

Research Article

Occurrence and Antibiotic Susceptibility of *Escherichia coli* from Door Handles in the Faculty of Pharmacy, Nnamdi Azikiwe University, Agulu

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Abstract

Door handles are high-contact surfaces that can serve as reservoirs for pathogenic microorganisms, including *Escherichia coli*, a significant cause of community- and hospital-acquired infections. Increasing antibiotic resistance among *E. coli* strains poses a public health challenge, necessitating local surveillance. This study investigated the prevalence of *E. coli* contamination on door handles within the Faculty of Pharmacy, Nnamdi Azikiwe University, Agulu, and assessed the antibiotic susceptibility patterns of the isolates. A total of 100 swab samples were collected from door handles across offices, classrooms, hostels, laboratories, clinic, and other locations. Samples were cultured in nutrient broth, subcultured on MacConkey agar, and pure isolates identified using Gram staining, cultural characteristics and biochemical tests. Antibiotic susceptibility testing was performed using the Kirby-Bauer disc diffusion method on Mueller-Hinton agar with 12 commonly used antibiotics. Data were analyzed using CLSI breakpoints and Multiple Antibiotic Resistance Index (MARI) was used to assess multidrug resistance. Seventeen *E. coli* isolates were recovered, with the highest prevalence from hostel door handles (59%). Antibiotic susceptibility testing revealed high resistance to β -lactams, including Ampicillin/Sulbactam (94%) and Piperacillin/Tazobactam (88%), as well as third- generation cephalosporins such as Cefotaxime (82%). Conversely, 100% susceptibility was observed for Gentamicin, Azithromycin, and Chloramphenicol, while Ofloxacin and Levofloxacin retained significant activity (88% susceptibility). Twelve isolates (71%) exhibited multidrug resistance, with MARI values >0.2 , suggesting exposure to high antibiotic pressure. The findings highlight the role of fomites in harboring multidrug-resistant *E. coli*, underscoring the need for improved hygiene practices, routine surveillance, and rational antibiotic use within academic environments.

Keywords

Escherichia coli, Antibiotic Resistance, Fomites, Door Handles, Multidrug Resistance

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1. Introduction

Microorganisms are found everywhere; in some cases, living as contaminants that cause health hazards and are sources of community and hospital-acquired infections. Hand railings, showers, chairs, tables and door handles of public places like schools and offices are a hotspot for different bacteria. As people come in contact with surfaces, there is a high chance of taking up the microorganisms deposited on them. This is so because not much attention is given to doorknobs while sanitising, unlike other items [1, 2].

Bacteria can survive for variable durations on surfaces like mobile devices, pens, and door handles. The risk of transmission depends on the duration of contamination and the microbial load on these surfaces. However, survival is determined by varying factors such as temperature, humidity, the presence of organic matter, the ability to form biofilms, and infection control techniques. Inadequate hand hygiene has the potential to lead to the proliferation of bacteria and thus, increase the risk of transmission [3, 4].

Escherichia coli is a rod-shaped, Gram-negative, facultative anaerobic bacterium belonging to the genus *Escherichia* and cause of illness ranging from mild gastrointestinal symptoms, urinary tract infections, meningitis and pneumonia [5-7].

Antibiotic resistance is a public health issue encountered worldwide, affecting a variety of disease conditions, including community-acquired infections and food-borne infections. The treatment of community-acquired infections is negatively impacted by antibiotic resistance, as it can result in increased rates of morbidity and mortality, higher treatment costs, poor treatment outcomes due to limited treatment alternatives, and a wider and more rapid spread of resistant bacteria [8-10]. This emergence of antibiotic resistance threatens the treatment of *Escherichia coli* infections and the worldwide increase of multidrug-resistant *Escherichia coli* strains is mostly associated to the dissemination of mobile genetic factors such as plasmids [11-13].

This study seeks to determine the extent of microbial contamination on door handles in the Faculty of Pharmacy, Agulu and to further isolate *Escherichia coli* and determine its antibiotic resistance and susceptibility pattern.

2. Method

2.1. Study Site

The study was conducted in the faculty of Pharmacy, Agulu, Nnamdi Azikiwe University, situated in Anambra State's Anaocha Local Government Area. The faculty is divided into six departments, including the Department of Pharmaceutics and Pharmaceutical Technology, Department of Pharmacognosy and traditional medicine, Department of Toxicology and Pharmacology, Department of Clinical Pharmacy and pharmacy management, Department of Pharmaceutical and

medicinal chemistry, and Department of Pharmaceutical microbiology and biotechnology. The faculty has specified buildings for each department and classrooms for the students. The population of students in the faculty is estimated to be above 700, thus creating an enabling environment for the transfer and proliferation of microorganisms, especially bacteria, through different sources, for instance, fomites like door handles.

2.2. Sample Size Determination

The sample size for the experiment was determined to be 95 using the Cochran Correction formula [14] and a prevalence rate of 9% from a similar study [15].

The calculation is illustrated below; Cochran's Formula is given by;

$$n = \frac{p*q}{\left(\frac{e^2}{Z^2}\right) + \left(\frac{p*q}{N^2}\right)}$$

where

n = sample size

p = prevalence rate = 9% = 0.09

q = 1-p = 1-0.09 = 0.91

e = maximum acceptable marginal error = 0.05 (at 95% confidence level)

z = 1.96 (at 95% confidence level)

N = population size = 384 samples

$$n = \frac{0.09*0.91}{\left(\frac{0.05^2}{1.96^2}\right) + \left(\frac{0.09*0.91}{384^2}\right)}$$

n=95 samples

Ninety-five (95) samples were obtained from the formula above. The number of samples was made up to 100.

2.3. Collection of Samples

A total of 100 samples were collected from different door handles in the Faculty of Pharmacy, Agulu. With the aid of a labelled swab stick soaked in normal saline, the samples were collected from the door handles.

A systematic approach was employed to identify and characterize door handles within the study environment. To ensure relevance and uniformity in sampling, only fully operational door handles installed on routinely accessed entry and exit doors were included. Handles on permanently locked doors, mechanical rooms, or restricted-access areas were excluded to minimize observational bias and enhance representativeness of user-contact surfaces.

The samples on the swab stick were aseptically inoculated into a sterile nutrient broth and incubated at 37°C for 24 hours.

2.4. Isolation, Microbial Identification and Characterisation of *Escherichia coli*

With the aid of a sterile wire loop, inocula from the broth culture that were incubated for 24 hours were spread on MacConkey agar plate and incubated at 37°C for 24 hours. A colony of *Escherichia coli* (identified as the pink growth on the agar) was picked from the MacConkey agar plate, after 24 hours and, using the streak method, re-inoculated to get a pure culture. Pure isolates were each cultured on agar slants in bijoux bottles and incubated at 37°C for 24 hours. The bottles were refrigerated at temperatures between 2°C and 8°C for storage.

The bacterial isolates were further identified using their Gram stain reaction (microscopy), the morphological appearance (macroscopic) of their colonies, and confirmatory biochemical tests [16].

2.5. Antibiotic Susceptibility Testing

The antibiotic susceptibility tests were performed following the Kirby-Bauer disc diffusion method using Mueller-Hinton agar [17, 18]. The standardised inocula were swabbed onto a Mueller-Hinton agar plate, and the Gram-negative antibiotic multidiscs were placed on the inoculated plates and firmly onto the agar plate to ensure complete contact. This process was carried out in triplicate. The bacterial isolates were tested against the following antibiotics; Ampicillin/Sulbactam (20mcg), Co-Trimoxazole (25mcg), Cefo-

taxime (30mcg), Piperacillin/Tazobactam (110mcg), Chloramphenicol (30mcg), Ciprofloxacin (30mcg), Ceftriaxone (30mcg), Tetracycline (30mcg), Ofloxacin (5mcg), Gentamicin (10mcg), Azithromycin (15mcg) and Levofloxacin (5mcg).

The plates were incubated in an inverted position at 37°C for 24 hours. The susceptibility of each isolate to each antibiotic on the multidisc was shown by a clear zone of inhibition, which was measured in millimetres using a meter rule. The diameter of each zone of inhibition was later interpreted using a standard CLSI interpretative chart [17].

2.6. Data Analysis

The Multiple Antibiotic Resistance Indices (MARI) were calculated for each isolate by dividing the number of antibiotics the isolates were resistant to by the total number of antibiotics that the isolates were subjected to [19].

3. Results

3.1. Collection of Samples

A total of 100 samples were obtained from door handles of offices, classrooms, restrooms and hostel rooms in the Faculty of Pharmacy Agulu (Figure 1). These Other places include: security post, canteen and generator houses.

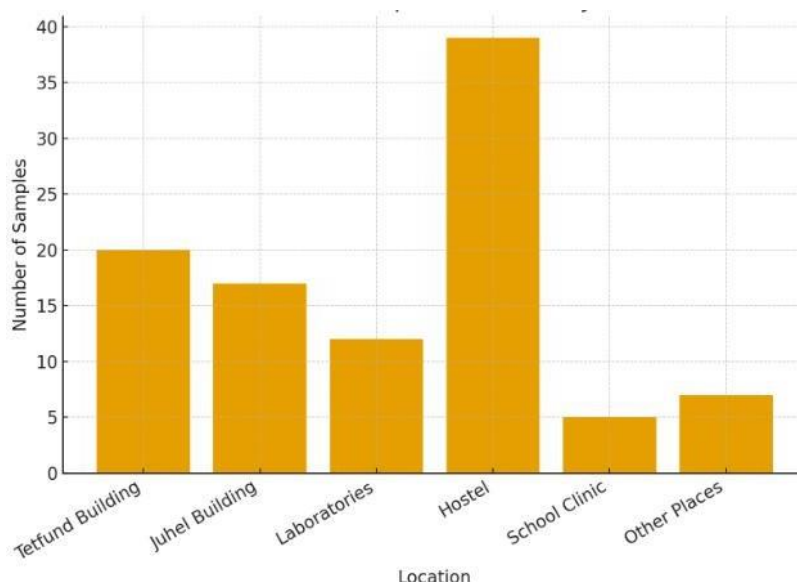


Figure 1. Distribution of samples collected by Location.

3.2. Isolation of Samples

Out of the 100 samples collected from different door

handles in the Faculty, only seventeen [17] *E. coli* isolates were obtained. The hostel door handles had the highest percentage of the isolates, while the Tetfund building had no isolates.

3.3. Antibiotic Susceptibility of *Escherichia coli*

82% of the *E. coli* isolates were resistant to Cefotaxime (3rd generation cephalosporin), 88% and 94% of the isolates were resistant to Piperacillin/Tazobactam and Ampicillin/Sulbactam, respectively. Ofloxacin and Levofloxacin had better activity against the isolates, as 88% were susceptible,

and 6% of the bacterial isolates were resistant to these two drugs. Chloramphenicol, Gentamicin and Azithromycin exhibited similar susceptibility patterns and had the best activity, as all (100%) of the isolates were susceptible. No isolate was susceptible to Cefotaxime, and only 1 isolate was susceptible to Ampicillin/Sulbactam, Piperacillin/Tazobactam and Ceftriaxone, respectively.

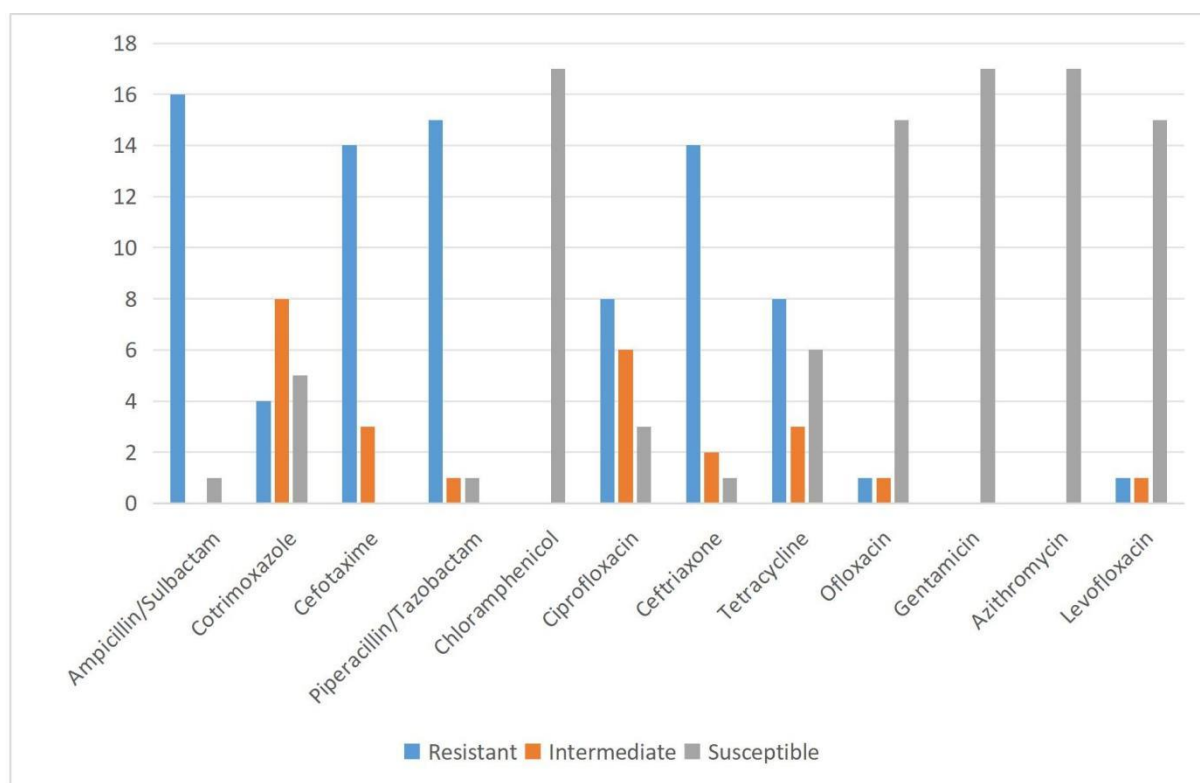


Figure 2. Antibigram of *Escherichia coli* isolates to different antibiotic agents.

The multidrug-resistant isolates were determined by their resistance to at least one agent in three different classes of antibiotics. Out of the *Escherichia coli* isolates obtained, twelve (12) isolates were multidrug resistant. Hostel doors had the highest prevalence of *Escherichia coli* (59%) and multidrug-resistant isolates (67%), while Tetfund doors had no *Escherichia coli* (0%) isolates at all. Other doors had one *Escherichia coli* isolate (6%) and no multidrug-resistant isolate at all (0%).

Table 1. Prevalence of MDR *Escherichia coli* isolates across the Faculty buildings.

Location	Number of <i>E.coli</i> isolates	MDR isolates
Tetfund building	0	0
Juhel building	3	2
Laboratories	1	1

Location	Number of <i>E.coli</i> isolates	MDR isolates
Clinic	2	1
Hostel	10	8
Others	1	0
Total	17	12

4. Discussion

This study was conducted to investigate the prevalence and antibiotic susceptibility patterns of *Escherichia coli* associated with fomites (door handles). Fomites like door handles of offices, classrooms, conveniences and hostels are a major source of disease spread as they are mostly neglected when cleaning or sanitising the environment, hence the heavy microbial load observed on them. The risk of disease transmission through fomites is dependent on certain factors like frequency of site contamination and exposure, level of

pathogen excreted by host, virulence of organism and immune-competence of infected persons [20].

In this study, door handles of offices, restrooms, classrooms and hostel rooms in the Faculty of Pharmacy, Agulu were swabbed and evaluated to determine the level of bacterial contamination and to isolate *Escherichia coli*. Out of the 100 samples collected, all of them showed some form of bacteria growth after culturing in nutrient broth and incubating for 24 hours, this further proves that door handles are significant fomites with a great potential of housing microorganisms, it also proves that microbes are ubiquitous and a high level of bacterial contamination on the door handles, which is mostly gotten from contact from human hands, is indicated.

However, only 17 *Escherichia coli* species were isolated from the 100 samples, which is a prevalence rate of 17%. This is less than what was reported in a similar study carried out on door handles in the University of Calabar [21]. The prevalence rate of *Escherichia coli* found in this study (17%) is greater than the 9% reported by Maina [15] in a similar study conducted in a district hospital, in Kenya. The difference in the prevalence of *Escherichia coli* in these studies could be attributed to the different locations of sample collection, varying levels of microbial load and hygienic practices of the occupants of the area [22, 23].

The female hostel doors had the highest number of *Escherichia coli* isolates, as the samples were collected from the toilet doors in the rooms, which have a potential for faecal contamination, which is a primary source of *Escherichia coli*, and it has a huge tendency to cause diseases by cross-contamination [20]. The high microbial load found here could also be attributed to certain habits of women that could enhance contamination. The doors in the Tetfund building had no *Escherichia coli* isolate at all, however, the samples collected from the Tetfund building still showed a level of bacterial contamination. The absence of *Escherichia coli* isolates here could be due to the absence of faecal contamination in the door handles that were swabbed.

An antibiotic susceptibility test was carried out on the isolates, using a Gram-negative antibiotic multidisc. From the results obtained, the majority of the isolates were susceptible to classes of antibiotics like Fluoroquinolones (Ofloxacin and Levofloxacin), Aminoglycosides (Gentamicin), Macrolides (Azithromycin), and Chloramphenicol. This corresponds with the results of Daoud *et al.* [24], which recorded the sensitivity of isolates to Gentamicin and Ofloxacin. It is also similar to studies by Wang *et al.* [25] and Adzitey *et al.* [26], which recorded a high level of sensitivity to Levofloxacin, Chloramphenicol and Azithromycin. The isolates were mostly resistant to Penicillin (Ampicillin/Sulbactam and Piperacillin/Tazobactam); this result is greater than the findings to results from a study by Aabed *et al.* [27]. The isolates were also resistant to Cephalosporins (Cefotaxime and Ceftriaxone) and Tetracycline. This finding is similar to the results from a study by Nwafia *et al.* [28]. The isolates also

demonstrated intermediate susceptibility patterns to Co-trimoxazole. This is dissimilar to the results from a study conducted on Penicillin by Humayun *et al.* [29]. These differences could be attributed to the variety of factors that confer resistance and the different mechanisms of action of the antibiotics [30]. Contributors to the antibiotic susceptibility pattern observed may include frequent consumption of antibiotics in our locality, as they are readily accessible, quite affordable and easy to administer. Incomplete antibiotic dose intake can result in these resistance patterns observed [31].

The MARI calculation revealed that 14 isolates (82%) had MARI values greater than 0.2. This further buttresses the point that the isolates were sourced from places where antibiotics are frequently used, as it is very likely for medical students to abuse/misuse antibiotics when they notice signs of illness because they assume they have the knowledge to prescribe and self-medicate [19].

Out of the *Escherichia coli* isolated, 12 (71%) isolates were observed to be resistant to drugs in more than two antibiotic classes. The prevalence of multidrug-resistant *Escherichia coli* in the faculty poses a high risk of causing resistant infection among students and probably other faculty members, which may lead to difficulty in treatment, low productivity and various health complications.

5. Conclusion

The prevalence of *Escherichia coli* isolated from door handles in the Agulu campus was 17%. They were all sensitive to Chloramphenicol, Gentamicin and Azithromycin. The results of this study showed that door handles in the Faculty of Pharmacy Agulu have the potential to harbour infectious agents and multidrug-resistant *Escherichia coli* strains. A greater percentage of the isolates were multidrug resistant, and this indicates the high tendency of antibiotic misuse among people in the faculty. The outcome of this study places a huge responsibility on students and members of staff in the Faculty of Pharmacy Agulu to improve their hygiene, especially handwashing, and other sanitary measures. Further studies should be carried out to identify the mechanisms of resistance of these *Escherichia coli* strains so that the most appropriate class of antibiotics can be used to control their spread.

Abbreviations

Mcg	Micrograms
MDR	Multi Drug Resistant
MARI	Multiple Antibiotic Resistance Indices

Conflicts of Interest

The authors declare no conflicts of interest.

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