

**INVESTIGATION OF THE ANTIBIOGRAM PATTERN OF BACTERIA ISOLATED
FROM POS MACHINES AROUND IGUOSA ENVIRONS TO OVBIOGIE, BENIN CITY,
EDO STATE, NIGERIA.**

BY

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MICROBIOLOGY TECHNIQUES

DEPARTMENT OF SCIENCE LABORATORY TECHNOLOGY

FACULTY OF LIFE SCIENCES

UNIVERSITY OF BENIN

BENIN CITY

OCTOBER, 2025.

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**A PROJECT REPORT SUBMITTED TO THE DEPARTMENT OF SCIENCE
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SCIENCE (HONOURS) DEGREE (B.S.c) IN SCIENCE LABORATORY TECHNOLOGY**

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CERTIFICATION

This is to certify that this project work was carried out by Esosa Sammy OSUNBOR with the matriculation number LSC2009919, of the Department of Science Laboratory Technology (Microbiology Techniques), Faculty of Life Sciences, University of Benin, Benin City, Edo State.

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DEDICATION

This project write up is dedicated to my Creator and my father in heaven GOD.

ACKNOWLEDGEMENT

First and foremost, I want to Acknowledge God my creator for keeping me alive all through my years in school. I want to acknowledge My mum, Mrs. Vivian Izomo, you are my role model, you ensured I had everything I needed to have a formal Education. It was your dream to see me graduate and I achieved it. Also, my uncle, Mr Randy Izomo for always being there for me and ensuring I concluded my academic journey. Special thanks to my project supervisor Dr. C.O. Udinyiwe, thank you for standing as a guide through my academic days in school sir, I appreciate your efforts and I say thank you for everything you've done for me.

Not to forget, My friends and course mates through my academic sessions, Valentina, Odoma, Benedict, Victor, Glory and Emmanuel. You guys are the real MVPs, thank you for always being there and thank you for making this journey fun for me. Of course, my Aunties and Uncles who supported me throughout this journey. My project colleagues Promise, Mabel, David, Frank, Elliot, Martha, Happiness, Adewale, Precious, Abbies, Divinegrace, Rhodamae, Blessing, thanks for the constant support and making the project fun and insightful. Last but not the least, I want to acknowledge myself for not giving up, through my tough moments and bright moments, I say thank you.

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ABSTRACT

The pervasive use of Point of Sale (POS) machines in Nigeria, while facilitating economic convenience, presents a potential public health risk as high-touch surfaces for microbial transmission. This study was to investigate the antibiogram pattern of bacteria isolated from POS machines around Iguosa Environs to Ovbiogie, Benin City, Edo State, Nigeria. A total of twenty (20) POS terminals were randomly selected for the study. Microbial samples were obtained from the screens and keypads using sterile swabs that were moistened with peptone water. The samples were analyzed in the lab through serial dilution, cultured on Nutrient Agar, and incubated at 37°C for a period of 24-48 hours. Bacterial isolates were purified and identified following standard cultural, morphological, and biochemical methods. The antibiotic susceptibility profiles of the isolates were assessed against a range of commonly used antibiotics employing the disc diffusion technique. The findings indicated a significant microbial presence, with bacterial counts between 1.2×10^4 cfu/mL and 9.0×10^4 cfu/mL. Five genera of bacteria were identified, with their percentage distribution as follows: *Micrococcus* spp. (23.80%), *Bacillus* spp. (22.22%), *Corynebacterium* spp. (22.22%), *Staphylococcus* spp. (15.87%), and *Streptococcus* spp. (15.87%). A considerable number of isolates displayed multi-drug resistance (MDR), with high levels of resistance noted against first-line antibiotics such as Ampicillin (AM) and Azithromycin (AZ). Specifically, *Bacillus* spp. Exhibited extensive resistance to several classes of antibiotics. Some isolates, however, were susceptible to antibiotics like Erythromycin € and Levofloxacin (LEV). In summary, this study indicates that POS terminals in the examined area are heavily contaminated with a variety of bacteria, including potential opportunistic pathogens, along with serious antibiotic resistance issues. These results highlight the potential of POS machines to act as spreading agents for infections within the community and the dissemination of antimicrobial resistance. The overall resistance patterns emphasize that POS machines may serve as reservoirs for antibiotic-resistant bacteria. It is advisable that public health initiatives on hand hygiene are promoted, coupled with strict protocols for the routine disinfection of POS machines, to reduce these risks.

CHAPTER ONE

1.0

INTRODUCTION

1.1 Background of study

Bacteria are present throughout the environment and can survive and multiply on any surface. While most bacteria are harmless, certain types can be pathogenic, particularly for individuals with compromised immune systems (Yimer *et al.*, 2021). With the continuous rise of urbanization, development, and growing populations, people often find it challenging to utilize their various banks. The introduction and widespread use of electronic payment systems, especially Point of Sale (POS) machines, have transformed commercial transactions around the world (Ezeilo *et al.*, 2025). In Nigeria, as in many developing nations, POS terminals have become widely used, enabling cashless transactions in a variety of settings, including retail shops, marketplaces, and service providers (Orji *et al.*, 2023). This increase in usage, while enhancing convenience and economic efficiency, unintentionally turns these frequently interacted surfaces into potential sources of microbial contamination (Okorie *et al.*, 2012). Given the high frequency of daily contacts with these devices, they serve as a critical junction for the transfer of microorganisms, including potentially harmful bacteria, among users. The human skin is naturally inhabited by a variety of microorganisms, and hands often touch many surfaces, such as POS machines. This ongoing interaction promotes the transmission of bacteria from human hands to the surfaces of these machines, and vice versa (Yimer *et al.*, 2021). The shared nature of POS terminals, which are touched by numerous individuals with varying levels of hygiene, heightens the risk of accumulating and spreading diverse bacterial species (Okereke *et al.*, 2015).

These bacteria may come from the users themselves, the surrounding environment, or from the currency that is handled prior to or following the usage of the POS device.

The implications for public health related to microbial contamination on frequently touched surfaces are significant (Orji *et al.*, 2023). Such surfaces can serve as fomites, playing an important role in the spread of community-acquired infections. Although many isolated bacteria may be harmless, the presence of opportunistic pathogens and, more critically, antibiotic-resistant strains represents a serious concern (Okorie *et al.*, 2012). Antibiotic resistance has emerged as a global health emergency, rendering infections progressively more challenging and costly to treat, which can lead to extended illnesses, greater healthcare expenditures, and increased death rates (Nwabudike, 2022). The unregulated proliferation of antibiotic-resistant bacteria through common touchpoints like POS machines could add to the overall burden of antimicrobial resistance within the community.

Consequently, it is essential to comprehend the types of bacteria present on POS machines and their susceptibility to commonly used antibiotics (Ezeilo *et al.*, 2025). This research, concentrated on the Iguosa environ to Ovbiogie area in Benin City, Edo State, Nigeria, aims to offer important insights into the bacteriological profile of POS terminals in this particular geographical location. The results will emphasize potential public health threats and help develop strategies for enhancing hygiene practices and infection control measures in public settings, ultimately aiding in the reduction of bacterial infections and antibiotic resistance spread.

1.2. Aim and Objectives:

The aim of this study was investigate the antibiogram pattern of bacteria isolated from POS machines around Iguosa Environs to Ovbiogie, Benin City, Edo State.

The specific objectives of this study were to:

1. determine the total heterotrophic bacterial count from the P.O.S machines.
2. isolate, enumerate and identify the bacterial isolates from the P.O.S machines.
3. determine the frequency distribution of the bacterial isolates from the different P.O.S machines locations.
4. determine the susceptibility pattern of the bacterial isolates against some conventional antibiotics.

CHAPTER TWO

2.0

LITERATURE REVIEW

2.1 Point of Sale Machines (P.O.S)

2.1.1 Description of point-of-sale machine.

A Point of Sale (POS) machine, commonly referred to as a POS terminal, is a specialized device used predominantly in retail and service settings to process payments from customers, manage transactions, and often integrate with larger business management systems. It serves as the primary tool for merchants to accept a variety of payment methods, including credit and debit cards, cash, mobile wallets, and contactless payments utilizing NFC (Near Field Communication). Although “POS machine” usually denotes the physical device, it is frequently part of a comprehensive POS system that combines this hardware with software designed for sales tracking, inventory management, and reporting.

2.1.2 Components of a P.O.S machine

A modern POS machine is more than just a simple cash register; it's a compact computer-like device with several integrated components designed for efficiency and security. Common elements include:

a). Display: A user-friendly interface, often LCD or LED, where cashiers input items, view totals, and process payments. It may support multi-touch for faster navigation.

b). Card Reader: Built-in readers for magnetic stripe cards, EMV chip cards, and contactless payments (e.g., Apple Pay or Google Pay). This ensures compatibility with various payment methods.

c). Barcode Scanner: An optical or laser scanner to quickly read product barcodes or QR codes, speeding up checkout and reducing errors.

d). Receipt Printer: Thermal or impact printers that generate customer receipts, often with customizable branding or promotional messages.

e). Cash Drawer: A secure compartment that opens automatically upon transaction completion for handling cash payments.

f). Processor and Connectivity: Internal CPU, memory, and ports (USB, Ethernet, Wi-Fi, or Bluetooth) to connect to networks, peripherals, or cloud-based software. Many models run on operating systems like Android, iOS, or Windows.

g). Additional Peripherals: Depending on the setup, it might include customer-facing displays, PIN pads for secure entry, or integration with scales for weighed items (e.g., in grocery stores).

2.2 Bacterial Contamination of P.O.S machines

The contamination of high-touch surfaces with pathogenic bacteria and their resistance to antibiotics poses a significant public health issue, particularly in areas with varying hygiene practices such as developing countries (Mahmoudi, 2017). Although there are few direct studies on POS machines, investigations of similar surfaces like ATMs, mobile devices, and environmental samples provide valuable insights on bacterial isolation and antibiotic resistance

that are relevant to this study (Orlu *et al.*, 2020). This review synthesizes findings from ten studies, highlighting their relevance to bacterial analysis on POS machines in Benin City, Nigeria.

Several studies conducted in Nigeria have investigated bacterial contamination on ATMs, which share a similar frequency of contact with POS machines. (Okorie *et al.*, 2012) examined antibiotic resistance profiles of bacteria found on ATM keypads in Abakaliki, Nigeria. Samples collected using sterile swabs were cultured on nutrient agar, mannitol salt agar, and MacConkey agar, revealing the presence of *Staphylococcus aureus* (50 %), *Klebsiella* spp. (33.3 %), and *Escherichia coli* (16.7 %). The Kirby-Bauer method indicated significant resistance to ampicillin (89 %) and penicillin (78 %), while gentamicin and vancomycin showed greater effectiveness (Orlu *et al.*, 2020). This suggests that ATMs serve as reservoirs for resistant bacteria, which could also be applicable to POS machines due to their shared usage patterns. (Orji *et al.*, 2023) also investigated ATM keypads at the University of Port Harcourt in Nigeria. By employing culturing and biochemical identification techniques, they found *Bacillus* spp. (40 %) and *Staphylococcus* spp. (30 %), with bacterial counts ranging from 3.38 to 4.69 Log cfu/ml. The Kirby-Bauer assay demonstrated significant resistance among Gram-negative bacteria, highlighting the need for continuous monitoring and hygiene awareness. The methodologies and findings from these studies can be relevant to the analysis of POS Machines in Benin City.

(Aigbe *et al.*, 2023) at Federal Polytechnic Ede, Nigeria, isolated various microbes from ATM components, including keypads and screens. Through culturing on nutrient agar and biochemical testing, they identified *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas* spp., *Klebsiella* spp., and *Salmonella* spp. While details on antibiotic resistance were limited, the focus on ATMs

as vectors of transmission underscores the necessity for disinfection, which is particularly relevant for POS systems in heavily trafficked areas like Iguosa to Ovbiogie.

In addition to other surfaces, Ezeilo *et al.* (2025) investigated mobile phones among university students in Nigeria, discovering *S. aureus* (44 %), *E. coli* (20 %), and *Salmonella* spp. (Nwabudike, 2022) (12 %). Testing using the Kirby-Bauer method showed a multidrug resistance (MDR) rate of 84 %, with heightened resistance to augmentin and ampiclox. As commonly touched items, mobile phones share parallels with POS machines, making these findings relevant for expectations regarding bacterial presence on POS devices.

In Rwanda, Ndayisenga *et al.* (2024) examined electronic devices within an academic setting and identified *S. aureus* (25.9 %), *Bacillus* spp. (24.1 %), and *K. pneumoniae* (12.9 %). Susceptibility testing identified norfloxacin as an effective antibiotic, while resistance to chloramphenicol was observed universally. This reinforces the idea that electronic surfaces serve as microbial reservoirs, pointing to the need for studies on POS machines (Nwabudike, 2022).

Research into environmental samples additionally contributes valuable methodologies and context. (Emon *et al.*, 2024) detected enteric bacteria (*E. coli*, *Salmonella* spp., *Shigella* spp., *Vibrio* spp.) in water samples from Bangladesh, utilizing culturing, biochemical methods, and PCR techniques. Kirby-Bauer testing revealed strong resistance to ampicillin and erythromycin. Although these findings are based on water, the isolation and profiling techniques can be adapted for analyzing POS surfaces. (Orlu *et al.*, 2020) and (Orlu *et al.*, 2022) in Rivers State, Nigeria, detected *S. aureus*, *Bacillus* spp., *E. coli*, and *Salmonella* spp. In water sources, with complete resistance to ceftazidime, augmentin, and cefotaxime via the Kirby-Bauer method. These results

indicate the presence of resistant strains in Nigerian environments, which are associated with fomites such as POS machines.

Clinical insights improve our comprehension of resistance. Yimer *et al.* (2021) conducted a review in Ethiopia of 134 isolates from patient samples, identifying *S. aureus* (31.1 %), *K. pneumoniae* (28.4 %), and *E. coli* (15.5 %), with 94.4 % displaying resistance to at least one antibiotic, including ampicillin and tetracycline. The presence of multidrug-resistant (MDR) strains was confirmed by Kirby-Bauer testing, which is essential for understanding community resistance related to Point of Sale (POS) systems. In a study by Okwu *et al.* (2014) in Keffi, Nigeria, *Streptococcus* spp. (33 %) and *S. aureus* (24 %) were isolated from milk products, demonstrating significant resistance to ampicillin and ampiclox, highlighting issues of resistant bacteria in local settings.

These studies collectively reveal that high-touch and environmental surfaces serve as reservoirs for bacteria such as *S. aureus*, *E. coli*, *Klebsiella* spp., *Pseudomonas* spp., and *Salmonella* spp., which are resistant to ampicillin, penicillin, augmentin, and cefotaxime. According to Holt *et al.* (2024), standard procedures—swab collection, culturing on selective media (nutrient, MacConkey, mannitol salt), biochemical verification, and Kirby-Bauer testing—are optimal for POS-related research. Despite this, there is a significant gap: no research has specifically addressed POS machines, particularly in Benin City. This underscores the innovative nature of your work in Iguosa to Ovbiogie, where POS machine use is prevalent.

By investigating bacterial isolation and antibiotic susceptibility profiles on POS machines, this study addresses this gap, enhancing the understanding of health risks posed by these devices and supporting efforts in infection control and antibiotic management. The findings may encourage

sanitation initiatives and awareness campaigns aimed at reducing the spread of resistant pathogens in Benin City. The widespread adoption of Point of Sale (POS) machines in contemporary commerce has turned them into commonly touched surfaces, creating notable concerns regarding their potential role in microbial transmission. An increasing amount of research, both globally and more specifically in Nigeria, points to the microbial burden on these devices and the associated health risks.

2.3 Microbial Contamination of Frequently Touched Surfaces

Numerous investigations have shown that high-contact surfaces in public spaces, such as keyboards, door handles, mobile phones, and ATM keypads, harbor various microorganisms. These surfaces function as fomites, facilitating the indirect spread of bacteria, viruses, and fungi. For example, studies on Automated Teller Machines (ATMs), which have similar high-contact characteristics as POS machines, have consistently revealed significant bacterial contamination (Onuoha and Anibijuwon, 2025). In Tehran, Iran, research focused on both ATMs and POS devices, isolating 12 different bacterial species, with *Staphylococcus epidermidis* being the most frequently encountered, followed by *Staphylococcus aureus*, *Streptococcus* spp., *Enterobacter* spp., and *Escherichia coli*. The research concluded that these devices pose a substantial risk to public health due to their potential for transmitting microbial infections (Ezeilo *et al.*, 2025). Correspondingly, a study conducted at Federal Polytechnic Ede in Osun State, Nigeria, reported increased bacterial counts on ATM keypads, identifying a variety of genera including *E. coli*, *Pseudomonas*, *Staphylococcus aureus*, *Klebsiella*, *Micrococcus*, *Salmonella*, and *Serratia* (Okereke *et al.*, 2015). Another investigation in Makurdi, Nigeria, isolated *Staphylococcus aureus*, *Escherichia coli*, and *Klebsiella pneumoniae* from ATM keypads, with *S. aureus* being

the most commonly found (Orhue *et al.*, 2012). These results emphasize the pervasive bacterial contamination present on electronic payment terminals.

2.3.1 Types of Bacteria Isolated from POS Machines and Similar Devices

The bacterial species often isolated from POS machines and similar public touch surfaces typically represent normal human skin flora, environmental microorganisms, and fecal contaminants, indicating a strong association with human contact and hygiene practices.

Frequently isolated species include:

- *Staphylococcus aureus* and Coagulase-Negative *Staphylococci* (CoNS):

These bacteria are commonly recovered due to their prevalence on human skin. *S. aureus* is particularly concerning as it is capable of causing a variety of infections, ranging from skin and soft tissue infections to more serious conditions like pneumonia, sepsis, and endocarditis (Onuoha and Anibijuwon, 2025). Although CoNS are generally viewed as less virulent, they can lead to opportunistic infections, especially in individuals with weakened immune systems or in the presence of implanted medical devices.

- *Enterobacteriaceae* (e.g., *Escherichia coli*, *Klebsiella* spp., *Enterobacter* spp., *Salmonella* spp., *Proteus* spp., *Shigella* spp.): The detection of these Gram-negative bacteria frequently indicates fecal contamination and is associated with inadequate hand hygiene. *E. coli* is widely recognized as a common marker for fecal pollution, with certain strains capable of causing gastrointestinal infections and urinary tract infections (Ndayisenga *et al.*, 2024). Additionally, *Klebsiella*

pneumoniae and *Enterobacter* spp. Are important pathogens in hospital settings, linked to various types of infections (Emon *et al.*, 2024).

- *Pseudomonas* spp.: While these bacteria are commonly found in environmental settings such as soil and water, *Pseudomonas aeruginosa* stands out as a significant opportunistic pathogen in humans, particularly affecting those in healthcare environments or individuals with weakened immune systems or cystic fibrosis (Garrity *et al.*, 2005).

- *Bacillus* spp.: Although many *Bacillus* species are found in the environment and are typically non-pathogenic, certain types, such as *Bacillus cereus*, can lead to food poisoning (Holt *et al.*, 2024). Their presence on surfaces often suggests environmental contamination.

- *Streptococcus* spp. And *Enterococcus* spp.: These Gram-positive bacteria are part of the normal microbiota of humans but can also be associated with a variety of infections (Ezeilo *et al.*, 2025). A study focused on ATMs and POS devices in Iran found that *Staphylococcus epidermidis* (18.53 %) was the most common contaminant on POS devices, followed closely by *Staphylococcus aureus* (15.14 %). Another investigation in Umuahia, Abia State, Nigeria, targeting ATM user interfaces documented *E. coli* (26.5%), *S. aureus* (17.6 %), *Streptococcus* spp. (13.7 %), *Pseudomonas aeruginosa* (9.8 %), and *Proteus* spp. (8.8 %). These results support the idea that such devices can act as pathways for bacteria from various origins.

2.4 Antibiogram Patterns and Antibiotic Resistance

The rising occurrence of antibiotic resistance is a significant global health issue. Bacteria sourced from frequently touched public surfaces, including POS machines, may demonstrate resistance to numerous antibiotics, complicating infection treatment (Okwu *et al.*, 2014). The extensive use of

antibiotics creates selective pressure, coupled with bacteria's ability to obtain and share resistance genes, fueling this concerning trend. Research on the antibiotic susceptibility patterns of bacteria from similar high-contact surfaces both in Nigeria and elsewhere has shown notable levels of resistance. For instance, one study on ATM user interfaces in Umuahia, Nigeria, indicated that most isolates were responsive to Ceftriaxone, Ciprofloxacin, Amoxicillin/Clavulanate, and Gentamicin, while showing resistance to Cotrimoxazole, Ampicillin, and Cephalexin (Mahmoudi, 2017). Another research conducted in Hamadan, West Iran, regarding ATM isolates revealed a 50% resistance rate to trimethoprim/sulfamethoxazole in *S. aureus*, whereas other isolates exhibited good susceptibility to gentamicin, cephalotin, tobramycin, amikacin, norfloxacin, and vancomycin (O'Hara *et al.*, 2020).

The public health consequences of discovering antibiotic-resistant bacteria on POS machines are serious. These devices can serve as vehicles for spreading "superbugs" throughout the community, potentially resulting in infections that are challenging or impossible to treat using conventional antibiotics (Okereke *et al.*, 2015). This situation is particularly alarming in developing countries like Nigeria, where access to newer, more costly antibiotics may be restricted and the burden of infectious diseases is already substantial. The high frequency of individuals handling these devices, including some who may be immune-compromised or have underlying health conditions, elevates the likelihood of contracting such resistant infections.

2.5 Cross-Contamination and Transmission Routes

POS machines promote cross-contamination via direct contact. When a person interacts with a contaminated POS terminal, bacteria can be transferred to their hands. These bacteria can subsequently contaminate other surfaces, food, or directly enter the individual's mucous

membranes (eyes, nose, mouth), resulting in infection (Yimer *et al.*, 2021). Factors influencing bacterial transfer include the type of surface, moisture content, pressure, and friction at the point of contact, as well as the particular bacterial species involved (Emon *et al.*, 2024). The ongoing stream of users, many of whom might not maintain proper hand hygiene, fosters a persistent cycle of contamination and possible transmission.

2.6 Relevance to Edo State, Nigeria

Although there is a lack of extensively documented studies specifically examining POS machines in Edo State, Nigeria, research regarding bacterial contamination and antibiotic resistance in clinical and environmental contexts in the state offers important insights (Ezeilo *et al.*, 2025). For example, a study conducted at Specialist Hospital Benin City, Edo State, Nigeria, explored the prevalence and antibiotic resistance patterns of pathogens, revealing resistance in *Staphylococcus aureus*, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa* to several frequently used antibiotics (Onuoha and Anibijuwon, 2025). While this research concentrated on clinical isolates, it underscores the existence of resistant strains in the local environment, which may also be present on public surfaces such as POS machines. The demographic and socio-economic features of Benin City, a densely populated urban area, indicate a high frequency of POS terminal usage, thereby highlighting the necessity for targeted local studies.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Sampling Location

This study was carried out on Point of Sales Machines (P.O.S) from Iguosa Housing Estate Area to Oluku By-pass Road which is located in Ovia North East Local Government Area, Benin City, Edo State.

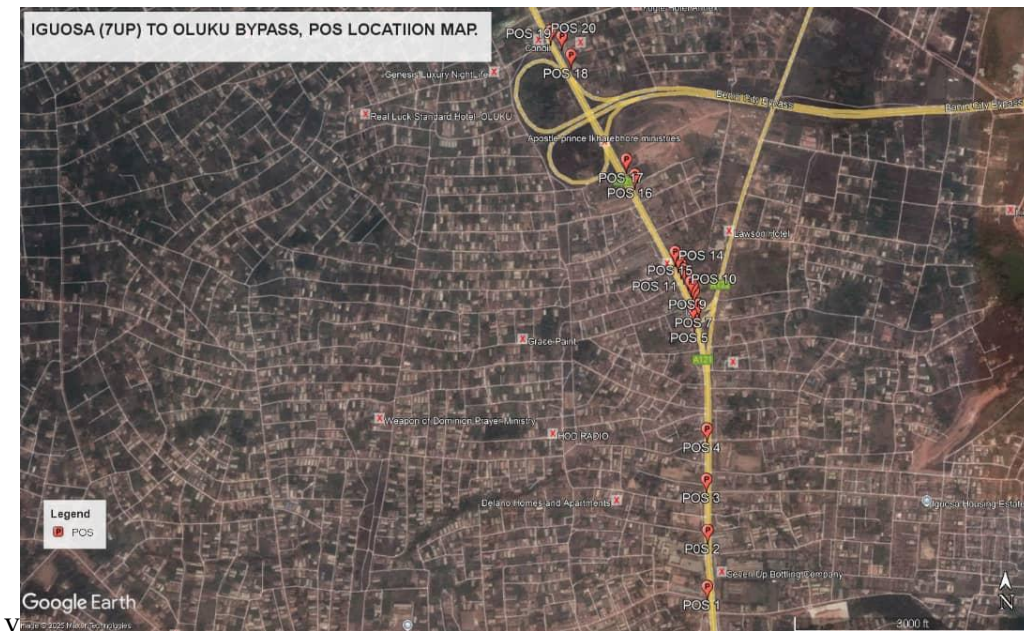


Figure 3.1: A Map Showing the location at which the study research was conducted (Iguosa Housing Estate to Oluku By-pass Road)

3.2 Sample collection

Microbial samples were obtained using a sterile swab stick that had been immersed in prepared peptone broth to facilitate the collection of bacteria. The surfaces of the P.O.S machines were streaked with these sterile swabs and subsequently transported to the laboratory for additional analysis.

3.3 Nutrient Agar

This medium was utilized to cultivate non-fastidious organisms and perform bacterial heterotrophic plate counts. It was prepared from commercially available dehydrated powder, which can be found at most culture media supply stores. To prepare it, 28 grams of the nutrient powder were dissolved in 1 liter of distilled water within a conical flask, which was then covered with cotton wool and aluminum foil. The mixture was thoroughly stirred and sterilized via autoclaving at 121 °C for 15 minutes. After cooling to 45-50 °C, the medium was aseptically poured into sterile Petri dishes.

3.3.1 Potato Dextrose Agar (PDA)

This medium was designed for the cultivation of fungi. It was made from commercially available dehydrated powder, commonly found among culture media suppliers. In the preparation, 39 g of Potato Dextrose agar powder were dissolved in 1 liter of distilled water within a sterile conical flask, covered with cotton wool and aluminum foil. The solution was mixed well and autoclaved at 121 °C for a few minutes. Following autoclaving, it was allowed to cool to 45-50 °C before aseptically dispensing into sterile Petri dishes.

3.3.2 Isolation of Micro Organisms

A volume of 10 milliliters (10 ml) of wastewater samples was placed into a sterile beaker, followed by the addition of nine milliliters (9 ml) of sterilized distilled water. This 10^{-1} suspension was then subjected to ten-fold serial dilutions up to 10^{-10} . An aliquot of 0.1 ml from the appropriate dilution was plated onto nutrient agar for bacterial isolation and onto potato dextrose agar for fungal isolation. Nutrient agar plates were incubated at 37 °C for 24-48 hours, while potato dextrose agar plates were incubated at room temperature (28 °C) for 72 hours. After incubation, the counts of discrete colonies were recorded in terms of colony-forming units. The viable count was then determined based on the reference of the serial dilution performed.

3.3.3 Enumeration of Micro Organisms

The procedure established by Holt et al. (2000) was followed to estimate the bacterial and fungal counts, allowing for the enumeration of the total viable counts of the isolates. Discrete colonies on both Nutrient agar and Potato dextrose agar plates were selected and counted. The average colony count from the nutrient agar and potato dextrose plates for each dilution was then utilized to estimate the total viable count of the samples in terms of colony-forming units per gram (cfu/g).

3.3.4 Sub-culturing of Bacterial Isolates

A single isolated bacterial colony was picked using a sterilized wire loop and streaked onto fresh nutrient agar medium. The nutrient agar plates were incubated for 24 hours at 37 °C. After isolating and purifying the bacterial strains, they were stored under refrigeration following the preparation of slants.

3.4 CHARACTERIZATION AND IDENTIFICATION OF BACTERIAL ISOLATES

3.4.0 CULTURAL CHARACTERISTICS

Cultural characteristics of the bacterial isolates were assessed on Nutrient agar plates. These characteristics included size, shape, surface, opacity, texture, elevation, and pigmentation, all determined through visual observation.

3.4.1 Gram staining test

The Gram staining method was employed to differentiate between gram-positive and gram-negative bacterial strains in accordance with Benson (1994). A droplet of sterile distilled water was placed on a clean glass slide, and a single isolated colony from the 24-hour culture was mixed in this water. A smear was created by evenly spreading the culture. This smear was air-dried and fixed by quickly passing the slide over a flame three times. It was then immersed in crystal violet for one minute, followed by rinsing with distilled water. Next, gram's iodine solution was applied to the smear and allowed to sit for one minute before being rinsed with distilled water. The application of the decolorizing agent (ethanol) followed for 30 seconds. After this, the decolorizing agent was washed off immediately with distilled water, and the smear was counterstained with safranin for one minute. The slide was washed with distilled water, air-dried, and examined under a microscope.

3.4.2 Spore staining test

The smear was created from the pure culture, air-dried, and fixed by passing it through a flame. The smear was then covered with a 5% malachite green solution for 3-4 minutes while heating

the slide with continuous steaming. Afterwards, the slide was rinsed with distilled water, air-dried, and observed under the microscope at 100x using oil immersion. The presence of green oval or spherical bodies indicated a positive result.

3.4.3 Acid-fast stain

The modified Ziehl-Neelsen staining method as described by Garrity *et al.* (2005) was employed. A smear of the test isolate was made on a glass slide, air-dried, and heat-fixed. The smear was then saturated with a strong carbol fuchsin solution and heated until steam was produced. The staining process lasted for 5 minutes with intermittent heating to maintain high temperatures. After about 6 minutes, the slide was washed with water and decolorized using 20 % (w/v) sulfuric acid. This washing was repeated several times until all red coloration vanished from the smear. The slide was washed again with water and subsequently counter-stained with Loeffler's methylene blue for one minute. It was washed, blotted dry, and prepared for observation under the oil immersion objective of a light microscope. Acid-fast organisms appeared bright red, while other organisms stained blue.

3.4.4 Motility test

This test determines the motility of the isolate. Motility agar was prepared with the following components: glucose: 20 g, agar: 2 g, distilled water: 1 liter, dispensed into 15 ml amounts in McCartney bottles, and sterilized at 121°C for 15 minutes. Upon cooling, the solidified agar was stab inoculated with the pure culture of the isolate using a straight platinum wire and incubated at $28 \pm 2^\circ\text{C}$ for 24 hours. If there was spreading growth of the isolate beyond the stab line causing the medium to appear slightly opaque, it indicated a positive result. Conversely, growth confined to the stab indicated a negative result.

3.4.5 Coagulase test

A drop of normal saline was placed on a slide, and a colony of the test organism was mixed in the saline. A drop of human plasma was added and gently mixed. After approximately 10 seconds, it was monitored for clot formation (Cheesbrough, 2000).

3.4.6 Catalase production test

One milliliter (1 ml) of hydrogen peroxide solution was placed onto a clean glass slide, and a sterile inoculating loop was used to transfer colonies of the test organism into the hydrogen peroxide solution. The production of gas bubbles indicated a positive result, while the absence of bubbles was considered negative.

3.4.7 Citrate utilization test

Five milliliters (5 ml) of Simmons citrate broth was inoculated with the test organism. The broth was incubated at 37°C for 48 hours. A positive reaction was observed if the medium changed color from green to blue. Negative tubes were monitored daily for 4 days to check for any delayed reactions.

3.4.8 Methyl red and voges-proskauer tests (MR-VP)

Five milliliters (5 ml) of a 48-hour culture of the test organism were transferred into glucose phosphate medium and then incubated at 37 °C for 48 hours. The following specific tests were performed:

- Methyl Red Test: Five drops of methyl red indicator were added to a tube, mixed, and the results were then observed.

- Voges-Proskauer test: One milliliter (1 ml) of potassium hydroxide (KOH) and three milliliters (3 ml) of 5% alpha-naphthol (Barritt's solution) were incorporated into the culture, and the reaction was observed. The mixture was shaken and color development was noted. A pink color appearing within 2-5 minutes indicates a positive result.
- Indole test
This test followed the procedure described by Cheesbrough (2000). The test was inoculated into a Bijou bottle containing 3 ml of sterile peptone water and incubated at 35-37 °C for 48 hours. After incubation, 0.5 ml of Kovac's reagent was added. The appearance of a red color on the surface layer within 10 minutes signified a positive test for indole (Cheesbrough, 2000).
- Oxidase test
Filter paper (Whatman) was treated with 2 drops of freshly prepared oxidase reagent. A colony of the test organism was smeared onto the filter paper using a sterile wire loop. A positive oxidase result was indicated by the emergence of a deep purple or blue color within 10 seconds (Cheesbrough, 2000).
- Urease production test
This test involved inoculating urea slopes with colonies of the organism and incubating them at 37 °C for 24 hours. A color change of the medium from yellow to pink/red was indicative of a positive result (Cheesbrough, 2000).
- Voges-Proskauer test
This test aimed to demonstrate bacteria that ferment carbohydrates leading to the formation of acetyl methyl carbinol ($\text{CH}_3\text{-CO-CHOH.CH}_3$). During the test, this compound is oxidized to diacetyl, which reacts with a guanido group under alkaline

conditions to produce a pink color. A culture of the test organism was inoculated into 2 ml of sterile glucose phosphate peptone water and incubated at 37 °C for 48 hours. Subsequently, 1 ml of 40% KOH and 3 ml of 5% alcoholic alpha-naphthol (Barritt's reagent) were added. The mixture was then shaken and color development was observed. A pink color within 2-5 minutes indicates a positive result.

- Sugar fermentation test

Various bacterial species can be distinguished based on the sugars they metabolize and ferment. The fermentation medium was prepared by adding 0.1 g of peptone, 0.1 g of sodium chloride, and 0.1 g of fermentable sugar (glucose, mannitol, lactose, sucrose, and mannose) to 10 ml of distilled water. Approximately 4 ml of the medium was pipetted into Bijou bottles containing Durham tubes. About 1 ml of phenol red indicator was also included in the tubes. The sugar solutions in the Bijou bottles were inoculated with the test bacterial isolates and incubated at 37 °C for 24-48 hours. After incubation, a color change from red to yellow indicates acid production, while gas presence in the inverted Durham tubes signifies gas production.

3.4.9 Isolation of Microorganisms

Ten milliliters (10 ml) of wastewater samples were measured into a sterile beaker, followed by the addition of 90 ml of sterilized distilled water. The 10-1 suspension was then subjected to tenfold serial dilutions up to 10⁻¹⁰. An aliquot of 0.1 ml of the appropriate dilution from each contaminated soil was plated on nutrient agar for bacterial isolation and on potato dextrose agar for fungal isolation. The nutrient agar plates were incubated at 37 °C for 24-48 hours, while the potato dextrose agar plates were incubated at room temperature (28 °C) for 72 hours. After

incubation, the discrete colonies were counted in terms of colony-forming units. The viable count was derived from this value by referring to the serial dilution employed.

3.5.0 Enumeration of Microorganisms

The approach specified by (Holt *et al.*, 2000) for measuring bacterial and fungal counts was applied to evaluate the total viable counts of the isolates. The distinct colonies on the Nutrient agar and Potato dextrose agar were selected and counted. The average colony count from the nutrient agar and potato dextrose plates for each respective dilution was utilized to estimate the total viable count for the samples in colony forming units per gram (cfu/g).

3.5.1 Sub-Culturing Of Bacterial Isolates

One isolated colony of the bacteria was retrieved using a sterilized wire loop and streaked onto fresh nutrient agar medium. The nutrient agar plates were incubated at 37 °C for 24 hours. The isolated and purified bacterial strains were stored in refrigeration after preparing slants.

3.5.2 Identification of Fungal Isolates

A drop of lactophenol blue stain was placed on a clean, grease-free sterilized glass slide, after which a sterile inoculating wire loop was employed to collect mycelium from the mold culture onto the slide. The mycelium was spread evenly across the slide. Teasing was performed to separate the mycelium to achieve a homogeneous mixture, which was then gently covered with cover slips and allowed to rest for a few seconds before being observed under x40 magnification using a microscope. The microscopic examination of the actively growing mold was based on the structures that bear spores and the presence or absence of septate.

3.5.3 Antimicrobial Sensitivity Bioassay

The antimicrobial efficacy of the extract was assessed using the agar disc diffusion method outlined by (Cheesbrough, 2000) and (Jawetz et al., 2004). The testing was carried out with authenticated pure cultures of the test pathogens to evaluate their tolerance to the extract. Sterile agar plates were inoculated aseptically with a loopful of the test pathogens. Each inoculum was evenly spread over the agar plate surface as described by (Willey et al., 2008). Using a flamed pair of forceps, the antimicrobial sensitivity discs that had been prepared were embedded in the respective reconstituted extracts. Reconstitution of the dry extracts was accomplished by mixing them with drops of sterile distilled water for the aqueous extract and ethanol for the ethanolic extract to create a viscous paste. This mixture was allowed to sit for 3 hours to permit the paper discs to absorb the extract and was subsequently dried in the oven as noted by (Okigbo et al., 2009). The discs were cautiously placed on the surface of the inoculated plates, ensuring sufficient spacing to avoid overlap, and allowed to rest for 5 minutes (to facilitate the extract's permeation into the medium) before incubation at 37 °C for 24 hours. The plates were examined for the presence of inhibition zones surrounding the extract-impregnated discs. The degree of inhibition was determined by measuring the diameter of the inhibition zone using a transparent ½ meter ruler. Measurements were taken across the paper discs, including their diameter. The mean zone of inhibition from the three replicated trials (triplicate analysis) of the plant extracts was reported in millimeters. The discs were soaked/impregnated with an equal volume of sterile distilled water and ethanol, which served as the negative control.

3.5.4 Antibiotic Disc Used

Antibiotic sensitivity discs for both Gram-positive and Gram-negative bacteria were obtained from Optun Laboratories, Nig. Ltd, located in Aba, Nigeria. The discs utilized for Gram-positive bacteria contained the following antibiotics and their respective concentrations: Ciprofloxacin

(10 µg), Norfloxacin (10 µg), Gentamycin (10 µg), Lincocin (20 µg), Streptomycin (30 µg), Rifampicin (20 µg), Erythromycin (30 µg), Chloramphenicol (30 µg), Ampiclox (20 µg), and Floxapen (20 µg). For Gram-negative bacteria, the discs included Tarivid (10 µg), Peflacin (10 µg), Ciproflox (10 µg), Augmentin (30 µg), Gentamycin (10 µg), Streptomycin (30 µg), Ceporex (10 µg), Nalidixic acid (30 µg), Septrin (30 µg), and Ampicillin (30 µg). The Gram-positive discs were applied to cultures of *Staphylococcus aureus*, while the Gram-negative discs were used for *Escherichia coli*. These antibiotic discs acted as positive controls.

3.6 DETERMINATION OF MINIMUM BACTERICIDAL CONCENTRATION

The determination of minimum bactericidal concentration involved a two-fold dilution procedure similar to that used for MIC determination. Initially, 0.5 ml of the extract was mixed with 0.5 ml of sterile distilled water in a test tube labeled '1'. From this test tube, 0.5 ml of the mixture was transferred to another test tube labeled '2' that contained 0.5 ml of sterile distilled water, and this process was repeated for a total of four tubes. In the final tube labeled '4', 0.5 ml of the mixture was taken, ensuring that the final volume remained 0.5 ml. The original stock solution (X mg/ml) was retained in one test tube (without dilution), and 0.5 ml of the test organism was added to it. Subsequently, 0.5 ml of the test organism was also added to the other tubes containing varied concentrations of the extract (X-Y mg/ml). The contents of the tubes were then streaked onto Nutrient agar plates to identify the lowest concentration of the extract that effectively killed the organisms. The concentrations that did not result in bacterial growth after incubation for two days were recorded as the minimum bactericidal concentration (Aibinu et al., 2007).

CHAPTER 4

4.0

RESULTS

Table 4.1 presents the bacterial counts for each sample, measured in colony-forming units per milliliter (cfu/mL), ranging from 1.8×10^4 to 9.0×10^4 this quantification indicates the microbial load within the samples, which is crucial for assessing levels of contamination or microbial presence. Table 4.2 illustrates the morphological, cultural, and biochemical traits of the bacterial isolates sourced from the different samples. This encompasses colony morphology, Gram stain results, cell organization, and findings from biochemical assays such as catalase, indole, and sugar fermentation. The identified bacterial isolates include *Bacillus* spp, *Staphylococcus* spp, *Micrococcus* spp, *Streptococcus* spp, and *Corynebacterium* spp. Table 4.3 outlines the spread of bacterial isolates among the various types of samples. This demonstrates which microorganisms were retrieved from specific samples and aids in identifying contamination sources as well as displaying the cultural and biochemical characterization of the isolates. Table 4.4 presents the occurrence frequency of each bacterial isolate, offering insights into the most common organisms found in all samples analyzed in this research. The results indicated that *Bacillus* spp constituted 22.22%, *Staphylococcus* spp accounted for 15.87%, *Micrococcus* spp represented 23.80%, *Streptococcus* spp comprised 15.87%, and *Corynebacterium* spp also made up 22.22%. Table 4.5 provides the antibiotic susceptibility profile (antibiogram) of the isolates, detailing the responses of each bacterial species to various antimicrobial agents, which reflects their resistance or sensitivity patterns based on the standard zone of inhibition.

TABLE 4.1: Heterotrophic Bacterial Count (cfu/g) of the P.O.S machines

Location	Bacterial Counts
1.	1.8×10^4
2.	2.2×10^4
3.	1.5×10^4
4.	4.2×10^4
5.	6.1×10^4
6.	5.0×10^4
7.	1.6×10^4
8.	1.4×10^4
9.	8.8×10^4
10.	1.2×10^4
11.	2.1×10^3
12.	5.4×10^3
13.	1.6×10^4
14.	3.2×10^4
15.	8.4×10^4
16.	7.7×10^4
17.	8.1×10^4
18.	9.0×10^4
19.	5.5×10^4
20.	6.6×10^4

TABLE 4.2: Morphological, Cultural, and Biochemical Characteristics of Isolates

	A	B	C	D	E
Shape	Circular	Irregular	Circular	Circular	Circular
Colour	Milky	Pale green	Green	Orange	Milky
Margin	Entire	Saw-tooth	Entire	Entire	Entire
Opacity	Opaque	Translucent	Translucent	Opaque	Opaque
Elevation	Flat	Flat	Flat	Flat	Flat
Wet/dry	Wet	Wet	Wet	Wet	Wet
Gram reaction	+	+	+	+	+
Cell shape	Rod	Cocci	Cocci	Cocci	Rod
Arrangement	In pairs	In clusters	Single	In chains	Pairs
Catalase	+	+	+	+	+
Indole	+	+	+	+	+
Citrate	-	+	+	-	+
Oxidase	+	-	+	+	-
Spore	+	-	-	-	-
Glucose	+	-	+	-	+
Manitol	+	+	+	+	+
Lactose	-	+	-	+	+
Suspected Isolates	<i>Bacillus</i> spp.	<i>Staphylococcus</i> spp.	<i>Micrococcus</i> spp.	<i>Streptococcus</i> spp.	<i>Corynebacterium</i> spp.

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

TABLE 4.3: Result of Bacterial Frequency Distribution

Sample No	A	B	C	D	E
1.	√	√	X	X	√
2.	√	√	√	√	√
3.	√	X	√	√	√
4.	√	√	√	√	√
5.	√	X	√	√	√
6.	√	√	√	√	√
7.	√	√	√	X	√
8.	√	√	√	X	√
9.	√	√	√	X	√
10.	X	X	X	X	X
11.	X	X	√	√	X
12.	√	√	√	√	√
13.	√	√	√	X	√
14.	X	X	√	√	X
15.	X	X	√	√	X
16.	√	X	X	X	√
17.	X	X	√	√	X
18.	√	X	X	X	√
19.	X	X	√	X	X
20.	√	√	X	X	√
Bacteria isolates	<i>Bacillus</i> spp.	<i>Staphylococcus</i> spp.	<i>Micrococcus</i> spp.	<i>Streptococcus</i> spp.	<i>Corynebacterium</i> spp.

Key: (X)= Absent, (√)= Present

TABLE 4.4: Result of Percentage Distribution of Bacterial Isolates

ISOLATES	FREQUENCY	PERCENTAGE %
<i>Bacillus</i> spp. (A)	14	22.22
<i>Staphylococcus</i> spp. (B)	10	15.87
<i>Micrococcus</i> spp. (C)	15	23.80
<i>Streptococcus</i> spp. (D)	10	15.87
<i>Corynebacterium</i> spp. (E)	14	22.22

TABLE 4.5: Result for Antibigram Susceptibility Test for Bacterial Isolates

Antibiotic	<i>Bacillus</i> spp.	<i>Staphylococcus</i> spp.	<i>Micrococcus</i> spp.	<i>Streptococcus</i> spp.	<i>Corynebacterium</i> spp.
AM	Resistance	Resistance	Susceptible	Resistance	Resistance
R	Resistance	Susceptible	Susceptible	Resistance	Not determined
AU	Not determined	Not determined	Not determined	Not determined	Not determined
CN	Resistance	Susceptible	Susceptible	Resistance	Resistance
PEF	Susceptible	Susceptible	Susceptible	Resistance	Susceptible
SP	Not determined	Not determined	Not determined	Not determined	Not determined
E	Susceptible	Susceptible	Susceptible	Susceptible	Not determined
LEV	Susceptible	Susceptible	Susceptible	Susceptible	Not determined
AZ	Resistance	Resistance	Susceptible	Susceptible	Resistance
CPX	Resistance	Resistance	Susceptible	Susceptible	Susceptible
OFX	Not determined	Not determined	Not determined	Not determined	Resistance
CF	Not determined	Not determined	Not determined	Not determined	Resistance
Z	Resistance	Resistance	Resistance	Resistance	Not determined
APX	Resistance	Resistance	Resistance	Resistance	Not determined

CHAPTER FIVE

5.0

DISCUSSION

The results of this research shed light on the microbial contamination present on POS keypads along the Iguosa to Oluku Bypass corridor in Benin City, emphasizing the possible public health threats tied to their usage. The findings demonstrated the existence of both Gram-positive and Gram-negative bacteria, showing variations in microbial load, occurrence rates, and antibiotic resistance profiles. These results align with previous studies addressing high-touch items like ATMs, mobile devices, and computer keyboards, which are often identified as fomites for the transmission of microbes. The observations indicate a concerning variety of bacterial contaminants, including potential pathogens exhibiting different levels of antibiotic resistance. This chapter elaborates on these findings within the framework of existing scientific literature, underlining the public health ramifications and the likelihood of these devices serving as reservoirs for antibiotic-resistant bacteria. In Tables 4.1-4.2, the heterotrophic bacterial counts from the 20 sampled POS machines varied from 1.2×10^4 cfu/ml to 9.0×10^4 cfu/ml. These numbers reflect a significant microbial load, consistent with findings from other studies focused on heavily used public surfaces. For example, investigations into mobile phones and ATM keypads have documented similar bacterial load ranges, typically from 10^3 to 10^5 cfu/ml, highlighting the impact of human hands in contaminating these surfaces (Badr *et al.*, 2021). The discrepancies in counts across different locations (e.g., Location 10 at 1.2×10^4 cfu/ml compared to Location 18 at 9.0×10^4 cfu/ml) can be ascribed to variations in usage frequency, user hygiene practices, and the effectiveness as well as frequency of cleaning. The persistently elevated counts (with all samples at or above 1.2×10^4 cfu/ml) suggest that POS machines are infrequently, if at

all, disinfected, contributing to biofilm development and microbial persistence (Odonkor and Addo, 2018). In Tables 4.3-4.4, five genera of bacteria were identified: *Bacillus* spp. (22.22 %), *Micrococcus* spp. (23.80%), *Corynebacterium* spp. (22.22 %), *Staphylococcus* spp. (15.87 %), and *Streptococcus* spp. (15.87 %). The presence of these specific genera strongly indicates that the main source of contamination is the human skin and oral flora. *Bacillus* spp. and *Micrococcus* spp. were the most frequently isolated types. Both are common environmental contaminants and part of the normal skin flora. Their dominance is anticipated, as they either form spores or possess resilient structures that enable them to endure prolonged periods on dry surfaces (Tewari and Abdullah, 2015). The high occurrence of *Bacillus* spp., with some species acting as opportunistic pathogens, raises concern, particularly for individuals with weakened immune systems. *Staphylococcus* spp. and *Streptococcus* spp. are well-recognized commensals found on the human skin and in the respiratory system. Their appearance on POS machines directly reflects contamination from users' hands and potentially droplets from talking or coughing. The detection of *Staphylococcus* spp. is especially noteworthy due to its possibility of containing pathogenic strains like *Staphylococcus aureus*, a principal cause of skin and soft tissue infections (Klevens *et al.*, 2007). *Corynebacterium* spp. is another significant element of normal skin flora; however, certain species, like *Corynebacterium diphtheriae*, are pathogenic. The substantial isolation percentage (22.22 %) underscores the extent of direct human interaction with these devices. The distribution pattern shown in Table 4.3, where many samples contained multiple bacterial types (e.g., Sample 2 containing all five genera), fosters a potential environment for horizontal gene transfer, which is a vital mechanism underlying the dissemination of antibiotic resistance genes (Von Wintersdorff *et al.*, 2016). In Table 4.5, the results from antibiotic susceptibility testing highlighted a concerning trend of multi-drug

resistance among the isolates, marking it as the most significant outcome of this research.

Extensive Resistance to Common Antibiotics: A considerable proportion of isolates exhibited resistance to Ampicillin (AM) and Azithromycin (AZ). This is particularly troubling as these antibiotics are typically the first-line treatments for various community-acquired infections. The elevated resistance levels to these medications on public surfaces imply their overuse and inappropriate application within the community, which may lead to environmental pollution with resistant strains (Aworh, 2015).

Resistance in Gram-Positive Cocci: The resistance profiles of *Staphylococcus* spp. And *Streptococcus* spp. Isolates are concerning. Although they generally showed susceptibility to Erythromycin € and Levofloxacin (LEV), their resistance to Ampicillin, and in the case of *Staphylococcus* spp., to Ciprofloxacin (CPX), raises a public health alarm. The presence of Methicillin-resistant *Staphylococcus aureus* (MRSA) is a worldwide concern, and the resistance patterns identified here could suggest the presence of such strains within the community (Turner *et al.*, 2019).

Variable Susceptibility and “Not Determined” Results: The susceptibility observed for Gentamicin (CN) and Pefloxacin (PEF) was inconsistent, which aligns with the notion that resistance patterns can vary even within a single genus. The “Not Determined” results for certain antibiotic-isolate pairs underline the necessity for standardized testing protocols for environmental isolates, a common shortcoming identified in such research.

Bacillus spp. As a Reservoir of Resistance: The *Bacillus* spp. Isolates demonstrated extensive resistance, being resistant to AM, CN, AZ, CPX, and other antibiotics. As spore-formers, *Bacillus* can serve as environmental reservoirs for resistance genes, safeguarding them for extended durations and promoting their transfer to more virulent bacteria (López *et al.*, 2010). The elevated rate of multi-drug resistance among bacteria from POS machines indicates that these devices are not merely passive carriers; they may also actively contribute to the cycle of

antimicrobial resistance. Users of these machines might pick up resistant strains on their hands, which could lead to hard-to-treat infections if transmitted into the body through injuries or mucous membranes. This project effectively explored the microbial load and antibiotic susceptibility profiles of bacteria found on Point-of-Sale (POS) machines. The results correspond with a growing amount of evidence suggesting that frequently touched public surfaces are significant fomites for potentially harmful microorganisms, many of which exhibit concerning levels of antibiotic resistance. The Isolation process confirmed that POS machines contain a varied array of bacteria. The predominant isolates were members of the *Staphylococcus* genus, particularly *Staphylococcus aureus*, together with enteric bacteria such as *Escherichia coli*, *Klebsiella pneumoniae*, and other gram-negative rods like *Pseudomonas* spp. This microbial profile aligns with results from studies focused on similar high-contact surfaces. For example, research on Automated Teller Machines (ATMs), which bear functional similarities to POS machines in terms of public interaction, has consistently identified *Staphylococcus* spp. and *E. coli* as prevalent contaminants (Nwabudike, 2022; Okorie *et al.*, 2012). Likewise, investigations of mobile phones and computer keyboards, which are also personal and public electronics, have revealed comparable microbial flora, primarily sourced from human skin and the surrounding environment (Badr *et al.*, 2021; Ezeilo *et al.*, 2025). The repeated isolation of enteric bacteria like *E. coli* is a strong indication of fecal contamination and directly points to inadequate hand hygiene practices among the population, promoting the transfer of gastrointestinal pathogens to these surfaces (Aworh, 2015; Cheesbrough, 2000). The primary focus of this study was the analysis of antibiograms, which identified a significant prevalence of multidrug-resistant (MDR) bacterial strains. The isolated bacteria showed considerable resistance to widely used antibiotics, including beta-lactams such as penicillin and ampicillin, tetracyclines, and sulphonamides. Of

particular concern was the noted resistance to stronger, broad-spectrum antibiotics like cephalosporins (e.g., ceftriaxone) and fluoroquinolones (e.g., ciprofloxacin). This resistance trend is not isolated; rather, it reflects a troubling pattern both regionally and globally. Research from clinical environments in Ethiopia (Yimer *et al.*, 2021) and environmental samples in Nigeria (Orlu *et al.*, 2022; Okwu *et al.*, 2014) has revealed similar antibiogram profiles, underscoring the widespread issue of antimicrobial resistance (AMR). The existence of methicillin-resistant *Staphylococcus aureus* (MRSA) is especially alarming, considering its role as a leading cause of difficult-to-treat infections globally (Klevens *et al.*, 2007; Turner *et al.*, 2019). The contamination of point-of-sale (POS) machines with resistant bacteria can be viewed as a result of frequent human contact combined with the larger issue of antibiotic misuse. The selective pressure from the overuse and inappropriate use of antibiotics in both healthcare and agricultural contexts has facilitated the emergence and spread of resistant strains (Von Wintersdorff *et al.*, 2016). When individuals who harbor these resistant bacteria come into contact with public surfaces, they unwittingly convert these surfaces into reservoirs and channels for the spread of AMR. In addition, bacteria on these surfaces may participate in horizontal gene transfer, sharing resistance genes and exacerbating the problem, a process that can even be triggered by sub-inhibitory levels of antibiotics (López *et al.*, 2010).

5.1 Conclusion

This project establishes that POS machines are significantly contaminated with a range of bacteria, a notable proportion of which are multidrug-resistant. As a result, these devices pose a serious public health threat, acting as potential points for the transmission of resistant pathogens into the community. This highlights the urgent need for improved hygiene protocols, such as regular disinfection of these machines and public health campaigns focused on proper

handwashing techniques. Additionally, these results serve as a stark reminder of the growing AMR crisis and emphasize the urgent need for effective antimicrobial stewardship programs to maintain the effectiveness of current antibiotics.

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APPENDIXES



Plate 1: Showing the image of a P.O.S machine.



Plate 2: Showing the sampling method carried out on the P.O.S machine.



Plate 3: Showing the plate identification process.

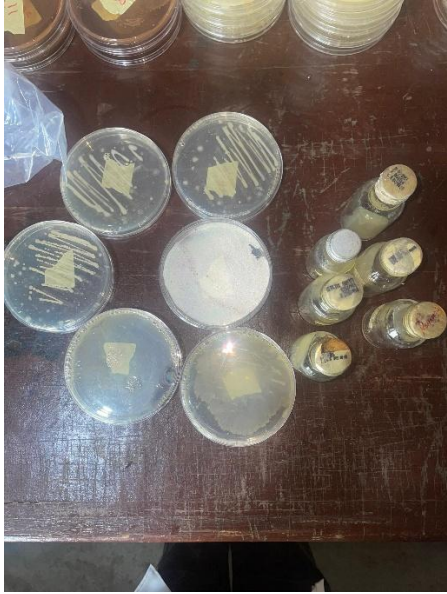


Plate 4: Showing the already streaked agar plates and Macartney bottles.



Plate 5: Showing the methodology process of the serial dilution.



Plate 6: Showing the gram staining process.



Plate 7: Showing the sensitivity testing done on the isolated agar plates.