ISOLATION AND IDENTIFICATION OF MICROBES ASSOCIATED WITH POS MACHINES OPERATED AROUND USELU TO TEXTILE MILL ROAD IN BENIN CITY, EDO STATE.

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Divinegrace Onyinyechi EZEOHA

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A PROJECT REPORT SUBMITTED TO THE DEPARTMENT OF SCIENCE LABORATORY TECHNOLOGY, FACULTY OF LIFE SCIENCES IN PARTIAL FULFILMENT OF THE REQUREMENT FOR THE AWARD OF BACHELOR OF SCIENCE (HONOURS) DEGREE (BSC.) IN SCIENCE LABORATORY TECHNOLOGY

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CERTIFICATION

This is to certify that this project work carried out by Divinegrace Onyinyechi EZEOHA (Miss) with the matriculation number, LSC2009985 of the department of Science Laboratory Technology (Biotechnology Technology), Life Sciences, University of Benin, Benin City, Edo State, Nigerian.

DR. O.C. UDINYIWE _____

(Project Supervisor) Signature and date

DR. P.O. ALONGE

_____

(Project Coordinator) Signature and date

PROF. J.O. OSARUMWENSE _____

(Head of Department) Signature and date

_____ ___ _____

(External Supervisor) Signature and date

DEDICATION

This book is dedicated to God Almighty whose unwavering love, guidance, direction and strength has fueled my journey of discovery.

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ABSTRACT

The rapid adoption of cashless transactions in Nigeria, particularly in bustling commercial areas like the Uselu to Textile Mill Junction, Benin City, Edo State, has led to the widespread use of Point-of-Sale (POS) machines. These devices, frequently handled by multiple users with varying hygiene practices, have raised concerns about their potential as fomites for microbial contamination and the spread of infectious diseases. The aim of this study was to isolate and identify bacteria associated with POS machines, determining their total heterotrophic count, assessing their distribution across different locations, and evaluating their susceptibility to commonly used antibiotics. Swab samples were collected from 20 POS machines (designated POS 1 to POS 20) during peak usage hour. The bacterial isolates were purified and identified based on standard cultural, morphological and biochemical characteristics. The results revealed significant bacterial contamination, with total viable counts ranging from 5.0×10^2 to 8.1×10^4cfu/ml. The predominant bacterial genera identified were *Micrococcus* spp., *Pseudomonas* spp., *Staphylococcus* spp., *Streptococcus* spp., and *Bacillus* spp. *Micrococcus* spp. and *Pseudomonas* spp had the same percentage occurrence of 21.67% being the most frequent bacterial isolates. The antibiotic susceptibility tests showed varying resistance patterns, with many isolates resistant to β-lactam antibiotics like ampicillin and ceftriaxone, raising concerns about the potential spread of antimicrobial resistance. This study highlights the urgent need for regular disinfection of POS machines, improved hygiene practices among users and operators, and public awareness campaigns to promote hand hygiene, the high microbial loads and presence of resistant strains underscore the public health risks posed by POS machines in high-traffic settings. These measures are critical to reducing the risk of fomite-mediated infections and combating the growing threat of antimicrobial resistance in community settings like Uselu to Textile Mill Junction.

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background of Study

Electronic banking, a product of Information and Communication Technology (ICT) has revolutionized the banking sector by enabling easier and more efficient financial transactions. E-banking systems technologies has developed to include, Automated Teller Machines (ATMs), Point of Sales (POS) device, Electronic Funds Transfer and Telebanking with the ATMs and POS machine as the most frequently used technologies (Folorunso *et al*., 2010). These services are provided within certain locations not only near the premises of banks but places such as market places, shopping centers/ malls, airports, petrol or gas stations, restaurants, hospitals or any place that large number of people gather (Okafor and Ezeani, 2012). Therefore, the development of e-banking services has not only affected economic status of countries but has had several deep social and cultural effects on the quality of individual lives. Studies indicate that increasing number of persons prefer to use the POS and ATMs machines than to queue at banking halls for financial transactions especially as most countries gravitate towards cashless economy (Abban and Thano Debbra, 2011).

A point-of-sale (POS) machine is a computerized telecommunications equipment driven by a software that processes transactions, manages data (e.g, sales, inventory), and interfaces with users via a display or touchscreen, it relies on a microprocessor or computing device to execute tasks, as a telecommunication equipment POS machines communicate with payment processors, banks, or card networks (e.g, Visa, Master-card) to authorize transactions in real-time. This requires an internet connection (Wi-Fi, cellular data, or wired) the machine transmits encrypted payment data over secure telecommunications networks to complete a sale (Kholod *et al*., 2024).

The advent of cashless transactions has led to the widespread use of Point of Sale (POS) machines across Nigeria, particularly in commercial hubs like Benin City, Edo State. In recent years, the rapid use of the point-of-sale (POS) machines in Nigeria has transformed financial transactions. These electronic devices, used for cashless payments, have become ubiquitous in markets, shops, and public spaces due to their convenience and efficiency in facilitating transactions (Dawodu and Akanbi, 2021).

However, the frequent handling of POS machines by multiple users, including vendors and customers with varying hygiene practices, raises significant public health concerns regarding microbial contamination. Microorganisms are defined as organisms which cannot be seen with the bare eyes but rather with the aid of a microscope, they include bacteria, yeasts, moulds, protozoa, algae, viruses etc. These microorganisms may be beneficial or disease causing (pathogenic) and need to be controlled in both cases. Microorganisms are prevalent and have an amazing ability to adapt to new environments and further multiply in large numbers within a limited time, they exist everywhere in the environment, in high populations in soil, in the air we breathe, the water we drink, the food we eat, on our skin, in our noses, throats, mouths and are able to persist or even grow on any surface (W.H.O., 2012).

There are reports indicating the presence of viable organisms on inanimate objects causing contamination, colonization and the spread of microbial infections (John and Adegoke, 2018; Otu-Bassey *et al*., 2022). Humans often acquire microorganisms from objects in their surroundings, with the hands playing a key role in contact and transmission. Frequently touched surfaces such as POS keypads, screens, and card slots can act as fomites,

hosting harmful microorganisms that may facilitate the spread of infectious diseases. These microbes can pose health risks, especially in communities with limited awareness of hygiene practices. Microbial contamination of shared surfaces has gained increasing attention in public health research due to the role of fomites, (objects capable of transferring pathogens, which cause disease spread). A representative number of microorganisms bear the potential to survive and are likely to persist on the surfaces of the POS machine especially if not regularly cleaned, thereby posing a risk of cross-contamination between users and potentially contributing to the spread of infectious agents in public settings as People come into contact with a variety of fomites on a regular basis, infections are becoming more common (Ya'aba *et al*., 2021).

The study area, Uselu to Textile Mill Junction is a busy commercial zone filled with active markets, numerous retail shops, and constant pedestrian movement. The frequent use of POS machines here reflects the nation's transition toward a cashless economy. Contamination of shared public surfaces by microbes is a recognized global issue, with the risk heightened in regions where environmental conditions such as high humidity, inadequate sanitation, and infrequent cleaning, favour the survival of microorganisms. In highly active trading centers like Uselu and Textile Mill Junction, the combination of dense human traffic, intense market activity, and continual handling of POS devices creates a suitable environment for microbial growth (Chawla *et al*., 2023). Public surfaces such as POS terminals, ATMs, currency notes, and market tools are known to host various bacteria, including *Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonas aeruginosa*, as well as fungi like *Aspergillus* species, all

of which can cause illnesses ranging from skin infections to gastrointestinal diseases (Otu-Bassey *et al*., 2022).

The significance of studying microbial contamination on POS machines lies in its implications for public health and safety. In Nigeria, where infectious diseases remain a leading cause of morbidity, understanding the microbial load on commonly used surfaces can inform preventive measures and policy recommendations. For instance, identifying the types and prevalence of microbes on POS machines can highlight the need for regular disinfection protocols or public awareness campaigns to promote hand hygiene among users (Obiebi *et al*., 2025; Sulaiman and Adam, 2025). Unlike ATMs, which are often maintained by banks, POS machines in Nigeria are typically operated by small-scale vendors with no interest for regular disinfection, the findings from this study could guide POS operators and users in adopting better hygiene practices, such as regular disinfection of machines (Fatehi *et al*., 2020). Furthermore, the results may provide evidence for policymakers to develop sanitation guidelines for shared devices in commercial settings, ultimately reducing the risk of microbial transmission in high-traffic areas like Uselu to Textile Mill Junction (Cobrado *et al*., 2017).

1.2 Aim of study

The aim of this research was to isolate and identity bacteria associated with POS machines operated around Uselu to Textile Mill Road Junction, Benin City, Edo state, Nigeria.

The specific objectives of this research were to:

i. determine the total heterotrophic bacterial count of the isolates from the POS machines operated around Uselu to Textile Mill junction.

ii. isolate, and identify the bacterial isolate from the POS machines.

iii. determine the frequency of the distribution of the bacterial isolates gotten from POS machine samples at different sampling location.

iv. determine the susceptibility pattern of the bacterial isolates against some known antibiotics.

CHAPTER TWO

2.0 LITERATURE REVIEW

A point-of-sale (POS) machine is a computerized telecommunications devices powered by a software that processes transactions, manages data (e.g, sales, inventory), and interfaces with users through a display or touch-screen, it relies on a microprocessor or computing device to execute task, as a telecommunication equipment, POS machines communicate with payment processors, banks, or card networks (e.g Visa, Master-card) to authorize transactions in real-time. This requires an internet connection (Wi-Fi, cellular data, or wired) the machine transmits encrypted payment data over secure telecommunications networks to complete a sale. The POS machine is a system that enables businesses to process transactions efficiently and accurately. It typically consists of a terminal, a card reader, and a printer. The terminal is the main component of the POS system, where transactions are processed and sales data is stored. The increasing reliance on electronic payment systems, such as point-of-sale (POS) machines, has transformed financial transactions globally, particularly in urban settings like Benin City, Nigeria. POS machines, widely used for cashless transactions, are high-contact surfaces that facilitate the transfer of microorganisms due to frequent handling by multiple users (Obiebi *et al*., 2025; Sulaiman and Adam, 2025). This chapter reviews existing literature on the microbial contamination of high-tough surfaces, with a focus on POS machines, to provide a foundation for the study on the isolation and identification of microbes associated with POS machines around Uselu to Textile Mill Junction, Benin City, Nigeria By synthesizing findings from related studies, this chapter highlights gaps in current

research and justifies the need for investigating microbial contamination on POS machines. The review is organized thematically, covering microbial contamination of public surfaces, specific studies on electronic payment devices, health implications, and factors influencing contamination (Cobrado *et al*., 2017; Fatehi *et al*., 2020).

2.1 MICROBIAL CONTAMINATION OF HIGH-TOUCH SURFACES

High-touch surfaces are objects or areas frequently contacted by multiple people, increasing the risk of germ transmission. Examples include doorknobs, light switches, countertops, currency notes, market utensils, handrails, elevator buttons, and shared equipment like keyboards or touchscreens and electronic devices; ATMs and POS machines. High-touch surfaces, such as POS machines, ATMs, doorknobs, and touchscreens, are frequently contacted by multiple individuals, making them potential fomites for microbial transmission (Kramer *et al*., 2006). These surfaces facilitate the spread of pathogens through direct contact, particularly in public settings where hygiene practices may be inconsistent. Studies have shown that surfaces in retail environments, healthcare facilities, and public transportation harbor diverse microbial communities, including bacteria, fungi, and viruses (Otter *et al*., 2011). POS machines, used in retail, banking, and hospitality, are high-touch surfaces due to their frequent use by customers and staff. Unlike ATMs, which have been studied for microbial contamination, POS machines have received limited attention. However, their design featuring touchscreens, keypads, and card slots, makes them comparable to other electronic devices prone to contamination. A study by (Dawodu and Akanbi, 2021) conducted on ATMs at Federal Polytechnic Ede, Nigeria, found high levels of *Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa, and Bacillus* spp suggesting that similar

contamination may occur on POS machines due to their shared characteristics as public-facing electronic interfaces. Similarly, a study on currency notes in Keffi Metropolis, Nigeria, identified *Staphylococcus aureus, Escherichia coli* and *Klebsiella* spp. as dominant contaminants, attributing their presence to frequent handling and poor storage conditions (Ngwai *et al*., 2011). Given the similarity between ATMs and POS machines as high-touch electronic interfaces, these findings suggest that POS machines in Uselu and Textile Mill Road Junction may harbor similar pathogens, necessitating targeted sampling protocols.

In the context of Nigeria, research on public surfaces has primarily focused on food-related environments, ATMs and healthcare settings. For example, a study by (Umeanaeto *et al*., 2022) investigated bacterial and parasitic contamination of sachet water sold in Ngwo, Enugu State, Nigeria, identifying pathogens such as *Escherichia coli* and *Salmonella* spp. These studies highlight the prevalence of pathogenic microbes on frequently handled items in Nigeria, suggesting that POS machines in busy commercial areas like Uselu to Textile Mill Junction could similarly harbor harmful microorganisms.

2.2 MICROBIAL CONTAMINATION OF ELECTRONIC PAYMENT DEVICES

The wide acceptance of the advancement in banking technology has led to new public health challenges in the banking system due to the use of plastic cards which enables hand transmission of pathogenic microorganisms through dermal contact (Okoro *et al*., 2018). A study by (Tekerekoglu *et al*., 2011) on ATMs found high levels of *Staphylococcus aureus* and *Escherichia coli*, suggesting that similar contamination may occur on POS machines due to their shared characteristics as public-facing electronic interfaces. Electronic payment devices, such as Point-of-sales (POS) and automated teller

machines (ATMs), are particularly susceptible to microbial contamination due to their frequent use by individuals with varying hygiene practices. A seminal study by (Dawodu and Akanbi, 2021) conducted at Federal Polytechnic Ede, Osun State, Nigeria, investigated microbial contamination of ATMs, identifying bacteria such as *E. coli, Staphylococcus aureus, Pseudomonas aeruginosa, Klebsiella* spp*, Salmonella* spp*.,* and *Serratia* spp. on ATM screens, buttons, and surrounding surfaces. The study emphasized that the hand, as the primary contact point, acts as a "vault" for microorganisms, transferring them to ATM surfaces. This is particularly relevant to POS machines, which are similarly handled by multiple users in quick succession.

Internationally, studies on electronic payment devices corroborate these findings. For instance, a study by (Abban and Thano Debbra, 2011), in Ghana identified ATMs as potential sources of foodborne pathogens, including *Staphylococcus aureus* and *E. coli,* due to their frequent contact with unwashed hands. Another study by (Hessling *et al.*, 2021) reviewed microbial contamination of touchscreens, recommending ultraviolet disinfection to reduce microbial loads. These studies highlight the global concern regarding electronic payment devices as fomites, particularly in developing countries where hygiene practices may be inconsistent.

In Nigeria, the microbial contamination of POS machines specifically has received limited attention. However, a study by (Onuoha and Fatokun, 2014) in Ebonyi State, Nigeria, on ATM contamination found high levels of *Staphylococcus aureus* and other pathogens, suggesting that similar risks may apply to POS machines. The lack of specific studies on POS machines in Nigeria, particularly in commercial hubs like Benin City, underscores the need for localized research to assess microbial risks in these settings.

2.3 HEALTH IMPLICATIONS OF MICROBIAL CONTAMINATION

Fomites play a critical role in the transmission of infectious diseases by facilitating the transfer of microbes from surfaces to human hands and subsequently to mucous membranes (Boone and Gerba, 2007). The presence of pathogenic microorganisms on POS machines poses significant public health risks, including skin infections, gastrointestinal diseases, and respiratory illnesses etc. *Staphylococcus aureus,* commonly identified on shared surfaces, is one the most common bacterial infections in humans and are the causative agents of multiple human infections, including bacteremia, infective endocarditis, skin and soft tissue infections (e.g., impetigo, folliculitis, furuncles, carbuncles, cellulitis, scalded skin syndrome, and others), osteomyelitis, septic arthritis, prosthetic device infections, pulmonary infections (e.g., pneumonia and empyema), gastroenteritis, meningitis, toxic shock syndrome, and urinary tract infections and can lead to severe conditions like methicillin-resistant *Staphylococcus aureus* (MRSA) infections (Tony *et al.,* 2015). Similarly, *E. coli* and *Salmonella spp*. are associated with gastrointestinal infections, which can be transmitted through hand-to-mouth contact after using contaminated POS machines (Umeanaeto *et al.,* 2022).

In Nigeria, the public health implications are exacerbated by limited access to hand hygiene facilities and low awareness of microbial risks associated with shared devices. A study by (Tunio *et al*., 2025) in Pakistan emphasized the importance of hand hygiene interventions to reduce microbial transmission in public settings, a recommendation applicable to Nigeria where hand-washing facilities near POS terminals are often absent. Furthermore, the high prevalence of antibiotic-resistant bacteria, as noted in studies by (Kramer *et al.*, 2006) and others, increases the risk of untreatable infections from contaminated surfaces.

2.4 FACTORS INFLUENCING MICROBIAL CONTAMINATION

Several factors contribute to the microbial contamination of POS machines, including user hygiene, environmental conditions, and cleaning practices. In tropical environments like Benin City, high humidity and temperatures provide favorable conditions for microbial growth, warm, moist environments support bacterial and fungal proliferation. Additionally, the lack of regular cleaning of POS machines, as observed in studies of ATMs in Nigeria, contributes to microbial persistence (Dawodu and Akanbi, 2021). User behavior, such as touching POS machines with unwashed hands or during peak transaction periods, further increases contamination risks, as demonstrated in the Federal Polytechnic Ede study, which found higher bacterial counts during peak afternoon periods.

Socioeconomic factors also play a role. In Nigeria, the widespread use of POS machines in informal markets, such as those along Uselu to Textile Mill Junction, means that devices are handled by diverse populations, including traders and customers with varying hygiene practices. This is similar to findings from a study on shared sanitation facilities in Ghana, which noted that high user traffic increases microbial contamination (Obeng *et al*., 2023). These factors highlight the need for context-specific studies in Nigeria to understand the unique environmental and social dynamics influencing POS machine contaminate.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 STUDY AREA

The study was conducted along the Uselu to Textile Mill Junction corridor in an urban setting, as depicted in the provided map. This area features a dense network of commercial activities, including markets and residential zones, with 20 identified POS locations (POS 1 to POS 20) along the main route. The selection of this area was based on its high human traffic and frequent use of POS machines as alternatives to banking halls and Automated Teller Machines (ATMs), particularly due to the increasing demand for cashless transactions, which increases the likelihood of bacterial contamination due to hand contact.

Figure 3.1: A Map Showing the Location at which the study research was conducted (Uselu road to Textile mill junction, Benin City, Edo State)

3.2. SAMPLE COLLECTION

Twenty POS machines were used along the designated location of study. The designated

location of study was depicted as POS 1 to POS 20, POS 1 to POS 20 represent a range of locations along the Uselu to Textile Mill Junction route. The samples were collected between 10:00 AM and 4:00 PM on the particular day of sampling to capture peak usage. Sterile cotton swabs moistened with sterile peptone broth water for easy picking of bacteria, the swab stick were used to swab the keypads and screens of each POS machine for 30 seconds. Each swab was placed in a sterile tube and transported to the laboratory immediately after sampling was done to maintain sample integrity. Aseptic conditions were maintained before and after collection of samples to avoid cross-contamination.

3.3 PREPARATION OF MEDIA

Media for microbiological analysis were weighed according to the manufacturer's specifications.

Nutrient agar

This was used to culture non-fastidious organisms and for bacterial heterotrophic plate count. This medium was prepared from commercially available dehydrated powder, available from most suppliers of culture media. In the preparation, 28g of nutrient powder was dissolved in 1 litre of distilled water in a conical flask covered with cotton wool and aluminium foil paper. It was mixed thoroughly and sterilized by /autoclaving at 121 °C for 15 minutes. The medium was cooled to 45-50 °C and then dispensed aseptically into sterile Petri dishes.

3.4. MICROBIAL ISOLATION

Inoculation: In the laboratory, each swab was vortexed to dislodge microbes into the saline. The isolate was spread onto nutrient agar, potato dextrose agar, and plates using a sterile spreader. The plates were incubated at 37 °C for 24-48 hours.

Streak Plate Technique: To obtain pure cultures, the streak plate method was employed. A sterile inoculating loop was used to streak samples from the initial spread plate across four quadrants of a fresh agar plate, sterilizing the loop between quadrants to ensure isolated colonies.

Incubation: Plates were incubated agar-side up to prevent condensation from interfering with colony growth, checked after 24 hours, and monitored for up to 72 hours if no growth was observed.

3.5. ENUMERATION OF MICROORGANISMS

The method described by Holt *et al*; (2000) for estimating bacterial and fungal counts was used to enumerate the total viable counts of the isolates. The discrete colonies on the Nutrient agar was selected and counted. The mean colony count on the nutrient agar plates of each given dilution was used to estimate the total viable count for the samples in colony forming units per gram (cfu/g).

3.6. SUB-CULTURING OF BACTERIAL ISOLATES

A single isolated colony of the bacteria was picked up with the help of sterilized wire loop and was streaked on fresh nutrient agar medium. The nutrient agar plates were incubated at 37 °C for 24 hours. The isolated and purified bacterial strains were then characterized to know the genus of the bacteria isolate.

3.7. CULTURAL CHARACTERISTICS

For the bacterial isolates, cultural characteristics were observed on Nutrient agar plates. The cultural characteristics include. Size, shape, surface, opacity, texture, elevation and pigmentation were determined by visual observation

3.7.1. GRAM STAINING TEST

The Gram staining technique was used for differentiation between gram positive and gram negative bacterial strains according to Benson (1994). A drop of sterile distilled water was placed on a neat and clean glass slide, and a single isolated colony of 24 hours old culture was mixed in it. The smear was made by spreading the culture. This smear was air dried and fixed by rapidly passing the slide three times over the flame. It was then flooded with crystal violet for 1 minute and then washed off with distilled water. Then gram's iodine solution was added to the smear and the glass slide was left for one minute and rinsed with distilled water. This step was followed by the application of decolorizing agent (ethanol) for 30 seconds. Decolorizing agent was immediately washed with distilled water and the smear was counter stained with safaranine for one minute. The slide was washed with distilled water; air dried and was observed under the microscope.

3.7.2. SPORE STAINING TEST

The smear was prepared from the pure culture, air dried and fixed by passing on the flame. The smear was covered with malachite green (5%) solution for 3 - 4 minutes, by heating the slide with continuous steaming. Then, slide was washed with distilled water, air dried and examined under the microscope at 100x, oil immersion objective. The presence of green colored oval or spherical bodies indicated a positive results

3.7.3 COAGULASE TEST

A drop of normal saline was put on a slide; a colony of the test organism was emulsified in the saline. A drop of human plasma was added and mixed gently. After about 10 seconds it was observed for cloting (Cheesbrough, 2000).

3.7.4. CATALASE PRODUCTION TEST

One (1) ml of hydrogen peroxide solution was discharged into a clean glass slide and a sterile inoculating loop was used to collect the colonies of the test organism which were subsequently immersed in the hydrogen peroxide solution. A positive result was indicated by the production of gas bubbles, while its absence was regarded as a negative.

3.7.5. CITRATE UTILIZATION TEST

Five millilitres (5 ml) of Simmon citrate broth was inoculated with the test organism. The broth was incubated at 37 °C for 48 hours. A positive reaction was indicated by a change in the color of the medium from green to blue color. Negative tubes were observed daily for 4 days to detect any delayed reaction.

3.7.6. METHYL RED AND VOGES-PROSKAUER TESTS (MR-VP)

Five (5ml) of a 48 hours culture of the test organism was put into glucose phosphate medium and incubated at 37 °C for 48 hrs. The following specific tests were conducted as follows:

Methyl Red Test: Five drops of methyl red indicator was then cultured in a tube, mixed and the results were observed.

Voges-Proskauer test: One (1) ml of potassium hydroxide (KOH) and 3 ml of 5 % alpha-naphthol (Barrittes solution) were added to the culture and the reaction observed. It was then shaked and observed for color formation. A pink color within 2-5 minutes indicates a positive result.

3.7.7. INDOLE TEST

This was carried out according to the protocol outlined by Cheesbrough (2000). The test was inoculated into Bijou bottle containing 3ml of sterile peptone water, incubated at 35-37 °C for

48hours. This was followed by the addition of 0.5ml Kovac's reagent. Red color on the surface layer within 10mins indicated positive test for indole (Cheesbrough, 2000).

3.7.8. OXIDASE TEST

Filter paper (Whatman) was soaked with 2 drops of freshly prepared oxidase reagent. Colony of the test organism was smeared on the filter paper using sterile wire loop. Positive oxidase was indicated by the production of a deep purple/blue color within 10 seconds (Cheesbrough, 2000).

3.7.9. SUGAR FERMENTATION TEST

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

3.8 ANTIMICROBIAL SENSITIVITY BIOASSAY

The antimicrobial activity of the extract was determined by the agar disc diffusion technique delineated by (Cheesbrough, 2000). The tests were conducted with the authenticated pure cultures of the test pathogens to determine their respective tolerance to the extract. Sterile agar plates were aseptically inoculated with a loopful of the test pathogens. Each inoculum

was spread evenly over the surface of the agar plate as described by (Willey *et al*., 2008). With a flamed pair of forceps, the antimicrobial sensitivity discs prepared were embedded in the respective reconstituted extracts. Reconstitution of the dry extracts was achieved by mixing it with drops of sterile distilled water for the aqueous extract and ethanol for ethanolic extract to form a viscous paste. The mixture was left to stand for 3 h to allow the paper discs absorb the extract and was allowed to dry in the oven delineated by. The discs were carefully placed on the surface of the inoculated plates at a distance away from each disc to prevent over lapping, and allowed to stand for 5 min (to enable the extract permeate into the medium) before being incubated at 37 ºC for 24 hrs. The plates were observed for the presence of inhibition zones around the extract-impregnated discs. The extent of inhibition was determined by measuring the diameter of the inhibition zone using a transparent ½ meter rule. Measurements were made across the paper discs thus including its diameter. The mean zone of inhibition of the three replicated tests (triplicate analysis) of the plant extracts was expressed in millimeters. The discs were soaked/ impregnated with an equivalent volume of sterile distilled water and ethanol. This was used as a negative control.

3.8.1. ANTIBIOTIC DISC USED

Gram-positive and Gram negative (Optun laboratories, Nig. Ltd, Aba, Nigeria) antibiotics sensitivity disc was bought from the pharmaceutical shopping store. Antibiotic disc used and their concentrations were as follows: Gram positive discs contained; Ciprofloxacin (10 µg), Norfloxacin (10 µg), Gentamycin (10 µg), Lincocin (20 µg), Streptomycin (30 µg), Riflampicin (20 µg), Erythromycin (30 µg), chloramphenicol (30 µg), Ampiclox (20 µg) and Floxapen (20 µg). The Gram negative discs contain, Tarivid (10 µg), Peflacine (10 µg),

ciproflox (10 µg), Augumentin (30 µg), Gentamycin (10 µg), Stretomycin (30 µg), ceporex (10 µg), Nalidixic acid (30 µg), Septrin (30 µg) and Ampicilin (30 µg). The Gram positive disc was used on the culture of *Staphylococcus aureus* while Gram negative disc was used on *Escherichia coli*. The Antibiotic discs served as positive control.

CHAPTER FOUR

4.0 RESULT

A total of 20 swab samples were processed, yielding a range of bacterial isolates primarily from the genera *Micrococcus* spp, *Staphylococcus* spp, *Pseudomonas* spp*, Streptococcus* spp and *Bacillus* spp.

Table 4.1 represents results of POS locations showing different range of bacteria population. The result showed that location 1 had 5.5×10^3 cfu/ml, location 2 had 1.0×10^3 cfu/ml, location 3 had 2.2×10^3 cfu/ml, location 4 had 1.8×10^4 cfu/ml, location 5 had 1.0×10^3 cfu/ml, location 6 had 3.2×10^4 cfu/ml, location 7 had 2.8×10^4 cfu/ml, location 8 had 4.9×10^4 cfu/ml, location 9 had 5.0×10^2 cfu/ml, location 10 had 6.1×10^4 cfu/ml, location 11 had 6.5×10^4 cfu/ml, location 12 had 6.8×10^4 cfu/ml, location 13 had 5.0×10^2 cfu/ml, location 14 had 1.5×10^3 cfu/ml, location 15 had 2.2×10^4 cfu/ml, location 16 had 5.6×10^4 cfu/ml, location 17 had 8.1×10^4 cfu/ml, location 18 had 1.8×10^4 cfu/ml, location 19 had 1.0×10^3 cfu/ml, location 20 had 1.0×10^3 cfu/ml. The colony count results from POS swabs revealed significant microbial loads across the sampled machines, such as samples 12 and 17, showing particularly high values, indicating heavy contamination.

Table 4.2 represent the cultural and biochemical characterization of the suspected isolates. The suspected bacteria are *Micrococcus* spp, *Pseudomonas* spp, *Staphylococcus* spp, *Streptococcus* spp, and *Bacillus* spp.

In Table 4.3 shows the frequency of distribution of each bacterial isolates across the different POS location with *Micrococcus* spp and *Pseudomonas* spp appearing 13 times, *Staphylococcus* spp and *Streptococcus* spp appearing 11 times and *Bacillus* spp appearing 12

times. These findings collectively provide insight into the level of contamination, the predominant organisms present, and their level of contamination.

Table 4.4 shows the percentage occurrence of each bacterial isolates where *Micrococcus* spp and *Pseudomonas* spp had the highest occurrence with 21.67 %, *Staphylococcus* spp and *Streptococcus* spp had the lowest occurrence of 18.33 % each and *Bacillus* spp had an occurrence of 20 %.

Table 4.5 reports the findings of antibiotic susceptibility pattern of the bacterial isolates against some known antibiotics such as pefloxacin (PEF) ciprofloxacin (CPX, levofloxacin (LEV) ampicillin (AMP) etc. The antibiotic susceptibility patterns revealed varied responses among the bacterial isolates.

TABLE 4.1 Heterotrophic Bacteria Count of the POS Machines

S/N	cfu/ml
1.	5.5×10^3
2.	1.0×10^3
3.	2.2×10^3
4.	1.8×10^4
5.	1.0×10^3
6.	3.2×10^4
7.	2.8×10^4
8.	4.9×10^4
9.	5.0×10^2
10.	6.1×10^4
11.	6.5×10^3
12.	6.8×10^3
13.	5.0×10^2
14.	1.5×10^3
15.	2.2×10^4
16.	5.6×10^4
17.	8.1×10^4
18.	1.8×10^4
19.	1.0×10^3
20.	1.0×10^3

TABLE 4.2: Cultural and biochemical characterization of isolates

	A	B	C	D	E
Shape	Circular	Circular	Circular	Circular	Circular
Colour	Milky	Pale green	Orange	Greenish	Milky
Margin	Entire	Entire	Entire	Entire	Entire
Opacity	Opaque	Translucent	Opaque	Opaque	Opaque
Elevation	Flat	Flat	Flat	Flat	Raised
Wet/dry	Wet	Wet	Wet	Wet	Wet
Gram stain	+	+	+	-	+
Arrangement	Single	Cluster	Chain	Pair	Single
Catalase	+	+	-	+	+
Indole	-	-	+	-	-
Citrate	-	-	-	+	-
Oxolase	+	+	-	+	-
Cell shape	Cocci	Cocci	Cocci	Bacilli/Rod	Short rod
Spore	-	-	-	-	+
Glucose	+	+	+	-	-
Mannitol	-	+	+	-	+
Lactose	+	-	-	-	-
Suspected isolates	*Micrococcus* spp	*Staphylococcus* spp.	*Streptococcus* spp	*Pseudomonas* spp	*Bacillus* spp.

TABLE 4.3 Frequency Distribution of Bacterial Isolates

S/N	A	B	C	D	E
1.	√	X	X	X	X
2.	X	X	X	X	X
3.	X	X	X	√	X
4.	√	X	√	√	√
5.	X	X	X	X	X
6.	√	√	√	√	√
7.	√	√	√	√	√
8.	√	√	√	√	√
9.	X	X	X	X	X
10.	√	√	√	√	√
11.	√	√	√	√	√
12.	√	√	√	√	√
13.	X	√	X	√	√
14.	√	X	X	X	X
15.	√	√	√	√	√
16.	√	√	√	√	√
17.	√	√	√	√	√
18.	√	√	√	√	√
19.	X	X	X	X	X
20.	X	X	X	X	X

KEY:

A- *Micrococcus* spp

B- Staphylococcus spp

C- Streptococcus spp

D- Pseudomonas spp

E-Bacillus spp

TABLE 4.4 Percentage Occurrence of Bacterial Isolates

BACTERIAL ISOLATES	FREQUENCY	% OCCURRENCE
Micrococcus spp	13	21.67
Staphyllococcus spp	11	18.33
Streptococcus spp	11	18.33
Pseudomonas spp	13	21.67
Bacillus spp	12	20.00

KEY:

A- *Micrococcus* spp

B- Staphylococcus spp

C- Streptococcus spp

D- Pseudomonas spp

E-Bacillus spp

TABLE 4.5 Susceptibility pattern of the bacterial isolates

ANTIBIOTICS	A	B	C	D	E
Pefloxacin	S	S	S	S	R
Gentamycin	S	R	R	R	R
Ampiclox	R	R	R	R	ND
Zinnacef (Defuroxine)	R	R	R	R	ND
Amoxicillin	R	R	R	R	R
Rocephin (Ceftriaxone)	R	R	R	R	ND
Ciprofloxacin	S	S	R	S	R
Azithromycin	S	R	R	R	R
Levofloxacin	S	S	R	R	R
Erythromycin	S	S	R	R	ND
Ofloxacin	ND	ND	ND	ND	R
Cefalexin	ND	ND	ND	ND	R
Spiramycin	ND	ND	ND	ND	R
Augmentin (Amoxicillin+Clavulanic acid)	ND	ND	ND	ND	R

KEY:

A- *Micrococcus* spp

B- Staphylococcus spp

C- Streptococcus spp

D- Pseudomonas spp

E-Bacillus spp

CHAPTER FIVE

5.0 DISCUSSION

The study revealed that POS keypads collected within the Uselu to Texile mill were contaminated with a variety of bacterial species. This finding indicates that POS machines, which are widely used in commercial transactions, can serve as potential reservoirs and vehicles for the transmission of pathogenic and opportunistic microorganisms due to their frequent handling by multiple users (Okoro *et al*., 2018).

The result in Table 4.1 revealed the heterotrophic bacteria count ranging from 5.0×10^2 to 8.1×10^4 cfu/ml, with samples 9 and 17 exhibiting particularly high microbial loads. These findings indicate significant contamination levels, likely influenced by factors such as usage frequency, hygiene practices, and environmental exposure. The high microbial loads on the sampled POS machines are consistent with previous studies on fomite-associated contamination, highlighting the public health implications of POS machines as fomites. Fomites are inanimate objects capable of transmitting infectious agents, and their role in the spread of pathogens in community settings is well-documented (Boone and Gerba, 2007). For instance, Rusin *et al*. (2002) reported that surfaces in public spaces, such as ATMs and POS terminals, can harbor significant microbial populations due to repeated handling by multiple individuals. The variation in colony-forming units (cfu) across samples suggests that some POS machines, particularly those in high-traffic areas, are more heavily contaminated, posing a greater risk for microbial transmission.

The frequent handling of POS machines by both operators and customers, often without proper hand hygiene, creates an ideal environment for microbial transfer. The methodology

employed, which included pour plate culture technique and serial dilutions, ensured that viable microorganisms were adequately quantified. The findings corroborate previous reports by Effiong *et al*. (2024), who observed high microbial counts on frequently touched devices in open markets. The high bacterial load detected on POS machines in this study suggests that such devices can serve as reservoirs for pathogens and facilitate their easy transmission, especially in crowded markets where people have varying hygiene practices. The findings suggest that these devices could contribute to the dissemination of both pathogenic and opportunistic microorganisms, potentially leading to infections, especially among immune-compromised individuals (Marbel *et al*., 2014).

The result in Table 4.2 revealed the following bacteria isolated from the swab samples included *Micrococcus* spp, *Staphylococcus* spp, *Pseudomonas* spp, *Streptococcus* spp, and *Bacillus* spp. These bacterial genera are commonly associated with the skin, respiratory tract, and environment, suggesting both human and environmental sources of contamination. The cultural and biochemical characterization of bacterial isolates (Table 4.2) revealed a diversity of colony morphologies, with all isolates exhibiting circular shapes but varying in color, elevation, and margin. The predominance of milky, pale green, orange, and grayish colonies, along with flat or raised elevations and entire margins, aligns with the characteristics of *Staphylococcus* spp., *Micrococcus* spp., and *Bacillus* spp. These morphological variations indicate that POS machines serve as reservoirs for multiple bacterial species, likely introduced through frequent human contact and environmental exposure.

The high level of contamination observed may be attributed to several factors, including poor hand-washing, practices, exposure to dust and environmental pollutants, and the absence of

routine disinfection of POS machines. Since these devices are constantly exchanged between cashiers and customers in markets, shops, and other crowded areas, they are easily contaminated with bacteria from human skin, money, air, and surrounding surfaces. ATM and POS machines once contaminated becomes vehicles for the transmission of infection, such that the user may succeed in picking these pathogens after making use of the device , since there is no restriction as to who has access to the facility, and no guidelines to ensure hygienic usage (Onuoha and Kayode, 2014).

The high level of microbial load seen in this study which includes: Micrococcus spp *pseudomonas* spp, *staphylococcus aureus*, *streptococcus* spp, *bacillus* spp, is in line with the study of Fraser and Girling (2009), who reported that keyboards of ATM harbored more bacteria than computer keyboards and this may be due to the fact that they are exposed to many users, environmental factors such as rain and climatic factors such as wind. The study is in conformity with Anastasiades *et al*. (2009) who reported that *Staphylococcus aureus* is prevalent in the environment.

The frequent isolation of *Staphylococcus aureus* from POS devices in this study is not unexpected, as it is a common microorganism found on human skin (Hardy *et al*., 2006). Around 20–40 % of healthy individuals carry *Staphylococcus aureus* at any given time. However, its presence should not be overlooked, given that *Staphylococcus aureus* is a major human pathogen responsible for a range of infections including boils, abscesses, wound infections, and pneumonia. The increasing occurrence of Methicillin-Resistant *Staphylococcus aureus* (MRSA) further raises concern. Although typically a normal skin

resident, *Staphylococcus aureus* is also known to cause serious conditions like endocarditis and other infections, particularly in immune-compromised individuals (Willey, 2008).

The isolation and detection of *Bacillus* species from the surfaces of Point-of-Sale (POS) keypads indicate that these devices can serve as potential reservoirs of environmental microorganisms. *Bacillus* spp are a diverse group of Gram-positive, rod-shaped, aerobic or facultatively anaerobic bacteria that are widely distributed in nature. They are commonly found in soil, dust, water, air, and on various inanimate objects (Prescott *et al.*, 2008). Their detection on POS keypads is not unexpected, considering that these devices are frequently handled by multiple users throughout the day and are often exposed to open air and dust particles in commercial environments.

One of the most important features of *Bacillus* spp. is their ability to form endospores, which are resistant dormant structures that enable them to withstand heat, desiccation, ultraviolet radiation, and chemical disinfectants (Cappuccino and Sherman, 2014). This spore-forming capability allows *Bacillus* cells to persist for extended periods on dry and hard surfaces such as plastic or rubber keypad materials used in POS machines. Consequently, even with occasional cleaning, *Bacillus* spores can survive and recolonize the surface once conditions become favourable.

Micrococcus species are common inhabitants of water, soil and dust. They are also found as commensals on the human skin where they break down compounds in sweat to generate unpleasant smell (Smith, 1999) The occurrence of *Micrococcus* in these environments explain its presence on POS machines, naira notes etc. which contact human skin, soil, dust and water

of doubtful source from time to time. Micrococcus is another opportunistic pathogen of man. From the foregoing discussion, it is obvious that bacteria isolated from naira notes in this study are known to be involved in causing human health challenges.

The result in table 4.3 and 4.4 revealed the frequency distribution and percentage occurrence of the bacteria. The study revealed that *Micrococcus* spp. and *Pseudomonas* spp. were the most prevalent, each accounting for 21.67 % of isolates, followed by *Bacillus* spp. (20 %), *Streptococcus* spp. (18.33 %), and *Staphylococcus* spp. (18.33%). The predominance of Gram-positive bacteria (*Micrococcus spp*., *Staphylococcus* spp., *Streptococcus spp*., and *Bacillus* spp.) is consistent with their ability to survive on dry surfaces and their association with human skin flora (Grice and Segre, 2011). The equal distribution of *Pseudomonas* spp., a Gram-negative bacterium, is notable, as it suggests environmental contamination in addition to human sources. *Pseudomonas* spp. are ubiquitous in soil, water, and moist environments, and their presence on POS machines may indicate exposure to contaminated hands or surfaces (Lister *et al*., 2009). The frequent handling of POS machines by multiple individuals likely facilitates the transfer of these organisms, contributing to their high prevalence. The comparison of contamination across different POS locations showed that microbial contamination was widespread, as bacterial growth was observed from all sampled sites. Although some variations in counts existed, the overall findings indicated that both Uselu and Texile mill POS terminals were similarly contaminated. This observation implies that contamination is not limited to specific locations but is a general problem in areas with high human interaction and limited sanitization practices. Similar observations have been made by Mbajiuka *et al*. (2015) who reported that the level of contamination of public

devices was strongly linked to the frequency of use rather than geographical differences. The results from this study further emphasize the public health risks posed by high-touch devices in densely populated commercial centers. The frequent occurrence of *Micrococcus* spp. and *Pseudomonas* spp. (21.67 % each) and the relatively high occurrence of *Bacillus* spp. (20 %) further corroborate their ubiquitous nature and adaptability to different environmental conditions (Iquo *et al*., 2015). The presence of Gram-positive bacteria, such as *Micrococcus* spp. (21.67 %) and *Staphylococcus* spp. (18.33 %), as shown in Table 4.4, is consistent with their ability to survive on dry surfaces and human skin (Otto, 2008). *Micrococcus* spp. and *Staphylococcus* spp. are known to thrive in environments with high human traffic due to their resilience to desiccation and ability to adhere to surfaces (Kloos and Musselwhite, 1975; Jaradat *et al.,* 2020). Similarly, *Bacillus* spp. (20 %), known for their spore-forming capabilities, can persist on inanimate surfaces for extended periods, contributing to their prevalence on POS machines (Nicholson *et al*., 2000; Enger *et al.,* 2018)

The identification of *Pseudomonas* spp. (21.67 %) as one of the most frequent isolates is particularly concerning due to its opportunistic pathogenic nature and intrinsic resistance to many antibiotics (Lister *et al*., 2009). The presence of *Streptococcus* spp. (18.33 %) further suggests potential health risks, as certain species within this genus are associated with respiratory and skin infections (Marks *et al*., 2014). The diversity of isolates reflects the complex microbial ecology of frequently handled surfaces and emphasizes the need for regular cleaning and disinfection protocols to mitigate transmission risks.

The antibiogram presented in Table 4.5 reveals the varying degrees of antimicrobial susceptibility and resistance profiles of bacterial isolates recovered from the surfaces of

Point-of-Sale (POS) machines. The *Micrococcus spp*. isolate (A) exhibited susceptibility to fluoroquinolones such as pefloxacin, ciprofloxacin, and levofloxacin, as well as macrolides like azithromycin and erythromycin, but showed resistance to several β-lactam antibiotics, including ampiclox, cefuroxime (Zinnacef), amoxicillin, and ceftriaxone. This resistance pattern aligns with the known characteristics of *Micrococcus* spp., which are resilient environmental organisms capable of producing β-lactamase enzymes that degrade β-lactam antibiotics (Kloos and Musselwhite, 1975). Although Micrococcus is generally non-pathogenic, its resistance to commonly used antibiotics suggests its potential role as a reservoir for antibiotic resistance genes that may be transferred to more pathogenic bacteria under favorable conditions.

The *Staphylococcus* spp. isolate (B) demonstrated a similar pattern of broad resistance to β-lactam antibiotics, including ampiclox, cefuroxime, amoxicillin, and ceftriaxone, but remained sensitive to some fluoroquinolones (pefloxacin and levofloxacin) and erythromycin. This finding is consistent with the known behavior of *Staphylococcus* aureus and related species, which frequently develop resistance through the production of penicillin-binding protein 2a (PBP2a) encoded by the mecA gene, conferring resistance to β-lactams and leading to methicillin-resistant *S. aureus* (MRSA) strains (Chambers and DeLeo, 2009). The persistence of such resistant S*taphylococcus* strains on POS surfaces is a public health concern because these devices are handled by numerous individuals daily, facilitating the potential spread of resistant pathogens (Kramer *et al*., 2006).

The *Streptococcus* spp. isolate (C) exhibited multidrug resistance, particularly to β-lactam antibiotics and macrolides such as azithromycin and erythromycin, but retained sensitivity to

pefloxacin. Although streptococci are traditionally sensitive to penicillin and other β-lactams, emerging reports indicate the occurrence of resistance among environmental and clinical isolates, often due to alterations in penicillin-binding proteins or efflux mechanisms (Fiedler *et al*., 2013). The resistance of Streptococcus isolates in this study suggests environmental adaptation and possible horizontal gene transfer in response to selective pressure from antibiotic misuse in the community.

The *Pseudomonas* spp. isolate (D) demonstrated high levels of resistance to multiple antibiotics, including ampiclox, cefuroxime, amoxicillin, ceftriaxone, azithromycin, and levofloxacin, but showed susceptibility to pefloxacin and ciprofloxacin. This resistance profile is characteristic of *Pseudomonas aeruginosa*, which possesses intrinsic and acquired mechanisms of resistance, including efflux pumps, low outer-membrane permeability, and the production of β-lactamases (Hancock and Speert, 2000; Lister *et al*., 2009). The recovery of multidrug-resistant Pseudomonas from POS machines is alarming, as such bacteria can cause severe opportunistic infections, particularly in immune-compromised individuals, and contribute to environmental reservoirs of antimicrobial resistance.

The *Bacillus* spp. isolate (E) displayed resistance to most of the antibiotics tested, including β-lactams and fluoroquinolones. This resistance is likely linked to the ability of *Bacillus* species to form endospores, which confer high resistance to adverse environmental conditions, including desiccation, heat, and antimicrobial agents (Nicholson *et al*., 2000). While *Bacillus* species are primarily environmental organisms, certain strains such as *B. cereus* are opportunistic pathogens capable of causing foodborne illnesses and nosocomial infections. The resistance pattern observed in this isolate underscores the persistence of spore-forming

bacteria on inanimate surfaces and their potential role in the spread of antimicrobial resistance genes.

Overall, the antibiogram indicates a widespread pattern of resistance among the bacterial isolates, particularly to β-lactam antibiotics such as ampicillin, amoxicillin, and ceftriaxone. This resistance may be attributed to the overuse and misuse of antibiotics in the community, which imposes selective pressure favoring the survival of resistant strains (O'Neill, 2016). The partial sensitivity of some isolates to fluoroquinolones such as pefloxacin and ciprofloxacin is encouraging, suggesting that these antibiotics may still be effective for certain bacterial populations. However, the emergence of resistant strains, especially among *Staphylococcus* and *Pseudomonas,* calls for caution and reinforces the need for prudent antibiotic use.

From a public health perspective, the presence of antibiotic-resistant bacteria on POS machines highlights their potential role as fomites in the dissemination of antimicrobial resistance. The World Health Organization (2023) has identified antimicrobial resistance as one of the greatest threats to global health, emphasizing the importance of hygiene interventions and environmental disinfection. Simple measures such as wiping POS terminals with alcohol-based disinfectants have been shown to significantly reduce microbial contamination (Kramer *et al.*, 2006). Furthermore, promoting hand hygiene among users and operators through the use of alcohol-based sanitizers or regular hand washing can help minimize microbial transmission (Curtis and Cairncross, 2003).

CONCLUSION

The microbiological analysis of POS machines along the Uselu to Textile Mill Junction in Benin City revealed significant bacterial contamination, with *Micrococcus* spp., *Pseudomonas* spp., *Bacillus* spp., *Streptococcus* spp., and *Staphylococcus* spp. being the predominant isolates. The high microbial loads and presence of antibiotic-resistant strains highlight the potential of POS machines as fomites for microbial transmission and the dissemination of antimicrobial resistance. These findings underscore the need for improved hygiene practices, regular disinfection of POS machines, rational antibiotic use and public awareness campaigns to promote hand hygiene and to curb the spread of antimicrobial resistance within the community. By addressing these issues, public health authorities can mitigate the risks associated with fomite-mediated infections and contribute to the global fight against antimicrobial resistance.

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Appendix 1

Plate 1: Point of sale machine

Appendix 2

Plate 2: Culture slants

Appendix 3

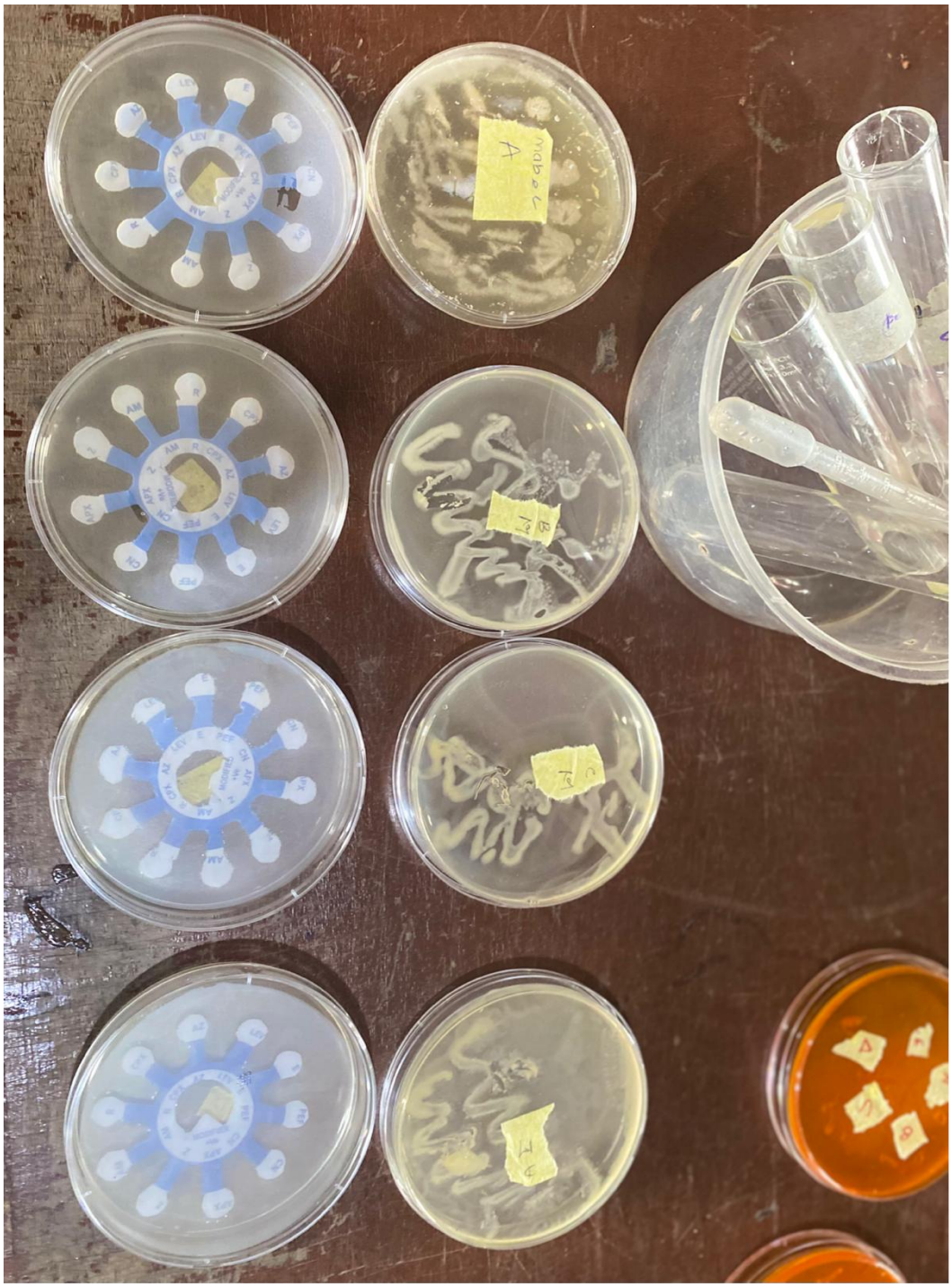

Plate 3: Antibiogram analysis of bacteria isolates.

Appendix 4

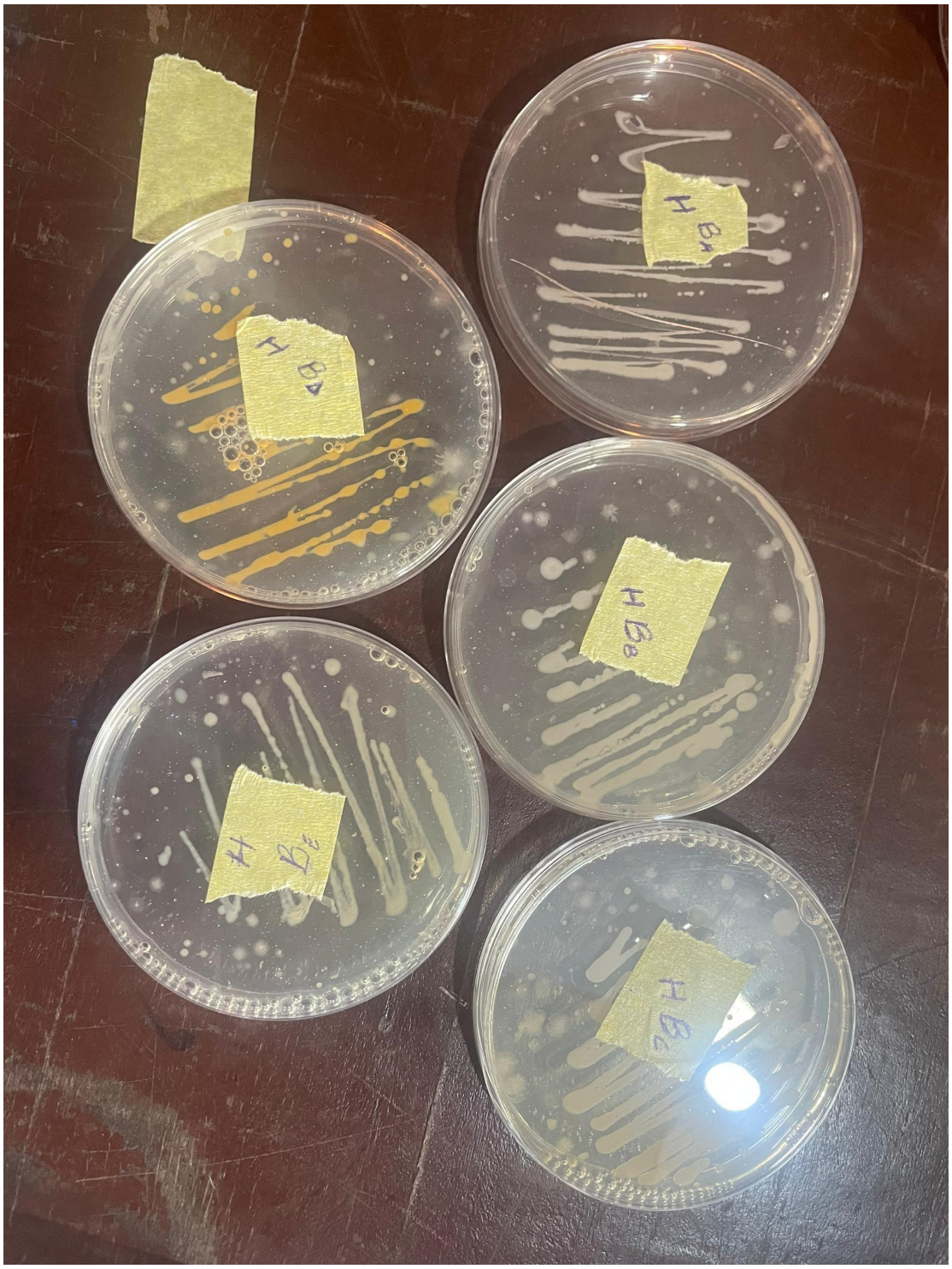

Plate 4: Picture of pure culture plates.

Appendix 5

Plate 5: Picture of experimental analysis in the laboratory.