogens to humans may occur via fecal-contaminated food, water, cutlery and fingers of food handlers (Walusansa, Asiimwe, Kafeero, et al., 2021). Bacterial strains of *Escherichia coli*, *Salmonella spp*, *Shigella spp* and *Campylobacter spp* are the most commonly reported causes of gastrointestinal disease in Sub-Saharan Africa (Hlashwayo et al., 2020). All these bacteria have been implicated in causing foodborne infections (Zong Minko et al., 2024). Emergence of drug resistance in these pathogens aggravates the burden as it makes the infections very expensive and difficult to treat (Walusansa, Asiimwe, et al., 2022).

In most parts of rural Uganda, reliable statistics about food borne infections, and the antibiotic sensitivity profiles of the pathogens involved, remain inadequate, due to poor or absence of surveillance systems; this limits the adoption of risk-based food safety policies. Though outbreaks of food borne bacterial illnesses, like *Salmonellosis* and *Cholera*, remain a serious problem in Mbale, one of the districts in Eastern Uganda (Manetu & Karanja, 2021), no recent studies have been conducted to examine the bacterial strains circulating in the food establishments in this area. In addition, surveillance data on antibiotic resistance remains limited in the face of reduced therapeutic options in food borne bacterial illnesses. This is attributed to limited microbiology laboratory capacity required for diagnosis, surveillance, and drug sensitivity testing which are vital in the effective treatment and prevention of drug-resistant infections. Food outlets in and around universities are potential sources of food borne bacterial pathogens (Abebe et al., 2020; Asfaw et al., 2022).

In the recent three decades, Nkoma area of Mbale city has faced a remarkable population increase following the introduction of the Islamic University in Uganda (IUIU) which is currently populated by over 10, 000 students and staff (Islamic University in Uganda (IUIU), 2023). The IUIU community and hundreds of resident youths depend mostly on food prepared in commercial

food outlets in the area. These food outlets are operated in hygienically challenged environment and by mostly illiterate personnel with untested competence and skills pertaining food safety. This provides a suitable platform for microbial contamination of food with a potential transfer risk of pathogenic contaminants to consumers. Indeed, food borne infections are prevalent in this area; the IUIU health center, a major place for seeking healthcare among the University students, staff and the surrounding communities registered over 60 cases of bacterial infections related to food contamination in the month of November 2023 alone (Islamic University in Uganda (IUIU), 2023). To elucidate the unending prevalence of food borne bacterial infections in the study area, it has become necessary to conduct a systematic survey of the species and drug susceptibility patterns of bacterial pathogens associated with food poisoning potentially circulating in the commercial food outlets. The purpose of this study was to examine the occurrence of common bacterial strains in the food sold in and around IUIU main campus in Mbale city, drug resistance among the bacterial contaminants, and the risk factors for contamination of food with bacterial pathogens in the selected food outlets. The information that was generated is vital in guiding the design of risk-based food safety interventions to address the burden of foodborne bacterial ailments in the IUIU and the general community at large. The antibiotic susceptibility data generated will guide antibiotic resistance stewardship.

2. Materials and Methods

2.1 Study design

A cross-sectional study was conducted between July 2021 and August 2022 among commercial food serving establishments within the selected IUIU and in the neighboring communities of this university.

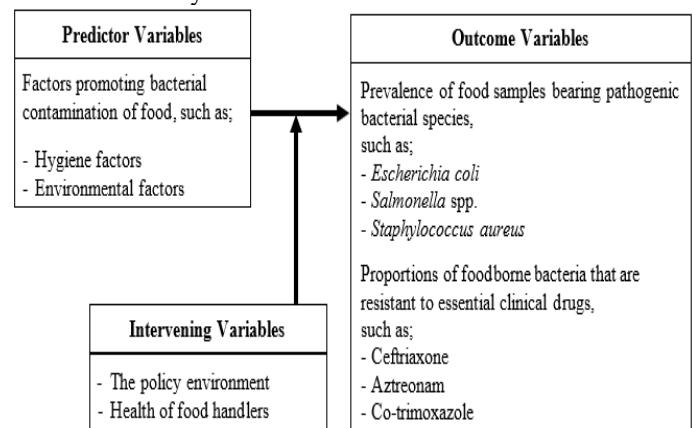


Fig. 1: Conceptual framework demonstrating how various factors contribute to drug resistant bacterial contamination of food in and around universities in Eastern Uganda.

2.2 Study sites and settings

The study was conducted in Nkoma Ward (Northern Borough division of Mbale Municipality), in and around the Islamic University in Uganda Mbale campus. Nkoma Ward is part of Mbale, the largest city in Eastern Uganda which is located in Mbale district (Fig. 2). The coordinates of the district are: 1.1190° N, 34.1275° E, 1.1190° N, 34.2165° E, 1.0330° N, 34.1275° E, and 1.0330° N, 34.2165° E (Warsame et al., 2021). The food Samples were randomly collected from all the 15 food serving outlets (Restaurants, Canteens, Footstalls, and Kiosks), within UIU and in the neighborhood.

Samples were then transported to the Medical Microbiology Laboratory at Makerere University College of Health Sciences (MakCHS) for microbiological analysis.

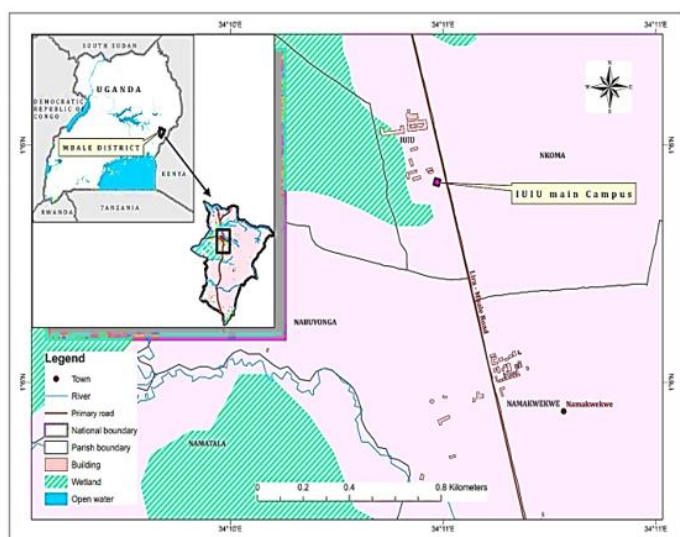


Fig.2: The map of Uganda showing the study area for this research

2.3 Study population

The targeted population includes all consenting individuals working in commercial food serving outlets in and around the Islamic University in Uganda (Mbale campus and Kampala campus). These individuals are those that make direct contact with food, e.g. via cooking, serving and preparation. In addition, selected raw ready to eat foods available in the food outlets will also be studied.

2.4 Inclusion and exclusion criteria

All consenting individuals directly engaged in food preparation and/or serving in these outlets were included in the study, and selected ready-to-serve foods in the selected food outlets were collected using aseptic procedures. Food handlers who did not consent to participate in the study, and those who were found absent from the respective food serving facilities were not

included in the study. All types of already cooked as well as the uncooked foods were selected as samples in the study.

2.5 Sample size and sampling technique

The sample size will be determined using the Leslie Kish formula (Singh & Masuku, 2014)

$$n = \frac{Z^2_{\alpha/2} p(1-p)}{d^2} \quad \text{- eqn. 1}$$

Where;

n - Sample size required, Z -Standard normal value corresponding to the set confidence level, e.g. if the confidence level is 95%, then Z is 1.96. *P*-estimated proportion of the population positive for one or more potential food borne bacterial contaminants. *d*-Tolerable sampling error (precision) usually 5% except when *p* < 0.1 then 3% is used. Using the prevalence of 49.6% (Tsegaye, *et al*, 2015), *P*= 0.11; *Z*_{α/2} = 1.96 (CI=95%) and *d* = 0.05.

Substituting into the sample size formula;

$$N = [(1.96)^2 \times 0.496 \times (1-0.496)] \div (0.05)^2 = [3.8416 \times 0.11 \times 0.89] \div 0.0025. n = 150.$$

Since the available population of food handlers is estimated at 120, a correction formula was used to adjust the sample size to the population size (*N*) as follows;

$$S = \frac{n}{1 + \frac{n}{N}} = \frac{150}{1 + \frac{150}{120}} \approx 67 \quad \text{-eqn. 2}$$

Hence the final sample size of participants was 67 Food handlers.

In addition, 79 food samples were collected. Out of the 79 ready-to-eat food samples collected, 39 were in the processed form (20 were cooked traditional food stuffs such as; Banana, Irish potatoes, Cassava, Posho, Rice, Beans, Meat, and ground nuts, and 19 were fried fast foods like Chips, Chapatis, Liver, Fish, and Chicken), while 40 were in the unprocessed / raw form (20 were fruit salads such as Avocado, Mangoes, Pawpaws, and 20 were vegetable salads like Cabbages, Collards greens, *Amaranthus* spp. A census sampling technique was used to include the food outlets into the study. A lottery sampling method was used to recruit the food handlers into the study. During the sampling each of the food handlers in a food serving outlet was assigned a unique number, the numbers were then thoroughly mixed by shaking in jar; without looking, the research assistant selected a number and the participant that was assigned that number was then included in the sample. This was repeated until the sample size required was obtained. The prospective participants were sensitized about the study, with exceptional emphasis on the objectives of the study, its benefits, potential risks and the available means to avert the risks. The participants were then taken through a consenting process.

2.6 Data collection instruments and procedures

Participants who gave informed consent were subjected to Focus Group Discussions (FGDs) using pre-validated FGD guides. In addition, the hygiene practices of the participants, and the sanitary qualities of the facilities in each food outlet were examined using an observation survey. The collection of the selected foods was done using sterile gloves, by handpicking of these samples; these samples were sliced into small pieces using sterile blades and introduced into labeled sterile specimen containers. The samples were then transported in cool boxes (immersed in ice to maintain potency of any contaminant microbes in food), to the Medical Microbiology Laboratory at Makerere University College of Health Sciences (MakCHS) for analysis within 24 hours since collection.

Culture and identification of selected bacteria from the food samples

Processing of the food samples was done as follows: on arrival at the laboratory, 5mls of sterile normal saline was added to each of the specimen containers, this was followed by vortexing of the container, to wash the surface of the foods and hence provide a 5ml solution of saline containing material washed from the surface of the foods, this solution also provided a 10^0 solution for the subsequent dilution, further dilutions were done to a 10^{-5} via 10^{-4} , 10^{-3} , 10^{-2} , 10^{-1} at each step, 0.5mls of the solution from the 10^0 were added to a container containing 4.5mls of sterile normal saline, this was followed by vortexing to mix uniformly, the 10^{-5} dilution was obtained by repeating the procedure, from each of the last dilution, 0.5mls was then inoculated onto MacConkey agar, Salmonella-Shigella agar, Violet Red Bile Agar, Mannitol Salt Agar, *Bacillus cereus* agar, and Kings agar. Inoculation of the culture media was done using spread plating with sterile beads. This was then followed by incubation of the culture plates at 37°C in ambient air for 24hrs. After 24 hours of incubation, the bacterial isolates were identified using gram-staining, microscopy, and biochemical methods (Walusansa, Nakavuma, et al., 2022).

Antimicrobial susceptibility testing of selected bacteria isolated from the food samples

Following identification of the isolates, antimicrobial sensitivity profiles of the isolates were then be determined using Disc diffusion-based methods. The outcome variables were presented as, susceptible (S); meaning that the bacteria are sensitive to the drug/or that the drug effectively kills the bacteria, intermediate (I); the drug kills some of the colonies but allows others to grow, meaning that resistance is evolving, resistant (R); meaning that the drug has completely lost its potential to kill or inhibit microbial growth. The antibiotics tested included; Ampicillin, Chloramphenicol, Nalidixic acid, Tetracycline, Ciprofloxacin, Amikacin, Amoxicillin, Ceftriaxone, Cefoxitin, Kanamycin, Streptomycin, Azithromycin Trimethoprim/ Sulfamethoxazole

and Gentamicin. The resistance phenotypes were interpreted according to the guidelines of Clinical Laboratory Standards Institute (Aruhomukama, 2020; Clinical and Laboratory Standards Institute (CLSI), 2020).

2.7 Data analysis plan

The data was entered into a computer using Microsoft Excel and analyzed using STATA versions 12. Descriptive statistics and thematic synthesis were used to analyze the data. Outcome variables were presence or absence of the target bacteria in food samples. The outcome variable were presented as the number of bacterial species isolated, and as a proportion of contaminated samples out of the total food samples tested. Data was entered into a computer using Microsoft Excel and analyzed using STATA version 12. The phenotypic resistance was presented as the percentage of the total isolates tested. Data was analyzed using descriptive statistics such as counts and frequencies.

3. Results and Discussion

3.1 Socio demographic characteristics of respondents.

Majority (n=46, 68.7%) of the respondents were female, hence males were 31.3% (n=29). Most of the participants were in the middle age category (n= 39, 58.2%), followed by the youths (n=25, 37.3%), while only three (4.5%) were elderly. Most of the participants (n=36, 53.7%) had attended secondary education, while very few (3, 4.5%) had attained tertiary education. The majority of the respondents had practiced commercial food supply for a duration of 5 to 15 years (n=47, 70.1%). Most participants earned a net daily profit of UGX. 7,220 (\$1.9) from the sale of food (Table. 1).

The socio-demographic profile of respondents highlights a predominantly female sample, emphasizing the critical role women play in food supply chains (Csordas et al., 2022). Most participants were middle-aged, while a smaller proportion were elderly, aligning with findings that aging populations often withdraw from commercial food supply (Jia et al., 2023). Educational attainment was generally at the primary level, reflecting Uganda's broader trends among informal business actors whose access to tertiary education remains limited (Uganda Bureau of Statistics (UBOS), 2016). Many respondents demonstrated significant experience in commercial food supply, underscoring their established presence in the market. However, low net daily profits place the majority of the respondents below Uganda's poverty level, pointing to economic challenges, revealing vulnerabilities to price volatility and market instability (Walusansa, Asiimwe, Ssenku, et al., 2021; World Bank Group, 2020).

Table 1. Socio-demographic characteristics of commercial food handlers in and around Islamic University in Uganda (N=67)

Variable		Frequency, n (%)
Gender	Male	21 (31.3)
	Female	46 (68.7)
Age (years)	18-24 (Youths)	25 (37.3)
	25-63 (Middle aged)	39 (58.2)
	≥ 64 (Elderly)	3 (4.5)
Nationality	Ugandan	66 (98.5)
	Non-Ugandan	1 (1.5)
Marital status	Married	43 (64.2)
	Single	24 (35.8)
Education	None	8 (11.9)
	Primary	36 (53.7)
	Secondary	20 (29.9)
	Tertiary	3 (4.5)
Years of experience in the food sector	5≤15	47 (70.1)
	16≤20	17 (25.4)
	>20	3 (4.5)
Type of food selling establishment	Restaurants	23 (34.3)
	Canteens	5 (7.5)
	Footstalls and Kiosks	39 (58.2)
Estimated daily net income from food sales	< UGX. 7,220 (\$1.9) (Poverty line)	58 (86.6%)
	≥ 7,220 (1.9)	9 (13.4%)

Key; UGX: Uganda Shillings, \$: United States Dollar

3.2 Characteristics of the food samples from which bacterial contaminants were isolated

Out of the 79 ready-to-eat food samples investigated in this research, 20 were cooked traditional food stuffs such as (Banana, Irish potatoes, Cassava, Posho, Rice, Beans, Meat, and ground nuts), 20 were fruit salads (Avocado, Mangoes, Pawpaws), 20 were vegetable salads (Cabbages, Collards greens, Amaranthus spp), while 19 were fried fast foods (Chips, Chapatis, Liver, Fish, and Chicken). The food samples were availed on market in different packaging materials namely; polyethene (n=32; 40.5%), Melamine plates (n=18, 22.8%), papers (n=14; 17.7%), buckets (9; 11.4%), baskets (3; 3.8%), and bare (unwrapped) (3; 3.8%). Forty-one (n=41, 51.9%) samples were visibly clean, while 38 (48.1%) appeared dusty. Fifty two (n=52, 65.8%) of the food samples were in absolute disconformity to the food safety guidelines of the World Health Organization (WHO, 2023).

This study therefore reveals a heterogeneous composition of commercial foodstuffs packaged in notably diverse packaging materials in the study community. A substantial fraction of the food packages exhibited surface dust accumulation, indicating potential exposure to environmental contaminants. These findings align with previous studies that have reported inadequate food hygiene practices among food vendors, contributing to microbial contamination (Azanaw et al., 2022; Kariuki, 2018). A significant number of samples did not comply with World Health Organization food safety guidelines (WHO, 2023), similar to findings in other studies where informal food markets and other ready-to-eat products have been identified as high-risk environments for foodborne illnesses (Kwiri et al., 2014; Walusansa, Asiimwe, et al., 2022; Walusansa, Asiimwe, Kafeero, et al., 2021; Walusansa, Nakavuma, et al., 2022). These findings suggest a need for targeted interventions, such as vendor training, stricter food safety regulations, and improved infrastructure for food handling. In addition, strengthening enforcement mechanisms and promoting consumer awareness could significantly reduce contamination risks and enhance food

safety thereby addressing foodborne illnesses (Naguib et al., 2021).

3.3 Bacterial contamination of food vended sold in and around IUIU

The bacterial contamination of foods was evaluated using three microbiological parameters namely; Total Coliform Counts of bacteria (TCC), prevalence of bacterial pathogens, and drug resistance profiles of the bacteria isolated from the food samples. The results are presented below.

Total Viable Loads (TVL) and Total Coliform Counts (TCC) of bacteria in food samples

Table. 2, presents the TVL and TCC analyses of food samples sold in and around Islamic University in Uganda. The results indicate that the mean TVL for cooked traditional foods such as Bananas and posho (0.91×10^1 CFU/g), was marginally higher than the mean TVL for deep-fried fast foods such as Irish potato chips and Chapatis (1.52×10^1 CFU/g). These TVL values were generally low and adhered to the international permissible levels of Total bacterial loads for processed foods set by World Health Organization (WHO, 2023). However, the mean TVL values for fresh fruit salads and fresh vegetable salads were higher than the internationally acceptable standards. All the mean TCC values were unsafe for human consumption with regards to the internationally permissible Total Coliform Counts (0.0×10^1 CFU/g), stipulated by WHO.

Table. 2: Average Total Viable Loads and Total Coliform Counts of bacteria in foods sold in and around Islamic University in Uganda

Type of food	TVL (CFU/g)	TCC (CFU/g)
Cooked traditional food stuffs	0.91×10^1	0.6×10^1
Deep-fried fast foods	1.52×10^1	0.38×10^1
Fresh Fruit salads	3.44×10^4	3.30×10^4
Fresh Vegetable salads	3.58×10^4	1.15×10^4

TVL: Total Viable Bacterial Counts, TCC: Total Coliform Count, CFU/g: Colony Forming Units per gram

The TVL and TCC analysis of food samples from in and around the Islamic University in Uganda has highlighted significant insights into food safety and hygiene practices. In this, the cooked traditional foods such as bananas and posho had slightly higher microbial loads compared to deep-fried fast foods like potato chips and chapatis, likely due to the high temperatures involved in frying, which reduce bacterial contamination (WHO, 2023b). The WHO has constituted a list of food safety guidelines including; cleaning the packages, washing hands, separating raw and cooked foods, cooking to safe temperatures, storing food properly, and compliance with permissible microbial loads (World Health Organization (WHO), 2023b). In the current study, while the TVLs of processed foods complied with those WHO standards, fresh fruit and vegetable salads exceeded permissible levels, suggesting inadequate washing, cross-contamination from handlers, or environmental exposure during preparation

and storage (Kpekurah, 2023). The Total Coliform Count (TCC) for all food samples was found to be above safe limits set by (World Health Organization, (2023b), indicating fecal contamination and unsanitary conditions during preparation or handling. These findings call for more stringent hygiene measures, including improved food handling practices, enhanced sanitation infrastructure, and regular microbial assessments to ensure food safety.

Prevalence of bacterial pathogens in the food samples

A total of 156 bacterial isolates were obtained from the 79 food samples. The cultural and cellular characteristics of these bacteria are shown in Fig. 3, and table 3. The most prevalent bacterial species was *Escherichia coli* (n=47, 30.1%), isolated from 39 samples, and *Staphylococcus aureus* (n=32, 20.5%), in 24 samples. The least prevalent bacteria were *Shigella* spp (n=2, 1.1%), which was isolated from two samples (Table 4).

Table 3: Cellular morphology, gram staining properties, and biochemical characteristics of representative bacteria isolated from commercial foods eaten by communities in and around Islamic University in Uganda

Gram negative bacterial isolates												
Isolate ID. No	Bacterial species isolated	Culture media (Agar)	Gram staining, Shape	Biochemical characteristics								
				TSI				Citrate	Urease	Indole	MR	VP
				B	S	G	H ₂ S					
X16	<i>Salmonella</i> spp	XLD	G-, Rods	A	K	-	-	+	-	-	-	-
X08	<i>K. pneumoniae</i>	MacConkey	G-, Rods	A	A	+	-	+	+	-	-	+
X07	<i>P. aeruginosa</i>	Kings' media	G-, Rods	NC	K	-	-	+	+	-	-	+
X13	<i>K. pneumoniae</i>	MacConkey	G-, Rods	A	A	+	-	+	+	-	-	+
X04	<i>B. cereus</i>	<i>B. cereus</i> agar	G-, Rods	A	K	+	+	-	+	+	+	-
X01	<i>P. aeruginosa</i>	Kings' media	G-, Rods	NC	K	-	-	+	+	-	-	+
X11	<i>Morganella morganii</i>	MacConkey	G-, Rods	K	A	-	-	-	-	+	+	-
X05	<i>K. pneumoniae</i>	MacConkey	G-, Rods	A	A	+	-	+	+	-	-	+
X12	<i>E. coli</i>	MacConkey	G-, Rods	A	A	-	-	-	-	+	+	-
Gram positive bacterial isolates												
Isolate ID. No	Species	Culture media	Gram staining, Shape	Biochemical characteristics				Mannitol fermentation	Coagulase	Catalase	DNase	
B20	<i>Staphylococcus aureus</i>	Mannitol Salt	G+ cocci arranged in grape-like clusters	+				+	+	+	+	
B21	<i>Staphylococcus aureus</i>	Mannitol Salt	G+ cocci arranged in grape-like clusters	+				+	+	+	+	
B27	<i>Staphylococcus aureus</i>	Mannitol Salt	G+ cocci arranged in grape-like clusters	+				+	+	+	+	

Key: ID. No; Identification Number, VP; Voges-Proskauer, MR; Methyl Red, H₂S; Hydrogen sulfide, B; Butt, S; Slant, G; Gas, K; Red/Alkaline, A: Yellow/Acid, NC; No colour change, TSI; Triple Sugar Iron, G-; Gram-negative, G+; gram-positive, +; Positive, -: Negative, *E. coli*; *Escherichia coli*, *P. aeruginosa*; *Pseudomonas aeruginosa*, *K. pneumoniae*; *Klebsiella pneumoniae*, *B. cereus*: *Bacillus cereus*, spp; Species, XLD: Xylose-Lysine-Deoxycholate

Table 4: Prevalence of bacteria in commercial foods eaten by communities in and around Islamic University in Uganda, Eastern Uganda

Bacterial species isolated	Number of bacterial Isolates obtained (N=156)	Percentage of bacterial isolates obtained (%)	Number of contaminated samples (N=79)	Percentage of contaminated samples (%)
<i>Escherichia coli</i>	47	30.1	39	49.4
<i>Staphylococcus aureus</i>	32	20.5	24	30.4
<i>Klebsiella pneumoniae</i>	16	10.3	10	12.7
<i>Pseudomonas</i> spp	16	10.3	12	15.2
<i>Morganella morganii</i>	14	9.0	10	12.7
<i>Pseudomonas aeruginosa</i>	12	7.7	9	11.4
<i>Bacillus cereus</i>	09	5.8	6	7.6
<i>Klebsiella oxytoca</i>	04	2.6	3	3.8
<i>Salmonella</i> spp	04	2.6	2	2.5
<i>Shigella</i> spp.	02	1.1	2	2.5

The fact that this study identified *Escherichia coli* as the most prevalent bacterial species in commercial foods, signals potential fecal contamination resulting from poor food handling practices (Mensah et al., 2002). *Staphylococcus aureus* was also frequently detected, consistent with findings from previous studies that document its common occurrence in ready-to-eat food (Mesbah et al., 2021). The presence of *E. coli* in nearly half of the contaminated samples poses significant public health risks, as certain strains such as Enterohaemorrhagic *E. coli* can cause severe foodborne illnesses (Walusansa et al., 2020). WHO guidelines emphasize that *E. coli* should not be present in ready-to-eat foods, underscoring the importance of stringent hygiene practices (World Health Organization (WHO), 2023b).

Other bacterial contaminants such as *Klebsiella pneumoniae*, *Pseudomonas spp.*, and *Morganella morganii* were also notable, with *K. pneumoniae* raising concerns due to its antibiotic resistance (Lupo, 2023). *Pseudomonas spp.* and *Pseudomonas aeruginosa* present spoilage risks and potential for foodborne illnesses (Bloomfield et al., 2024). Although *Shigella spp.* and *Salmonella spp.* were less common, their presence remains alarming due to their association with severe gastrointestinal infections (Tilahun et al., 2025). The bacterial species isolated in the current study were present both in both the processes (cooked or fried), as well as the unprocessed food samples. The observed contamination in the cooked foods would potentially mean that these foods are not thoroughly heated during cooking hence some pathogenic contaminants survive the heat, or that the food is served after it has cooled to low temperature that tolerate cross contamination from the food handlers, the environment, and utensils. Previous studies in Uganda have reported higher *Salmonella spp.* contamination rates, particularly in foods of animal origin (Kakooza et al., 2021). The detection of these bacteria indicates non-compliance with international food safety standards set by WHO and FAO, which advocate for zero tolerance to pathogenic bacteria in consumable food products (Food and Agriculture Organization/World Health Organization (FAO/WHO), 2016). These findings highlight the critical need for improved hygiene practices, better food storage, and stricter regulatory enforcement to address foodborne illnesses in such communities.

3.4 Antimicrobial resistance profiles of some bacteria isolated from the food samples

Two species of the most prevalent bacterial contaminants (one gram negative and one gram positive), were subjected to drug susceptibility. The inhibition zones from these experiments are shown in Fig. 4.

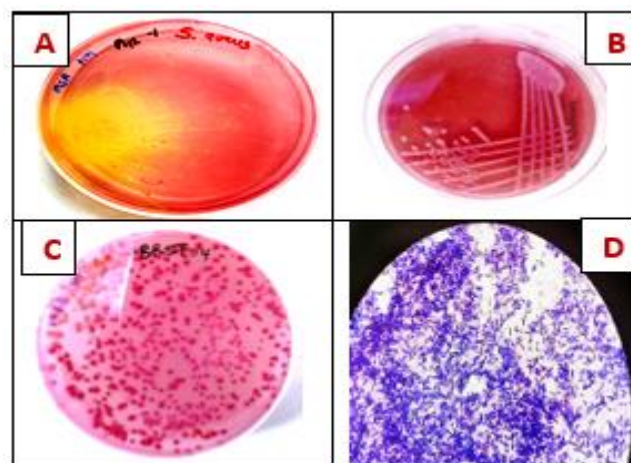


Fig 3: Colony and cellular characteristics of bacteria isolated from foods sold in and around Islamic university in Uganda. A: Golden yellow colonies of *Staphylococcus aureus* on Mannitol Salt Agar. B: Mucoid colonies of *Klebsiella pneumoniae* on MacConkey agar. C: Deep pink colonies of coliform bacteria on Violet Red Bile Agar. D: Gram-positive bacilli of *Bacillus spp.* observed in a light microscope.

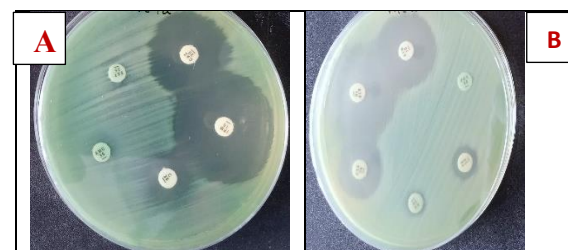


Fig. 4: Inhibition zones against selected conventional drugs on Mueller Hinton Agar for (A) *Escherichia coli* and (B) *Staphylococcus aureus* isolated from commercial foods in and around Islamic University in Uganda.

Though the majority of the bacterial isolates tested were susceptible to the respective clinical drugs, there was generally high resistance to high powered drugs such as Carbapenems and Cephalosporins. The 18% of the *Escherichia coli* isolates were resistant to Imipenem antibiotics. Also, 18% were resistant to Ertapenem, while 48% were resistant to Cefoxitin and 27% to Co-trimoxazole. The least resistance was evoked against Ceftazidime which is a 3rd generation cephalosporin. Similarly for *Staphylococcus aureus*, there was generally high resistance to critical drugs such as Nalidixic acid (38%), Gentamycin (26%), and Ertapenem (26%). The results are shown in Fig. 5.

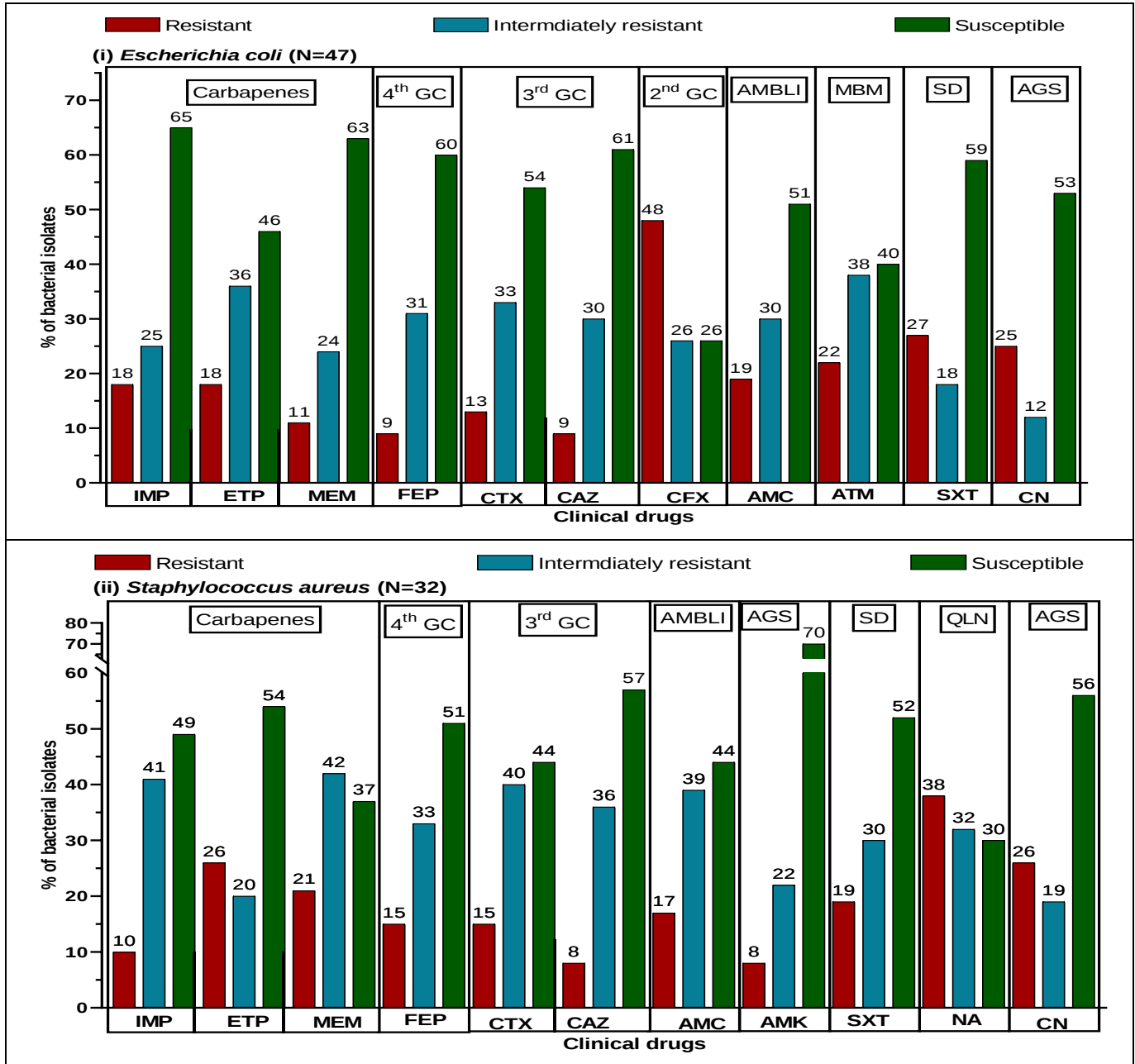


Fig. 5: Drug susceptibility profiles of *Escherichia coli* and *Staphylococcus aureus* isolated from food sold to communities in and around Islamic University in Uganda. IMP: Imipenem, ETP: Ertapenem, MEM: Meropenem, FEP: Cefepime, CTX: Ceftriaxone, CAZ: Ceftazidime, CFX: Cefoxitin, AMC: Amoxicillin+Clavulanic acid, ATM: Aztreonam, SXT: Co-trimoxazole, CN: Gentamycin, MBM: Monobactams, AGS: Aminoglycosides, SD: Sulphur Drugs, AMBLI: Amoxicillin+ β -Lactamase inhibitor, 2nd GC: Second Generation Cephalosporin, 3rd GC: Third Generation Cephalosporin, 4th GC: Fourth Generation Cephalosporin.

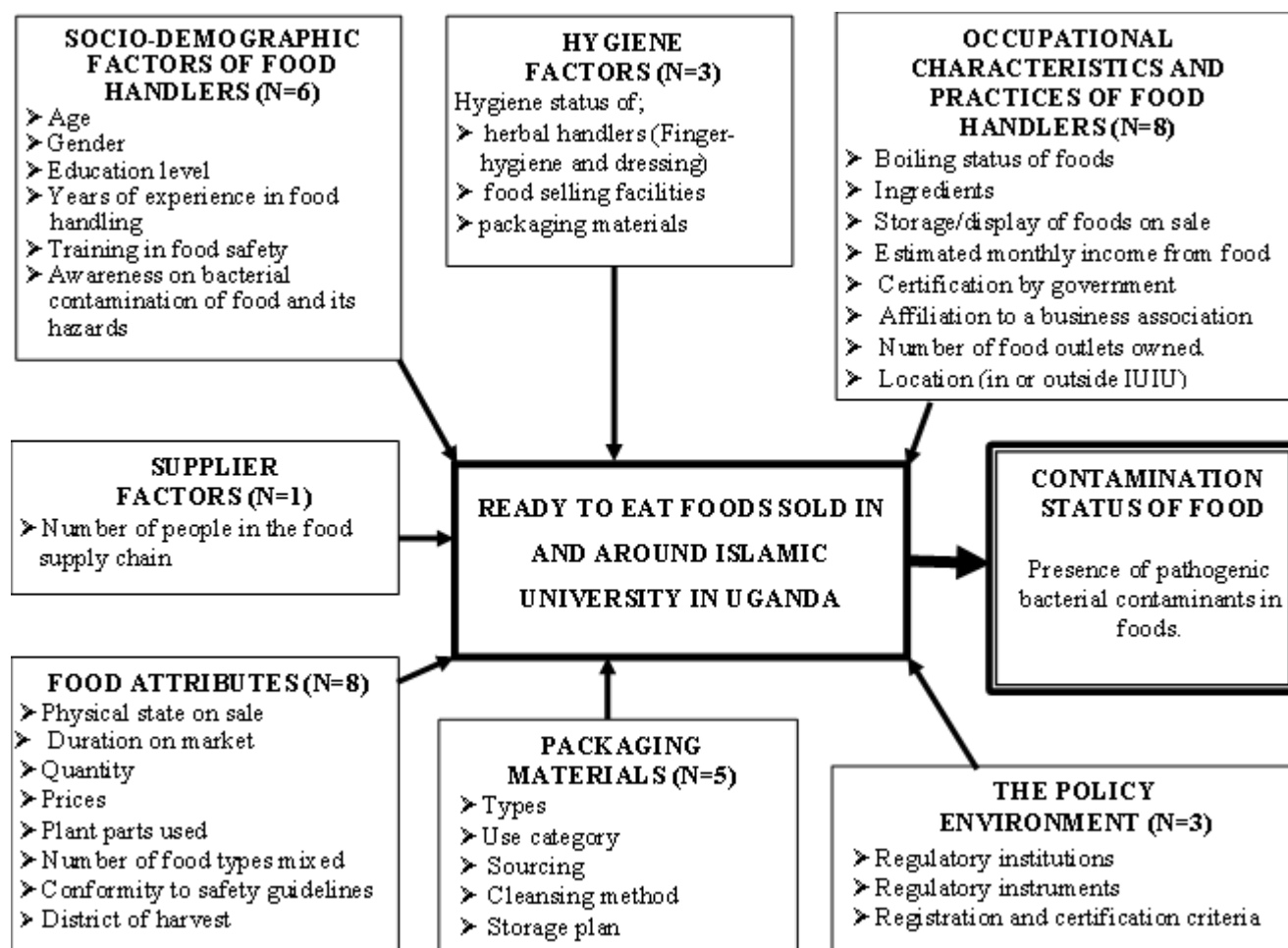


Fig. 6: Factors underlying the bacterial contamination of foods sold in and out of Islamic University in Uganda. The factors were conceptualized into seven themes; Socio-demographic factors, environmental factors, hygiene factors, policy factors, and occupational characteristics and practices of food handlers, among others.

This study underscores the alarming prevalence of antimicrobial resistance (AMR) among bacterial isolates, particularly against highly powered antibiotics like Carbapenems and Cephalosporins. *Escherichia coli* demonstrated resistance to multiple antibiotics, including last-resort options such as Imipenem and Ertapenem, consistent with prior findings in Sub-Saharan Africa, where unregulated antibiotic use has fueled rising resistance (Ventola, 2015). Similarly, *Staphylococcus aureus* exhibited high resistance to critical antibiotics, including Nalidixic acid and Gentamycin, mirroring global trends in methicillin-resistant *S. aureus* (MRSA) infections (World Health Organization (WHO), 2018). Resistance to aminoglycosides further suggests adaptive mechanisms that diminish the effectiveness of widely-used treatments (Saroj, 2022). Both the WHO and CDC emphasize the need for routine surveillance and antibiotic stewardship programs to address this escalating challenge (Centers of Disease Control and Prevention (CDC), 2029; World Health Organization (WHO), 2019).

3.5 Factors enabling bacterial contamination of the food vended in and around Islamic university in Uganda

The focus group discussions revealed a total of 34 factors enabling the bacterial contamination of ready-to-eat foods sold in and around Islamic University in Uganda. These factors conceptualized into seven themes (Fig. 6). The majority of the contamination enablers were occupational characteristics and practices of food handlers, and characteristics of the food stuffs (N= 8, 23.5% each). These were followed by the sociodemographic characteristics of the food handlers (N=6, 17.6%), while the least were supplier factors (N=1, 2.9%). Food contamination in this study was largely influenced by the occupational characteristics and handling practices of food handlers, along with the intrinsic properties of the food itself. These results align with prior research that identifies improper handling, poor hygiene, and environmental exposure as key contributors to microbial contamination in food establishments (Kamboj et al., 2020; Manning et al., 2019). Sociodemographic factors, such as the education and training levels of food

handlers, also played a notable role, corroborating the findings of other studies that link food safety knowledge to contamination risks (Da Vitória et al., 2021). Supplier-related factors had minimal influence, supporting evidence that contamination is more commonly introduced during food preparation and handling than at the supply stage (Lues & Van Tonder, 2007). Globally, the WHO emphasizes the critical need for robust food safety protocols, comprehensive training, and consistent monitoring to reduce contamination risks across diverse settings (World Health Organization (WHO), 2023b). Effective strategies should prioritize enhanced training for food handlers, stricter enforcement of hygiene regulations, and better oversight of supply chains to improve overall food safety standards.

Conclusion

This study highlights serious food safety and hygiene challenges in the food sold around the Islamic University in Uganda. A significant proportion of the tested food samples failed to meet World Health Organization safety standards, with many showing microbial contamination and unsafe total coliform counts, particularly in fresh fruit and vegetable salads. The detection of harmful pathogens such as *Escherichia coli* and *Staphylococcus aureus*, coupled with notable antimicrobial resistance, emphasizes the pressing need for improved hygiene practices in food handling. These findings reveal the substantial public health risks associated with inadequate food safety measures, particularly in informal food markets. To address these concerns, the implementation of strict food safety protocols is essential. This includes comprehensive training for food vendors on proper hygiene practices, improved sanitation infrastructure, and routine microbial assessments of food samples. Enhancing food packaging, minimizing environmental exposure, and ensuring adherence to WHO food safety guidelines could considerably lower the risk of foodborne illnesses. Additionally, stronger regulatory enforcement and increased consumer awareness are crucial steps in reducing contamination risks and protecting public health.

Ethical considerations

The study obtained approval from the institutional review board of the Islamic University in Uganda. Permission to conduct the study was also sought from relevant authorities including the village leaders. The study was conducted while following the national guidelines for the conduct of research in the post COVID-19 era designed by the Uganda National Council for Science and Technology (UNCST), and the Ministry of health of the republic of Uganda (Uganda National Council for Science and Technology (UNCST), 2020). All prospective respondents were subjected to written, voluntary informed consent procedures before participation. Respondents' identifiers were recorded in form of codes instead of names for purposes of anonymity, and the information was kept confidential during and after data analysis.

Data Availability statement

The data used in this study is available upon request from the author.

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Conflict of interests

Authors have declared that no competing interests exist.

Authors' Contributions

Abdul Walusansa (AW), and Shaban A. Okurut (SAO) conceived the research idea, and were involved in field data collection and manuscript writing. Jamilu. E. Ssenku (JES) and Ali Kudamba (AK), Nulu Nansikombi (NN), Bruhan Kitandwe, and Sumin Lunkuse (SL), were involved in data analysis and manuscript writing. Hannington Oryem-Origa (HOO), was the overall supervisor and was involved in manuscript writing. All the authors proofread and approved the final draft of the manuscript.

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