PREVALENCE OF EXTENDED-SPECTRUM-BETA-LACTAMASE-PRODUCING BACTERIAL IN HANDBAGS OF WOMEN IN ABEOKUTA, NIGERIA.

ABSTRACT

BACKGROUND: Multiple studies have revealed information about living, harmful microorganisms on inanimate objects. A woman handbag, which serves as a flexible private device, frequently harbors an array of microbes, including bacteria. This study aims to determine the prevalence, antibiogram, and dispersal of Extended Spectrum Beta Lactamase-producing isolates recovered from females' handbags, as well as their sensitivity and resistance pattern to selected common antibiotics in Abeokuta Nigeria.

Methods: This cross-sectional research studied a total of 300 samples recovered from various female handbags. Antibiotic susceptibility testing was done using the Kirby-Bauer disc diffusion technique on all the isolates and Extended Spectrum Beta-Lactamase was detected using the double disc synergy test on isolates that showed resistance to standard antibiotics.

Results: A total of 300 samples were collected, (59%). Of the samples yielded no growth, while (41%). Yielded bacteria growth. The prevalence of ESBL-producing bacteria showed that *Escherichia coli* had the highest production of ESBL, followed by *Klebsiella pneumoniae* (56.3%). while *Pseudomonas aeruginous* had the least production of Extended-Spectrum-Beta-Lactamase with (1.49%). Ceftazidime (19%). Was the most active antimicrobial agent, followed by Cefotaxime (13%). Augmentin (6%). was the least active specifically against ESBL-producing bacteria. Generally against all the isolates, Azithromycin (77.8%). Was the most active antibiotic, while Ceftriaxone (33.3%). Was the least active.

Conclusion: Conclusively, Women handbags, accommodate various multidrug-resistant bacteria, and serve as media for the transmission of pathogenic bacteria to their users. Therefore, awareness should be raised to educate females on this potential route of disease transmission to curb the spread of multi-drug-resistant organisms.

Keywords: Extended-Spectrum-Beta-Lactamase, Bacterial, Handbags, Cross infections, Antibiotic resistance, *Escherichia coli*, *Klebsiella pneumonia*, *Pseudomonas aerugionsa*, Women, Nigeria

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INTRODUCTION

Microorganisms are ubiquitous. Multiple studies have revealed information about living, harmful microorganisms on inanimate objects. Numerous studies have shown that items such as doorknobs, cell phones, money, and so on become colonized and contaminated (1). Eighty percent of diseases are passed on via tandem interaction with the hands of others or items (2). A fomite is an inanimate thing or material that may pass on pathogenic organisms to humans (3). Moisture, regular use, and overall cleanliness can all have an impact on fomite infection rates. According to (4), *Staphylococcus aureus, Escherichia coli, and Klebsiella spp.*, among others, were found to be infecting numerous surfaces, such as furniture, Figures, openings, hinges on doors, and a lot more. Such surroundings and objects, mostly those that get touched frequently and located near individuals, constitute a danger to human health and should be carefully taken into consideration.

A woman handbag, which serves as a flexible private device, frequently harbors an array of microbes, including bacteria. Numerous researchers discovered live, harmful germs on inanimate objects (2). Females' handbags, due to their frequent usage, may provide a perfect atmosphere for the growth of bacteria. These bags have been reported to contain mobile phones, cosmetics such as face creams, lip gloss, and powder, partially eaten food, fresh or used diapers, and milk bottles in the case of nursing mothers (1). Handbags are frequently stored in germ-infested areas such as restroom countertops, fast food counters, and kitchen Figures. As a result, handbags are highly susceptible to infection and may serve as conduits for disease transmission. Bacteria have been found in the purses and handbags of healthcare workers in healthcare facilities (5). Dust can sometimes be evacuated inside handbags by wiping them down with a damp cloth. If a handbag is damp, it will be left open overnight to dry and then used in the morning. Most women do not use natural methods of sunlight sterilization, which may slow the growth of bacteria in handbags (6).

Extended-spectrum beta-lactamases (ESBL) are enzymes that hydrolyze oxyimino-beta-lactam antibiotics, which are important therapeutic medicines for treatment of severe human and animal infections. ESBL production is common in *E.coli* and *Klebsiella pneumoniae* but were first discovered in 1983 in *Enterobacteriaceae*, and since then, scientific research has revealed that ESBL-producing *Enterobacteriaceae* (E-ESBL) pose major concern and threat to human health, accounting for approximately 1700 deaths in the United States due to therapeutic failure in severe infections in 2013 (28,31). The World Health Organization identified ESBL-producing Enterobacteriaceae as one of the "Highest Priority" pathogens in a list published by them (32).

Additionally, *Escherichia coli*, which produces extended-spectrum β-lactamase (ESBL), is a growing global pathogen Unlike most other antimicrobial-resistant bacteria that are associated with admission to hospitals (29). *E. coli*, a Gram-negative facultative anaerobe found primarily in the distal intestines of people and animals, is the leading cause of urinary tract infections and urosepsis in humans. *E. coli* has acquired antibiotic resistance genes, making treatment of these infections problematic. ESBL-producing *E. coli* are resistant to many β-lactam antibiotics, such as penicillins, aztreonam, and cephalosporins (30). The clinical impact of infections caused by ESBL-producing *E. coli* strains has primarily been studied in hospitalized patients. These infections have a greater mortality rate associated with a delay in administering an appropriate antimicrobial therapy, as empirically given antibiotics may not be effective in this scenario (28). By breaking down these antibiotics, ESBL produces beta-lactamase, which may produce resistance to penicillins, first-, second-, and third-generation cephalosporins, and aztreonam. Beta-lactamase inhibitors, like clavulanic acid inhibit these enzymes (7). Extended-spectrum penicillins, cephalosporins, and aztreonam are blocked by -lactamase inhibitors such as clavulanic acid, but not cephamycins or carbapenems, are rendered ineffective by ESBL (8).

Researchers found that 454 out of 572 mobile phones contained Gram-negative bacteria (79.4%). Women's mobile phones were more frequently affected (82.3%, or 275 phones) than men's (75.2%). 179 mobile phones (9). Female handbags are an important reservoir of bacterial contamination, which can cause serious problems. It is critical to understand the prevalence of ESBL-producing bacteria, as well as their susceptibility to certain conventional antibiotics (8).This study aims to determine the prevalence of Extended Spectrum Beta Lactamase (ESBL)

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bacterial in clinical isolates recovered from females' handbags in Abeokuta, Nigeria, as well as their sensitivity pattern and resistance pattern to some selected common antibiotics, to promote greater awareness of microbial contaminants, especially those with the potential for multidrug resistance infection.

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MATERIALS AND METHODS

The current cross-sectional study was carried out in June 2023 at the Federal Medical Centre Abeokuta in Nigeria. Participant were given a questionnaire containing Specific questions about the materials of their handbags, usage, and storage of their handbags, their house environments, items being left in the bags most of the time, and the frequency of washing, cleaning, and airing of their handbags. These data were used descriptively and inferentially in the analysis. A total of 300 samples were collected from various female handbags using saline-soaked swabs. The Medical Microbiology Laboratory at the Federal Medical Center in Abeokuta received these samples for culturing and susceptibility testing. The isolates were identified using the standard microbiological technique described by (Aflakian et al., 2022). including Colonial Morphology, wet preparation, Gram Stain, Indole Test, Simmons Citrate Test, Christensen's Urease Test, Oxidase Test, Catalase Test, Coagulase test, Methyl red, Voges-Proskauer test, Triple Sugar Iron test and Motility Test. All isolates were kept at -70°C in trypticase soy broth with 15% (v/v). Glycerol for 6 months. The culture media used for culturing and identification include MacConkey agar, blood agar, and Muller Hilton Agar. The counting of viable colonies was done manually by examining the plats under suiFigure lightning. Antibiotic sensitivity testing was performed using the Kirby-Bauer disc diffusion technique as recommended by (CLSI 2020). for the following disks: Cefuroxime (30 µg), Ofloxacin(5 µg), Erythromycin (15 µg), Azithromycin(15 µg), Ceftriaxone (30ug), Cefixime, (5 µg), Levofloxacin, (5 µg) Ciprofloxacin(5 µg), and 30ugGentamicin(10 µg), and Amoxicillin-clavulanate. (30 µg) The presence of ESBL in all isolates was detected using the double disc synergy test, as described by Livermore and Brown (2001). using 30ug augmentin and 30ug ceftazidime.

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STATISTICAL ANALYSIS

The data were analyzed using the statistical software INSTAT (Graph pad Software Inc, La Jolla, CA, USA) with chi-square and odd ratio analyses. The results are presented as descriptive statistics based on relative frequency. Continuous variables were expressed as mean ± SD, while categorical variables were represented as group percentages. P≤ 0.05 was considered statistically significant.

RESULTS

The findings from this study are presented in Figures 1-9. 178 (59%). samples showed no growth, while 122 (41%). showed bacteria growth. This high percentage of samples with no bacterial growth may suggest that the majority of women practice safe hygiene concerning their handbags. This can effectively reduce microbial contamination and mitigate health risks (4). It could also be a result of environmental factors, such as the materials used in making the handbags or the atmospheres under which they are stored. Certain materials may inhibit bacterial survival and growth, suggesting that women should consider these factors when selecting handbags (2). Figure 1 shows a significant prevalence of bacteria microbes in female handbags

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(P<0.0001).

Figure 2 shows that *Escherichia coli* had the highest percentage occurrence, followed by *Bacillus species, Staphylococcus aureus, Klebsiella pneumonia, Acinetobacter sp.*, and *Pseudomonas aerurginosa*. The implication of this is important for understanding the clinical implications of bacterial infections. The presence of ESBL-producing bacteria, such as *Escherichia coli and Klebsiella pneumonia*, indicates a significant concern for public health due to their ability to confer resistance to a wide range of beta-lactam antibiotics, including penicillins and cephalosporins (1, 3). The resistance patterns observed in the study suggest that infections caused by these resistant strains may lead to prolonged illness, increased healthcare costs, and a higher risk of treatment failure. Clinicians must be aware of local resistance patterns to make informed decisions regarding empirical therapy (4).

Figure 3 shows the prevalence of bacteria isolated from different types of handbag materials. The findings indicate that leather bags had the highest rate of bacteria contamination, while synthetic bags had the lowest rate. (Figure 4) The prevalence of bacteria isolated by usage duration; the results showed that frequently used handbags had the highest contamination percentage, compared to rarely used bags.

Figure 5 revealed that students' handbags with never-emptied contents had a higher percentage of contamination, while those that were frequently emptied had bacteria contamination as well.

Figure 6 Handbags stored in lockers had the highest bacteria contamination, followed by those stored on bunk beds, Figures, and wall nails.

Figure 7 shows the prevalence of ESBL-producing bacteria. *Escherichia coli* had the highest production, followed by *Klebsiella pneumoniae* (56.3%), and *Pseudomonas aeruginosa* had the lowest production (1.49%). The prevalence of ESBL-producing isolates was not significantly different (P = 0.3011).

Ceftazidime was the most active antimicrobial against ESBL-producing bacteria. Tab 8 showed that Cefotaxime and Augmentin were the least effective. Figure 9 shows the antibiotic susceptibility profile of the bacterial isolates from female student handbags. The most active antibacterial agent was azithromycin (77.8%), while Ceftriaxone (33.3%) was ineffective against the various isolates. This procedure was performed in vitro using standard laboratory procedures.

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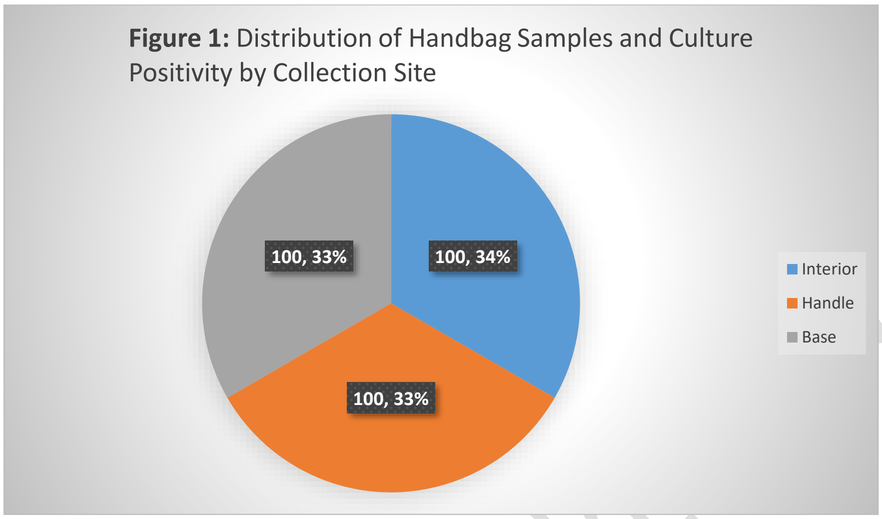

Figure 1: Distribution of Handbag Samples and Culture Positivity by Collection Site

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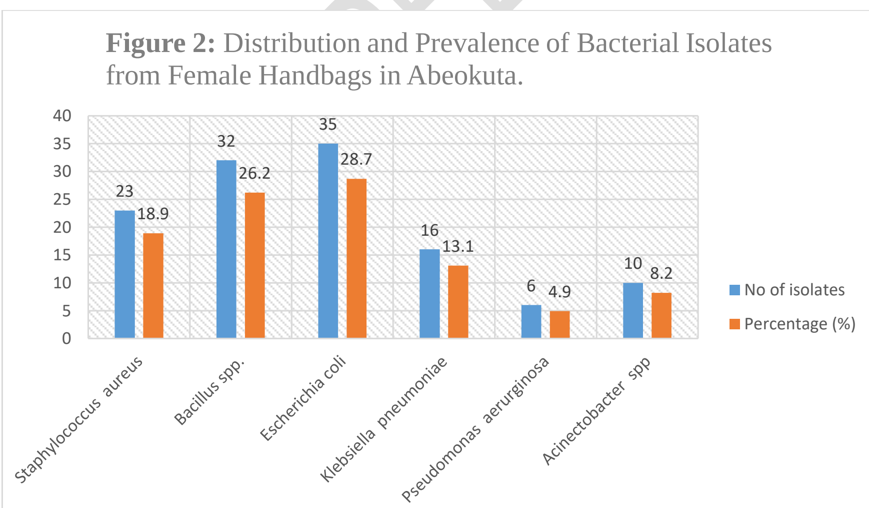


Figure 2: Distribution and Prevalence of Bacterial Isolates from Female Handbags in Abeokuta.

Figure 3: Prevalence of Bacterial Isolates According to Handbag Material Type

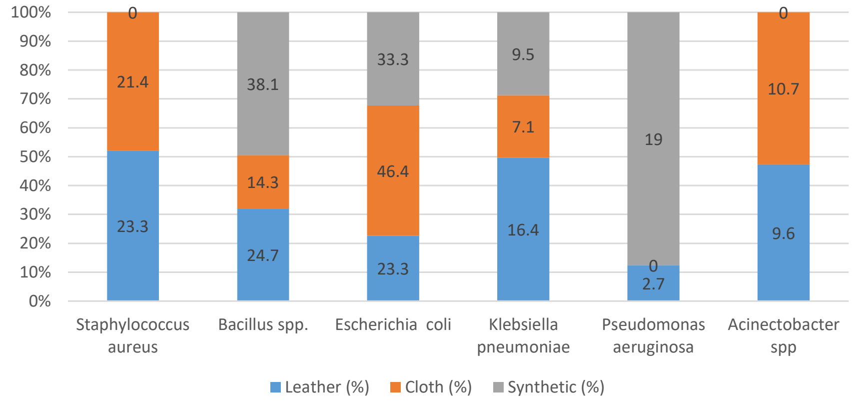
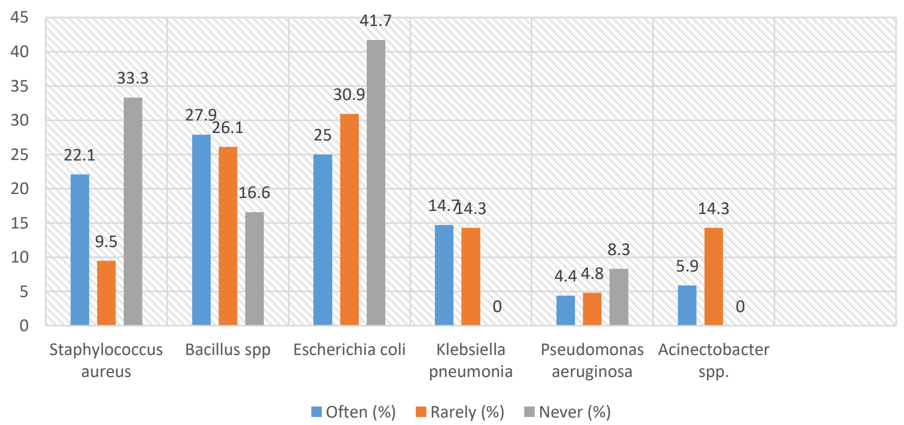


Figure 4: Prevalence of Bacterial Isolates from Handbags Based on Usage Duration



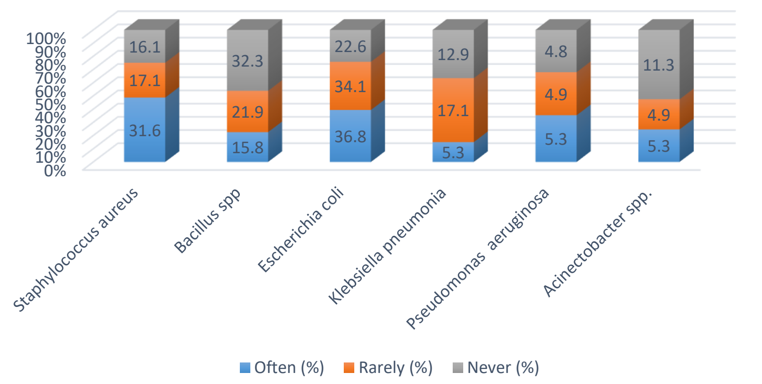


Figure 5: Prevalence of Bacterial Isolates from Handbags in Relation to Content Removal

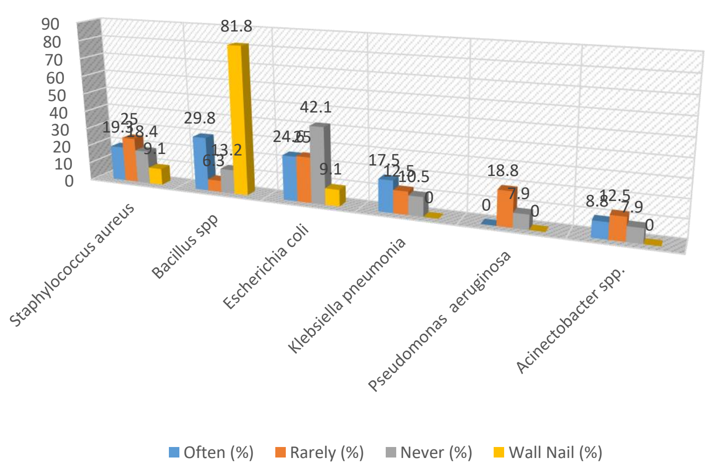


Figure 6: Prevalence of Bacterial Isolates from Handbags Based on Storage Conditions

Figure 7: Prevalence of ESBL-Producing Bacteria Isolated from Female Handbags in Abeokuta

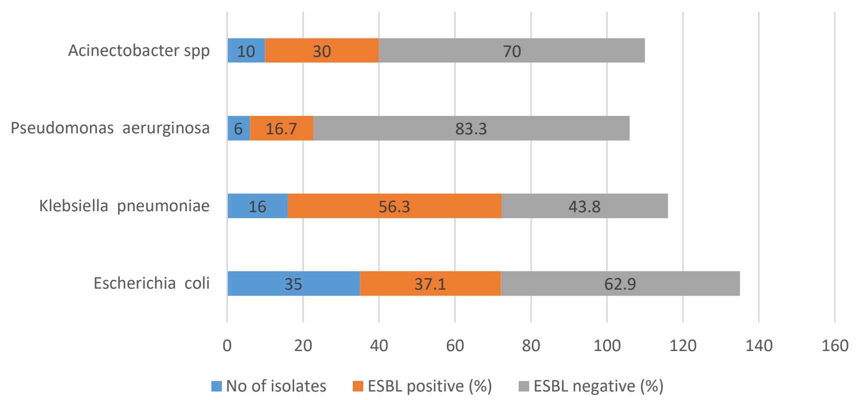


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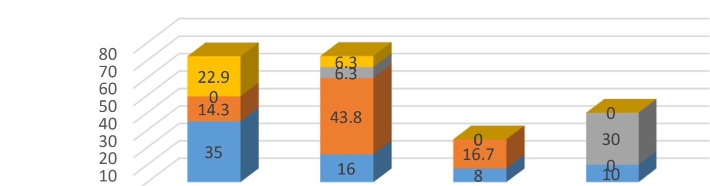
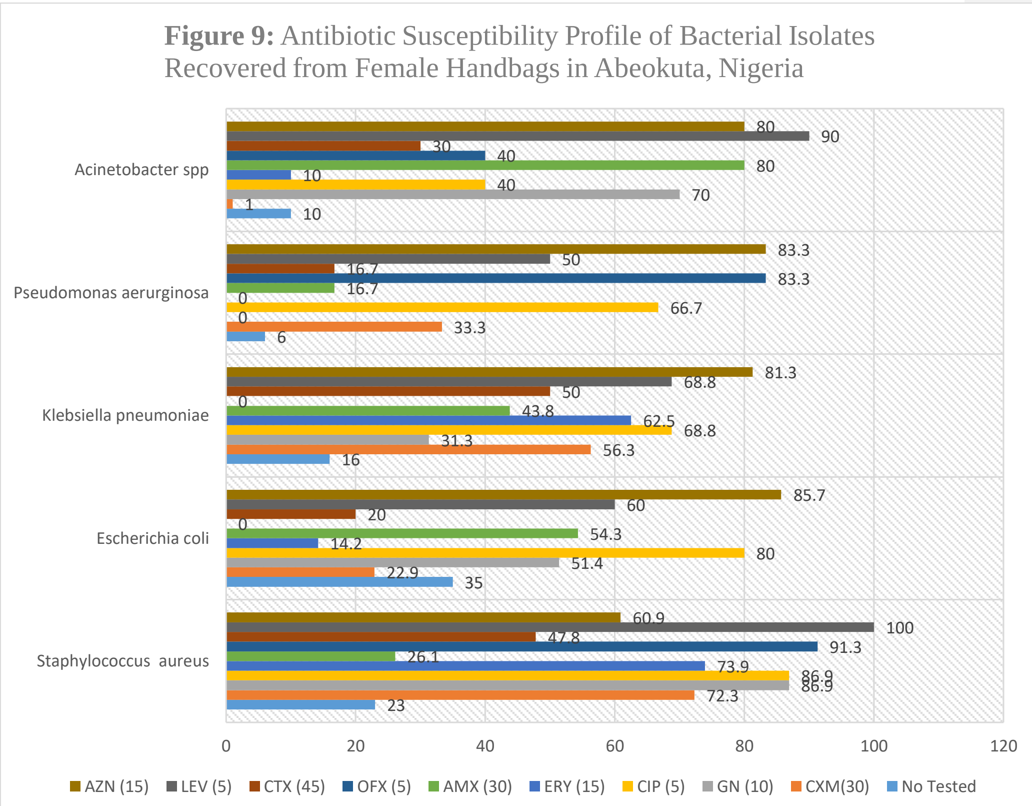
Figure 8: Susceptibility Profile of ESBL-Producing Isolates Recovered from Female Handbags in Abeokuta



	Escherichia coli	Klebsiella pneumoniae	Pseudomonas aerurginosa	Acinectobacter spp.
CTX(30µg)	22.9	6.3	0	0
AUG(30µg)	0	6.3	0	30
CAZ(30µg)	14.3	43.8	16.7	0
No of isolates	35	16	8	10

CAZ =Ceftazidime, CTX = Cefotaxime, AUG = Augmentin.

Bacteria isolates disk (µg)



No = Number, CXM = Cefuroxime, GN = Gentamicin, CIP = Ciprofloxacin, ERY = Erythromicin, AMX = Amoxicilin – clavulanate, OFX = Ofloxacin, CTX = Ceftriaxone, CFT = Cefixime, LEV= Levofloxacin, AZN = Azithromycin

DISCUSSION

In this study, 122 bacteria isolates from 300 swab samples collected from female handbags in Abeokuta, Nigeria, were used to determine their prevalence and susceptibility, and then screened for ESBL production. The interior of the handbags contained a higher percentage of bacterial contaminants than the handles and base. This is consistent with a study, which found that the interior surface of handbags may provide a favorable environment for microbes due to the nature of their frequent use (1). The handbags examined were found to be contaminated with a high concentration of Gram-positive and Gram-negative bacteria. The Gram-positive bacteria were mostly normal body flora, which explains the presence of *Staphylococcus aureus* in handbags. According to Itah et al., (4). Bacteria species such as *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella spp.*, and others were discovered to contaminate various surfaces, particularly those near people and frequently touched. *Bacillus spp.* was isolated from female handbags, which could explain why this organism is common and can withstand environmental changes for a while. *Escherichia coli*, *Klebsiella spp.*, and *Acinetobacter spp* were detected, indicating contamination and poor hygiene practices among the individuals. These microbes are present in feces, soil, and water. They can cause infection if hand hygiene and handbag hygiene are not practiced correctly (13). *Pseudomonas aeruginosa's* isolation may be due to its ability to live on both living and dead organisms, making it ubiquitous (14). The study revealed that leather bags displayed higher contamination rates compared to cloth or synthetic bags. Several potential mechanisms can be responsible for this including the material properties of leather, the usage patterns of handbags, and the environmental factors associated with their maintenance. (15). Leather is permeable and has a coarse surface that can harbor dirt, moisture, and bacteria. This environment can facilitate microbial proliferation, as bacteria thrive in warm, moist conditions (16). Leather can also provide a nutrient-rich environment for bacteria due to residual proteins and fats from the tanning process. This organic matter can serve as a food source for bacteria, promoting their survival and growth (17).

The prevalence of bacteria isolated from student handbags regarding content removal revealed that 19(15.6%) of female students who frequently empty their bags had low contamination of bacteria organisms, whereas 41(33.6%) and 62(50.8%) of female students who rarely or never empty contents from their handbags had a higher contamination rate of bacteria organisms. This could be because bacteria are everywhere, and contents such as face cream, handset, lipstick, face powder, and even money (currency notes) in female handbags may have been placed on contaminated surfaces such as the kitchen counter, bathroom sink, shopping carts, and even held by hands that have come into contact with dirty door knobs, toilet sinks, and dirty surfaces. When these items are left in handbags for an extended period, the bag provides an ideal environment for their proliferation (18).

Of the 122 samples tested, 26 (38.8%) produced ESBL. *Escherichia coli* had the highest ESBL production (19.4%), followed by *Klebsiella pneumoniae* (13.4%), and *Pseudomonas aureginosa* had the lowest (1.49%).

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A study conducted in the United States of America by Feldman and Feldman (2012) (5), found that women's handbags contained a variety of bacteria, including S*taphylococcus aureus* and *Escherichia coli*. The study reported that 100% of the handbags tested were contaminated, with leather bags showing higher levels of contamination. This aligns with this present study where leather bags were also associated with higher bacterial loads. The findings of this present study are also to the findings of Kaur et al., (2018) (21), that 85% of handbags tested were contaminated with pathogenic bacteria, including Escherichia coli and Klebsiella spp. Kaur et al in India also reported that leather bags had higher contamination rates compared to synthetic and cloth bags. Research by Mavhandu et al. (2019) (22), in South Africa indicated that handbags, particularly those made of leather, harbored significant levels of bacteria, with contamination rates reaching up to 90%. According to a study conducted in Adama Ethiopia (19). *Escherichia coli* and *Klebsiella pneumoniae* were the primary producers of ESBL. This suggests a consistent trend across different regions regarding the susceptibility of leather materials to bacterial contamination.

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The susceptibility profile of the Extended-Spectrum-Beta-Lactamase-producing isolate revealed Ceftazidime to be the most active antimicrobial agent, as it was least affected by ESBL enzymes, whereas Cefotaxime and Augmentin were the least active agents against these ESBL-producing bacteria. The antibiotic susceptibility profile of these bacteria isolates from female student handbags revealed that azithromycin (77.8%) was the most active antibacterial agent, while ceftriaxone (33.3%) was ineffective against the various isolates.

In this study, Gram-negative isolates were more common than Gram-positive isolates. According to Jorg and Thomas (20), Gram-negative organisms are the most common primary pathogens and are frequently found in the normal flora.

The higher percentage of samples yielding no growth (59% in the study) suggests that many individuals may be practicing effective hygiene measures, such as regular cleaning and maintenance of their handbags. This is crucial as it indicates that awareness of hygiene can lead to lower levels of bacterial contamination (21). The significant percentage of samples with no growth indicates a need for further research to understand the factors contributing to this outcome. Future studies could explore the relationship between handbag materials, usage frequency, and microbial presence to develop more effective hygiene guidelines (22). The absence of bacterial growth in a substantial number of samples may reflect variability in microbial presence due to factors such as the handbag's material, usage patterns, and environmental conditions. This variability can indicate that not all handbags are equally prone to contamination, which can inform future studies and hygiene recommendations (23). The findings from the study emphasize the importance of infection control practices beyond healthcare settings. Handbags, often used to carry personal items, can serve as vectors for the transmission of resistant bacteria. This highlights the need for public health campaigns aimed at educating individuals about hygiene practices, such as regular cleaning of personal items and hand hygiene. Infection control measures should extend to community education on the risks associated with contaminated personal items. The CDC emphasizes the importance of hand hygiene and cleaning

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frequently touched surfaces to prevent the spread of infections, particularly in light of rising antibiotic resistance (24).

The study's findings regarding extended-spectrum beta-lactamase (ESBL)-producing bacteria underscore the need for awareness of resistance mechanisms. ESBLs can inactivate a wide range of beta-lactam antibiotics, making infections caused by these organisms particularly challenging to treat. The presence of ESBL-producing bacteria in community settings indicates a potential reservoir for these pathogens. Clinicians must consider local resistance patterns when selecting empirical therapy for infections. The Infectious Diseases Society of America (IDSA) recommends that healthcare providers utilize local antibiograms to guide antibiotic selection (25). The study highlighted a significant prevalence of antibiotic-resistant bacteria, particularly *Escherichia coli* and *Klebsiella pneumonia*, isolated from female handbags. These organisms are known to cause a range of infections, including urinary tract infections and bloodstream infections. The presence of such resistant strains in everyday items suggests a potential for transmission to individuals, leading to infections that are increasingly difficult to treat. The emergence of antibiotic-resistant bacteria is a major public health concern. According to the Centers for Disease Control and Prevention (CDC), at least 2.8 million antibiotic-resistant infections occur in the United States each year, leading to more than 35,000 deaths (26). Infections caused by resistant strains often require more complex treatment regimens, which can increase healthcare costs and patient morbidity and mortality (27).

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LIMITATIONS OF THE STUDY

While this study sheds light on the prevalence of Extended Spectrum Beta-Lactamase (ESBL)-producing bacteria in female handbags, several limitations should be noted to ensure transparency and help readers understand potential biases and constraints that may have influenced the results.

1. Sample Size and Selection: The study included 300 female handbag samples, which may not be representative of the overall population. Participants were chosen from a specific geographic area in Abeokuta, Nigeria, which may not reflect the prevalence of ESBL-producing bacteria in other regions or demographics. A larger and more diverse sample size may provide a more complete picture of the problem.
2. The study's cross-sectional design limits the ability to establish causal relationships between bacteria and handbag usage or hygiene practices. While associations can be identified, the study cannot conclude whether specific behaviors directly contribute to bacterial contamination.
3. Self-Reported Data: The study used questionnaires to collect information about participants' handbag usage, cleaning habits, and environmental factors. Self-reported data is susceptible to bias because participants may not accurately recall or disclose their behaviors. This may result in an underestimate or overestimation of the actual practices that contribute to bacterial contamination.

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4. Microbiological Techniques: Standard microbiological methods were used to identify bacterial isolates and determine antibiotic susceptibility. While these methods are widely accepted, they may have inherent limitations, such as the possibility of misidentifying specific bacterial species or varying susceptibility testing results. Furthermore, the study did not investigate the presence of other potentially pathogenic microorganisms, such as viruses or fungi, which may also pose health risks.
5. Temporal Factors: Data collected over a specific time frame may not account for seasonal variations in bacterial prevalence or changes in public health practices that impact contamination levels. Longitudinal studies would help to assess trends over time.

By addressing these limitations and pursuing the suggested future research avenues, researchers can contribute to a more nuanced understanding of the risks associated with microbial contamination in everyday items such as handbags. Finally, this knowledge can help shape public health strategies aimed at reducing the spread of multidrug-resistant organisms and improving overall community health.

CONCLUSION

The current study found a high prevalence of ESBL among Gram-negative bacteria isolated from female students' handbags in Abeokuta, Nigeria. *Pseudomonas aeruginosa* was the most common producer of Extended-Spectrum-Beta-Lactamase. The majority of the isolates were sensitive to azithromycin and resistant to ceftriaxone. Female handbags contain a variety of multidrug-resistant bacteria that can contaminate other items in them and serve as a conduit for the transmission of pathogenic bacteria to users. As a result, awareness should be raised to educate females about this potential route of disease transmission to help prevent the spread of multidrug-resistant infections.

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RECOMMENDATIONS

To effectively raise awareness and promote better hygiene practices, implement the following practical recommendations and interventions:
1. Educational Campaigns: Public health organizations and educational institutions should launch campaigns to educate people about the risks of handbag contamination. These campaigns can use social media, workshops, and informational brochures to spread awareness about proper handbag hygiene.

2. Guidelines for Handbag Maintenance: Create and distribute clear guidelines for cleaning and maintaining handbags. Regular cleaning with disinfectant wipes, avoiding placing handbags on potentially contaminated surfaces (such as public restrooms or kitchen counters), and using protective pouches for cosmetics and food items are all possible recommendations.

3. Incorporating Hygiene into Fashion: Work with handbag manufacturers and retailers to promote hygiene-friendly designs. This could include easier-to-clean materials or the use of

antimicrobial coatings. Marketing campaigns can highlight the importance of hygiene in handbag selection.

4. Workshops and Demonstrations: Hold community workshops to demonstrate effective cleaning techniques for handbags and their contents. These hands-on sessions can provide individuals with useful skills and knowledge for maintaining hygiene.

5. Regular Reminders: Use digital platforms to send out regular reminders about handbag hygiene, especially during peak usage periods such as back-to-school or holiday shopping. These reminders can help people remember to clean their handbags regularly.
6. Research and Feedback: Encourage ongoing research into handbag hygiene and its impact on public health. Gathering feedback from educational program participants can help to refine strategies and improve outreach efforts.

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