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THE ROLE OF DOOR HANDLES IN THE PROLIFERATION OF MICROORGANISMS OF PUBLIC HEALTH INTEREST IN MALE STUDENT HOSTELS, UNIVERSITY OF BENIN, UGBOWO CAMPUS, BENIN CITY, NIGERIA

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ABSTRACT

Contact with contaminated door handles is a means of infection transmission. This study investigated the role of door handles in the proliferation of microorganisms in male student hostels at the Ugbowo Campus of University of Benin. A total of 378 swab samples from the rooms' door and toilet door handles in 3 hostels were randomly collected using sterile swab sticks between April, 2015 and September, 2015. The samples were processed using standard microbiological and molecular techniques. The bacterial counts for the period of 6 months ranged from 3.1 ± 0.52 to $6.4 \pm 0.94 \times 10^3$ cfu/cm², $4.4 \pm 0.37 \times 10^3$ to $8.5 \pm 0.55 \times 10^3$ cfu/cm² and $4.2 \pm 0.55 \times 10^3$ - $7.2 \pm 0.85 \times 10^3$ cfu/cm² for Halls 3, 4 and 5 respectively. The bacterial isolates identified were *Staphylococcus aureus*, *Bacillus subtilis*, *Streptococcus pyogenes*, *Aerococcus viridians*, *Corynebacterium sp.*, *Staphylococcus epidermidis* *Micrococcus sp.*, *Klebsiella pneumonia*, *Proteus vulgaris*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Enterobacter aerogenes*, *Acinetobacter haemolyticus*, and *Serratia marcescens*. *Staphylococcus aureus* was recorded the most predominant isolate with percentage frequency of occurrence at 23.61% prevalence while the least percentage frequency of occurrence was recorded for *Aerococcus viridians* at 0.24%. The pre and post curing susceptibility test of the isolates revealed that most of the antibiotics used like chloramphenicol, augmentin, ampiclox, streptomycin, septrin, pefloxacin and amoxicillin were resisted by all the isolates except *S. epidermidis* which showed increased susceptibility to Gentamycin, ciprofloxacin and amoxicillin, after curing. There were detectable plasmid fragments in *E. coli*, *P. aeruginosa*, *Corynebacterium sp.*, *S. epidermidis*, *S. aureus*, *P. vulgaris*, *E. aerogenes* and *K. pneumonia*. Control measures should thus be established to reduce the spread of these antibiotic resistant isolates through door handles.

1. INTRODUCTION

Microorganisms are universally widespread and make up a very significant part of every ecological system. They exist freely or are dependent on a host in these environments (Sleigh and Timbury, 1998). At times, they could be temporary contaminants in hands or fomites in which case they can pose a serious health hazard as a medium through which infections are transmitted (Pittet et al., 1999).

Even though the possibility of infection transfer in a community setting is indisputably not as high as that connected with patients in healthcare facilities, the annual increase in cases of gastrointestinal disorder, in which outbreaks in a family unit are major determinant, needs investigation into the likely sources and causes (Deodhar, 2003). Other than the everyday dealings between people, which constitute a means of infection transfer, the main medium of transfer of community acquired infections is contact with contaminated fomites (Prescott et al., 1993; Li et al., 2009). Fomites, when in constant contact with people or natural habitats of pathogenic organisms constitute a significant medium of spread of infectious diseases (Osterholm et al., 1995). Such fomites can be found on door handles of hostels, showers, toilet seats, spigots, sinks, seats, and tables, particularly those found in public places like hospitals, eateries, restrooms and schools (Bright et al., 2010). School lodgings have large number of students, who come in with their diverse microbial flora and other organisms they have picked somewhere else, these are then deposited on door handles / knobs in the process of going in and out of the accommodation space (Goldhammer et al., 2006). This study was therefore aimed at investigating the role of door handles in the proliferation of microorganisms in male student hostels, University of Benin, Ugbowo Campus, Benin City.

2. MATERIALS AND METHODS

2.1. Study Area

The study area, University of Benin, Benin-City lies within longitude 5.62°E and latitude 6.4°N, approximately 350 km SW of Abuja, the Federal Capital Territory. It is located in the capital of Edo State in the South-South region of Nigeria. The student population is about 70,000.

2.2. Sample Collection

A total of 378 samples comprising of 21 samples per hostel resulting in a total of 63 samples from the 3 male student hostels (Halls 3, 4 and 5) per month were collected during the sampling period of April, 2015 to September, 2015. Samples were collected from surfaces of room door handles and toilet door handles. A surface area of 10 cm² was swabbed with moistened swab sticks (inserted into 10 ml of sterile peptone water) and transported to the Laboratory for bacteriological analyses.

2.3. Enumeration of Bacteria

The enumeration of the microorganisms was carried out in accordance to the methods described by Chesebrough (2000) as well as Public Health England (2000) using standard plate count procedures. Serial dilution of the sample was aseptically carried out by pipetting 1 ml from the inoculated peptone water into a test tube (10^{-1}) containing 9 ml of sterile distilled water. The process was repeated until a dilution of 10^{-3} was obtained. A quantity (1 ml) of 10^{-3} dilution was aseptically introduced into sterile Petri dishes and prepared nutrient agar added using Pour plate techniques. The plates were incubated at 37 °C for 24-48 hrs. Colonies were counted and the mean counts recorded and expressed in colony forming units per centimeter square (cfu/cm²).

2.4. Isolation, Characterization and Identification of the Bacterial Isolates

This was carried out using cultural, morphological and biochemical methods. A sterile wire loop was used to pick distinct colonies and transferred into prepared nutrient agar (oxid) plates using streaking method and incubated at 37 °C for 24-48 hrs. A loopful of the culture was streaked on sterilized slant bottles containing prepared nutrient agar (oxid) medium using streaking plate techniques. The slants were incubated at 37 °C for 24-48 hrs after which Gram's reaction, spore staining, motility test and biochemical tests (catalase, coagulase, sugar fermentation, oxidase, urease, indole and citrate test) as well as hemolytic pattern on blood agar and growth on some differential media (MacConkey and Mannitol salt agar) were used to validate the identity of the isolates (Chesebrough, 2000)..

2.5. Antibiotic Susceptibility Testing

A sterile cotton swab was dipped into the standardized suspension and used to evenly inoculate the entire surface of nutrient agar and allowed to dry for five minutes after which sterile forceps were used to place the antibiotic test disks (Maxicare Medical Laboratory) onto the agar surface depending on whether the test organism plated was a Gram negative or Gram-positive organism (Kirby et al., 1966). The plates were incubated at 37°C for 24 hrs. The zones of inhibition were measured and recorded in millimeter. The results were interpreted on the basis of Clinical and Laboratory Standards Institute Guidelines (2012).

2.6. Plasmid Extraction

A measured volume of 2-3 ml of freshly grown culture was centrifuged, and the pellet was resuspended in 1 ml of a solution containing 0.04 M Tris-acetate, (pH 8.0) and 2 mM EDTA. Also, a volume of 2 ml lysis buffer (0.05 M Tris, 3% SDS, pH 12.5, adjusted with 2 N NaOH and mixed gently, the suspension was incubate at 60-68°C for 30-45 min. An amount (6 ml) of phenol/chloroform (1:1) was added to the hot samples and mixed gently to complete emulsification. Phases were separated by centrifugation at $10.000 \times g$ for 15-20 min at RT (where R is the universal gas constant ($8.314 \text{ 511 J K}^{-1} \text{ mol}^{-1}$), T the absolute temperature) and the upper aqueous phase was transferred carefully to new microfuge tube containing 1 volume (400 μ l) of chloroform. This was mixed and centrifuged again for separation of

phases. The aqueous phase was recovered and used directly for agarose gel electrophoresis (Kado and Liu, 1981).

2.7. Plasmid Curing

The Isolates that showed resistance to the antibiotics used (Septin (30 µg), Chloraphenicol (30 µg), Sparfloxacin (10 µg), Ciprofloxacin (10 µg), Amoxicilin (30 µg), Augmentin, Gentamycin (10 µg), Perloxacin (10 µg), Streptomycin (30 µg), Zinnacef (20 µg), Rocephin (25 µg), Erythromycin (10 µg), Ofloxacin (10 µg), Ampiclox (30 µg), were subjected to standard plasmid curing method (Sijhary et al., 1984). Overnight broth culture was inoculated into 4.5 ml nutrient broth and 0.5 ml of sodium dodecyl sulphate was added and incubated at 37°C for 48 hrs. An aliquot, 0.5 ml of the broth was added to a freshly prepared 4.5 ml nutrient broth, incubated for another 24 hrs at 37 °C after which post plasmid antibiotic susceptibility was carried out. The purpose of this repeated susceptibility test was to determine whether the eliminated plasmids were responsible for resistance to the antibiotics used.

2.8. Statistical Analyses

The statistical analyses of the data obtained were performed using Microsoft office Excel 2007. Data generated from the study was analyzed using the parametric test of analysis of variance (ANOVA), at $P < 0.05$ confidence limits for all parameters (Ogbeibu et al., 2015).

3. RESULTS AND DISCUSSION

The door handles sampled for the period of six months in the three (3) male student hostels showed bacterial contamination. The total viable bacterial counts on door handles in Halls 3, 4 and 5, between April, 2015 and September, 2015 ranged from $3.1 \pm 0.52 \times 10^3$ to $6.4 \pm 0.94 \times 10^3$ cfu/cm², $4.4 \pm 0.37 \times 10^3$ to $8.5 \pm 0.55 \times 10^3$ cfu/cm², $4.2 \pm 0.55 \times 10^3$ to $7.2 \pm 0.85 \times 10^3$ cfu/cm² for Hall 3, Hall 4 and Hall 5 respectively (Tables 1 to 3). There was no statistical significance ($p > 0.05$) of bacterial loads on door handles in the 3 male student hostels but significance difference was observed in the 6 months of study ($p < 0.05$). It was revealed that toilet door handles presents higher level of bacterial contamination compared to the door room handles of the sampled door handles. This difference in the level of bacterial contamination of the door handles could be attributed to the vulnerability of the toilets than rooms and as a result, larger population usage of toilets than rooms, coupled with the poor hygienic conditions of the toilets. There was variation in the bacterial counts on the door handles during the sampling periods, which could be attributed to the student's activities in the male student hostels at the time of sampling. The high bacterial counts in some months, specifically during examination period in the months of April, 2015 and September, 2015 in Halls 3, 4 and 5 could as well be attributed to increased activities of students in the hostels, resulting to frequent usage of door handles. These result findings are in agreement with the findings of Boone and Gerba (2010), who reported that the levels of contamination of conveniences differ depending on traffic, exposure and environment. Bacterial contamination of door handles and knobs are very much reported and these fomites in turn serve as vehicles for cross-infections and recontamination of washed hands (Monarca *et al.*,

2000; Otter and French, 2009; Bright *et al.*, 2010). Several of the bio-contaminants can be pathogenic and can be transferred starting from one individual then to the next or may bring about auto-inoculation (Kennedy *et al.*, 2005; Li *et al.*, 2009). Otter and French, (2009), reported 95% positive cultures in similar environments. The result of increase in high bacterial load may be attributed to the poor hygienic conditions in the Student Hostels environment. It could also be attributed to the unhygienic handling of the facilities by students with varying hygiene profile and lack of proper cleaning of contact surfaces engaged by the institution. Despite the use of a variety of methods and techniques for cleaning and sterilization of environmental surfaces, the door handles still play an important role in transmission of pathogens.

Table 1: Total viable bacterial counts on door handle in Hostel A

Samples	April	May	June	July	August	September	Mean \pm S.E
T1	19.3 $\times 10^3$	9.2 $\times 10^3$	1.02 $\times 10^3$	8.1 $\times 10^3$	13.4 $\times 10^3$	20.9 $\times 10^3$	13.5 $\times 10^3 \pm 2.21$
RM1	4.5 $\times 10^3$	3.7 $\times 10^3$	4.3 $\times 10^3$	6.5 $\times 10^3$	5.2 $\times 10^3$	4.8 $\times 10^3$	4.8 $\times 10^3 \pm 0.39$
RM2	3.1 $\times 10^3$	3.1 $\times 10^3$	2.6 $\times 10^3$	3.6 $\times 10^3$	5.9 $\times 10^3$	3.5 $\times 10^3$	3.6 $\times 10^3 \pm 0.48$
RM3	4.8 $\times 10^3$	2.3 $\times 10^3$	1.7 $\times 10^3$	2.9 $\times 10^3$	6.3 $\times 10^3$	5.1 $\times 10^3$	3.9 $\times 10^3 \pm 0.74$
T2	12.3 $\times 10^3$	7.6 $\times 10^3$	16.8 $\times 10^3$	11.9 $\times 10^3$	11.2 $\times 10^3$	12.6 $\times 10^3$	10.2 $\times 10^3 \pm 2.15$
RM4	2.5 $\times 10^3$	2.1 $\times 10^3$	3.4 $\times 10^3$	6.6 $\times 10^3$	6.9 $\times 10^3$	4.3 $\times 10^3$	4.3 $\times 10^3 \pm 0.84$
RM5	4.9 $\times 10^3$	3.9 $\times 10^3$	5.5 $\times 10^3$	5.8 $\times 10^3$	3.7 $\times 10^3$	5.6 $\times 10^3$	4.9 $\times 10^3 \pm 0.37$
RM6	6.5 $\times 10^3$	2.3 $\times 10^3$	3.2 $\times 10^3$	5.1 $\times 10^3$	2.1 $\times 10^3$	7.1 $\times 10^3$	4.4 $\times 10^3 \pm 0.88$
RM7	2.3 $\times 10^3$	1.8 $\times 10^3$	2.9 $\times 10^3$	4.1 $\times 10^3$	4.8 $\times 10^3$	2.9 $\times 10^3$	3.1 $\times 10^3 \pm 0.46$
RM8	3.6 $\times 10^3$	1.1 $\times 10^3$	1.5 $\times 10^3$	6.0 $\times 10^3$	2.4 $\times 10^3$	5.2 $\times 10^3$	3.3 $\times 10^3 \pm 0.81$
RM9	3.8 $\times 10^3$	2.5 $\times 10^3$	6.0 $\times 10^3$	4.2 $\times 10^3$	6.0 $\times 10^3$	4.4 $\times 10^3$	4.5 $\times 10^3 \pm 0.55$
RM10	5.4 $\times 10^3$	1.4 $\times 10^3$	4.6 $\times 10^3$	5.5 $\times 10^3$	4.7 $\times 10^3$	5.1 $\times 10^3$	4.5 $\times 10^3 \pm 0.63$
RM11	6.0 $\times 10^3$	1.9 $\times 10^3$	5.2 $\times 10^3$	7.7 $\times 10^3$	3.1 $\times 10^3$	6.6 $\times 10^3$	5.1 $\times 10^3 \pm 0.90$
RM12	7.6 $\times 10^3$	3.3 $\times 10^3$	3.3 $\times 10^3$	5.8 $\times 10^3$	6.0 $\times 10^3$	4.2 $\times 10^3$	5.0 $\times 10^3 \pm 0.70$
RM13	2.8 $\times 10^3$	1.7 $\times 10^3$	5.9 $\times 10^3$	5.8 $\times 10^3$	4.7 $\times 10^3$	5.5 $\times 10^3$	4.4 $\times 10^3 \pm 0.72$
RM14	6.4 $\times 10^3$	2.4 $\times 10^3$	1.8 $\times 10^3$	4.3 $\times 10^3$	3.1 $\times 10^3$	4.4 $\times 10^3$	3.7 $\times 10^3 \pm 0.68$
T3	14.4 $\times 10^3$	8.8 $\times 10^3$	9.1 $\times 10^3$	11.2 $\times 10^3$	4.0 $\times 10^3$	13.8 $\times 10^3$	10.2 $\times 10^3 \pm 1.56$
RM15	7.1 $\times 10^3$	1.6 $\times 10^3$	3.8 $\times 10^3$	3.3 $\times 10^3$	4.5 $\times 10^3$	6.5 $\times 10^3$	4.5 $\times 10^3 \pm 0.84$
RM16	5.7 $\times 10^3$	1.2 $\times 10^3$	2.2 $\times 10^3$	5.8 $\times 10^3$	3.8 $\times 10^3$	4.8 $\times 10^3$	3.9 $\times 10^3 \pm 0.77$
RM17	6.6 $\times 10^3$	2.3 $\times 10^3$	4.7 $\times 10^3$	3.7 $\times 10^3$	2.3 $\times 10^3$	3.1 $\times 10^3$	3.8 $\times 10^3 \pm 0.67$
RM18	4.9 $\times 10^3$	1.5 $\times 10^3$	2.8 $\times 10^3$	5.7 $\times 10^3$	4.8 $\times 10^3$	4.2 $\times 10^3$	3.9 $\times 10^3 \pm 0.63$
	6.4 $\times 10^3 \pm$ 0.91	3.1 $\times 10^3 \pm$ 0.52	4.8 $\times 10^3 \pm$ 0.77	5.9 $\times 10^3 \pm$ 0.51	5.2 $\times 10^3 \pm$ 0.59	6.4 $\times 10^3 \pm$ 0.94	

Key: RM-Room, T-Toilet, Hostel A – Hall 3

The bacterial isolates were characterized and identified, which include the following *Staphylococcus aureus*, *Klebsiella pneumonia*, *Acinetobacter haemolyticus*, *Bacillus subtilis*, *Streptococcus pyogenes*, *Pseudomonas aeruginosa*, *Aerococcus viridians*, *Corynebacterium* sp., *Proteus vulgaris*, *E. coli*, *Staphylococcus epidermidis*, *Micrococcus* sp., *Enterobacter aerogenes* and *Serratia marcescens*. This report is similar to the report by Lynn *et al.*, (2013), who isolated *Staphylococcus aureus*, *Klebsiella* sp., *E coli*, *Proteus* spp. from door handles in a public secondary school. All isolates except for *Micrococcus* spp., *Acinetobacter* sp. and *Aerococcus* sp. are basic commensals or normal flora in or on the body while *Bacillus* spp., *S. aureus* and *S. epidermidis* are found on the skin and *Pseudomonas* spp. on the outer ear. This supports the claim that, these organisms are normally not pathogenic, with low potential to cause diseases except when introduced into body tissues which could lead to bacteremia (Schito, 2006) and this can be as a result of poor personal hygiene and lifestyle as supported by Deodhar, (2003). *Bacillus* spp., have been linked to gastrointestinal disorders

when ingested by the production of toxins. Both *S. aureus* and *S. epidermidis* are abundant flora on the skin surface, and there is the tendency to drop them on environmental surfaces. Strains of *Staphylococcus aureus* which produce enterotoxin have been implicated in food poisoning. *Klebsiella pneumoniae*, *Serratia marcescens*, *Enterobacter aerogenes* as well *E. coli* has a fecal origin. These bacteria may result in urinary tract infection and when consumed through contaminated food, can cause gastroenteritis and diarrhea. *Klebsiella pneumoniae* causes a severe pneumonia. The likely reason is poor hand washing practice after usage of toilet which means that people spread fecal bacteria to everything else they touch around them. The predominant bacterial isolates from the door handles was *Staphylococcus aureus* 23.61% while *Aerococcus viridians* had the least occurrence of 0.24% (Table 4). The results revealed that the high load bacteria on door handles could be attributed to increase student population using door handles during periods of high school activities hence, the high frequency of *S. aureus* occurrence. Jawad *et al.* (1996) also noted the effect of relative humidity on the survival of *Staphylococcus aureus* and concluded that *S. aureus* can survive on dry surfaces and be transferred not only by moist factors but also in dry conditions in the hospital environment during nosocomial infection outbreaks. *S. aureus* is innocuous in most environment, but with remarkable adaptability and versatility which has equipped the organism as a commensal and pathogen.

Table 2: Total viable bacterial counts on door handle in Hostel B

Samples	April	May	June	July	August	September	Mean \pm S.E
RM1	9.2×10^3	2.8×10^3	6.3×10^3	2.8×10^3	3.3×10^3	3.5×10^3	$4.7 \times 10^3 \pm 1.06$
RM2	6.7×10^3	4.6×10^3	5.5×10^3	3.6×10^3	4.2×10^3	5.1×10^3	$4.9 \times 10^3 \pm 0.44$
RM3	8.8×10^3	7.1×10^3	5.6×10^3	7.1×10^3	4.5×10^3	6.8×10^3	$6.7 \times 10^3 \pm 0.59$
RM4	5.9×10^3	3.1×10^3	4.9×10^3	2.9×10^3	3.7×10^3	5.2×10^3	$4.3 \times 10^3 \pm 0.49$
T1	11.2×10^3	7.7×10^3	10.8×10^3	7.3×10^3	9.4×10^3	16.4×10^3	$10.5 \times 10^3 \pm 1.35$
T2	13.7×10^3	7.5×10^3	12.2×10^3	7.8×10^3	9.2×10^3	1.5×10^3	$8.7 \times 10^3 \pm 1.75$
RM5	7.8×10^3	4.4×10^3	3.1×10^3	4.4×10^3	3.6×10^3	7.8×10^3	$5.2 \times 10^3 \pm 0.85$
RM6	9.7×10^3	4.8×10^3	5.8×10^3	6.2×10^3	7.7×10^3	9.2×10^3	$7.2 \times 10^3 \pm 0.80$
RM7	8.2×10^3	2.3×10^3	9.1×10^3	3.7×10^3	2.5×10^3	4.8×10^3	$5.1 \times 10^3 \pm 1.19$
RM8	5.6×10^3	1.7×10^3	5.3×10^3	2.9×10^3	3.1×10^3	6.3×10^3	$4.2 \times 10^3 \pm 0.75$
RM9	7.7×10^3	3.5×10^3	8.0×10^3	4.5×10^3	4.8×10^3	7.7×10^3	$6.0 \times 10^3 \pm 0.81$
RM10	8.9×10^3	3.4×10^3	4.3×10^3	3.4×10^3	2.6×10^3	4.5×10^3	$4.5 \times 10^3 \pm 0.92$
RM11	6.1×10^3	4.8×10^3	2.1×10^3	4.8×10^3	3.3×10^3	6.3×10^3	$4.6 \times 10^3 \pm 0.66$
T3	14.8×10^3	6.2×10^3	11.5×10^3	11.2×10^3	10.3×10^3	15.5×10^3	$11.6 \times 10^3 \pm 1.37$
RM12	9.9×10^3	3.6×10^3	3.3×10^3	4.5×10^3	4.7×10^3	5.4×10^3	$5.2 \times 10^3 \pm 0.98$
RM13	5.3×10^3	2.9×10^3	7.5×10^3	3.3×10^3	2.5×10^3	6.1×10^3	$4.6 \times 10^3 \pm 0.82$
RM14	8.7×10^3	5.4×10^3	3.1×10^3	5.4×10^3	4.4×10^3	7.2×10^3	$5.7 \times 10^3 \pm 0.81$
RM15	9.6×10^3	3.1×10^3	6.7×10^3	4.2×10^3	2.8×10^3	7.5×10^3	$5.7 \times 10^3 \pm 1.1$
RM16	7.4×10^3	5.5×10^3	7.1×10^3	6.6×10^3	4.5×10^3	8.3×10^3	$6.6 \times 10^3 \pm 0.56$
RM17	6.5×10^3	4.8×10^3	4.0×10^3	5.1×10^3	3.8×10^3	5.4×10^3	$4.9 \times 10^3 \pm 0.40$
RM18	5.7×10^3	3.4×10^3	6.2×10^3	2.6×10^3	4.4×10^3	6.2×10^3	$4.8 \times 10^3 \pm 0.62$
	$8.5 \times 10^3 \pm 0.55$	$4.4 \times 10^3 \pm 0.37$	$6.3 \times 10^3 \pm 0.61$	$4.9 \times 10^3 \pm 0.46$	$4.7 \times 10^3 \pm 0.51$	$7.0 \times 10^3 \pm 0.75$	

Key: RM-Room T-Toilet Hostel B – Hall 4

Table 3: Total viable bacterial counts on door handle in Hostel C

Samples	April	May	June	July	August	September	Mean \pm S.E
RM3	4.4×10^3	2.7×10^3	5.1×10^3	1.5×10^3	4.3×10^3	1.5×10^3	$3.3 \times 10^3 \pm 0.64$
RM4	3.1×10^3	1.5×10^3	2.7×10^3	3.2×10^3	5.4×10^3	3.7×10^3	$3.3 \times 10^3 \pm 0.52$
T2	8.3×10^3	6.1×10^3	6.3×10^3	9.0×10^3	6.8×10^3	11.9×10^3	$8.1 \times 10^3 \pm 0.89$
T6	5.6×10^3	1.7×10^3	7.4×10^3	10.8×10^3	7.6×10^3	13.1×10^3	$7.7 \times 10^3 \pm 1.63$
T7	8.2×10^3	6.8×10^3	9.0×10^3	2.1×10^3	2.5×10^3	9.8×10^3	$6.4 \times 10^3 \pm 1.36$
RM10	4.2×10^3	1.7×10^3	4.8×10^3	1.2×10^3	8.0×10^3	5.5×10^3	$4.2 \times 10^3 \pm 1.03$
RM8	2.5×10^3	8.3×10^3	5.1×10^3	1.6×10^3	3.4×10^3	6.7×10^3	$4.6 \times 10^3 \pm 1.05$
T10	6.0×10^3	6.8×10^3	10.2×10^3	9.5×10^3	7.1×10^3	8.8×10^3	$8.1 \times 10^3 \pm 0.68$
RM2	3.3×10^3	1.7×10^3	3.7×10^3	7.6×10^3	4.9×10^3	2.5×10^3	$3.9 \times 10^3 \pm 0.85$
RM1	2.9×10^3	3.1×10^3	2.3×10^3	1.8×10^3	6.8×10^3	4.5×10^3	$3.6 \times 10^3 \pm 0.75$
T3	8.3×10^3	6.7×10^3	8.1×10^3	6.2×10^3	7.9×10^3	11.0×10^3	$8.0 \times 10^3 \pm 0.68$
T8	5.1×10^3	6.3×10^3	9.6×10^3	5.1×10^3	6.3×10^3	10.8×10^3	$7.2 \times 10^3 \pm 0.99$
T5	6.4×10^3	5.1×10^3	7.8×10^3	4.6×10^3	5.5×10^3	9.2×10^3	$6.4 \times 10^3 \pm 0.72$
T9	10.4×10^3	4.6×10^3	9.3×10^3	4.1×10^3	8.0×10^3	10.2×10^3	$7.8 \times 10^3 \pm 1.14$
RM7	7.9×10^3	2.1×10^3	3.8×10^3	6.2×10^3	7.4×10^3	5.5×10^3	$5.5 \times 10^3 \pm 0.90$
RM11	1.6×10^3	1.0×10^3	4.4×10^3	4.3×10^3	3.1×10^3	2.8×10^3	$2.8 \times 10^3 \pm 0.56$
RM5	2.2×10^3	4.4×10^3	3.7×10^3	1.5×10^3	2.0×10^3	3.9×10^3	$3.0 \times 10^3 \pm 0.49$
T4	4.3×10^3	8.0×10^3	10.6×10^3	6.2×10^3	5.1×10^3	12.8×10^3	$7.8 \times 10^3 \pm 1.35$
T1	5.5×10^3	6.3×10^3	8.3×10^3	5.8×10^3	1.7×10^3	11.2×10^3	$6.5 \times 10^3 \pm 1.29$
RM6	1.8×10^3	1.2×10^3	3.6×10^3	7.1×10^3	3.5×10^3	2.0×10^3	$3.2 \times 10^3 \pm 0.87$
RM9	2.4×10^3	1.4×10^3	4.8×10^3	5.3×10^3	2.9×10^3	3.4×10^3	$3.4 \times 10^3 \pm 0.60$
	$4.9 \times 10^3 \pm 0.55$	$4.2 \times 10^3 \pm 0.55$	$6.2 \times 10^3 \pm 0.57$	$4.9 \times 10^3 \pm 0.62$	$5.2 \times 10^3 \pm 0.46$	$7.2 \times 10^3 \pm 0.85$	

Key: RM- Room, T- Toilet. Hostel C – Hall 5

Table 4: Percentage occurrence of the bacterial isolates of door handles

Bacterial isolates	April	May	June	July	August	September	Frequency N=1245	Frequency (%)
<i>Streptococcus pyogenes</i>	42	26	47	47	40	46	248	19.92
<i>Klebsiella pneumonia</i>	29	22	21	26	32	22	152	12.21
<i>Staphylococcus aureus</i>	43	41	52	54	51	53	294	23.61
<i>Pseudomonas</i>	18	32	31	15	17	27	140	11.24
<i>Staphylococcus</i>	40	10	40	41	47	39	217	17.43
<i>Micrococcus</i> sp.	7	13	4	0	6	0	30	2.41
<i>Corynebacterium</i> sp.	0	7	0	0	0	0	7	0.56
<i>Bacillus subtilis</i>	6	28	10	0	0	0	44	3.53
<i>E.coli</i>	0	15	3	3	0	10	31	2.49
<i>Proteus vulgaris</i>	15	9	7	10	7	7	55	4.42
<i>Acinetobacter</i>	0	4	0	0	0	0	4	0.32
<i>Serratia marcescens</i>	0	0	0	0	5	0	5	0.40
<i>Enterobacter aerogenes</i>	0	0	0	10	0	5	15	1.20
<i>Aerococcus viridians</i>	0	0	0	0	0	3	3	0.24

The results of the pre and post curing susceptibility test of the isolates revealed that most of the antibiotics used like chloramphenicol, augmentin, ampiclox, streptomycin, septrin, pefloxacin and amoxicillin were resisted by all the isolates except *S. epidermidis* which

showed increased susceptibility to Gentamycin, ciprofloxacin and amoxicillin after curing (Tables 5 and 6).

Table 5: Inhibition zones (diameter in mm) for the sensitivity testing of bacterial isolates against antibiotics before curing

G+ve	PEF	CN	APX	Z	AM	R	CPX	S	SXT	E
<i>Bacillus subtilis</i>	R	S	R	R	R	R	S	R	R	S
<i>Streptococcus</i>	R	S	R	S	R	S	S	R	R	S
<i>Corynebacterium</i> sp.	R	S	R	R	R	R	S	R	R	R
<i>S.aureus</i>	R	S	R	S	R	S	S	R	R	R
<i>Micrococcus</i> sp.	R	S	R	S	R	R	S	R	R	R
<i>Aerococcus viridians</i>	R	S	R	S	R	R	S	R	R	R
<i>S.epidermidis</i>	R	R	R	R	R	R	R	R	R	R
Gram –ve	SXT	CH	SP	CPX	AM	AU	CN	PEF	OFX	S
<i>Pseudomonas</i>	R	R	R	R	R	R	S	R	S	R
<i>Serratia mercesens</i>	R	R	R	S	R	R	S	R	S	R
<i>Enterobacter</i>	R	R	R	S	R	R	S	R	S	R
<i>Klebsiella pneumoniae</i>	R	R	R	R	R	R	S	R	S	R
<i>Proteus vulgaris</i>	R	R	R	R	R	R	S	R	S	R
<i>Acinetobacter</i>	R	R	R	S	R	R	S	R	S	R
<i>E.coli</i>	R	R	R	S	R	R	S	R	S	R

Key: SXT- Septrin(30µg), CH- Chloraphenicol(30µg), SP- Sparfloxacin(10µg), CPX- Ciprofloxacin(10µg), AM- Amoxicillin(30µg), AU- Augmentin, CN-Gentamycin(10µg), PEF- Perloxacin(10µg), S – Streptomycin(30µg), Z- Zinnacef(20µg), R- Rocephin(25µg), E- Erythromycin(10µg), OFX- Ofloxacin(10µg), APX- Ampiclox(30µg), S-Susceptible (15- 23) R-Resistant (≤15)

Table 6: Inhibition zones (diameter in mm) for the sensitivity testing of bacterial isolates against antibiotics after curing

G+ve	PEF	CN	APX	Z	AM	R	CPX	S	SXT	E
<i>Bacillus</i>	R	S	R	S	R	R	S	S	R	S
<i>Streptococcus</i>	R	S	R	S	R	S	S	R	R	S
<i>Corynebacterium</i> sp.	R	S	R	S	R	R	S	S	R	R
<i>S.aureus</i>	R	S	R	S	R	S	S	S	R	R
<i>Micrococcus</i> sp.	R	S	R	S	R	R	S	S	R	R
<i>Aerococcus viridians</i>	R	S	R	S	R	R	S	R	R	R
<i>S.epidermidis</i>	R	S	R	R	S	R	S	R	R	R
Gram –ve	SXT	CH	SP	CPX	AM	AU	CN	PEF	OFX	S
<i>Pseudomonas aerogunosa</i>	R	R	R	R	R	R	S	R	S	R
<i>Serratia mercesens</i>	R	R	R	S	R	R	S	R	S	S
<i>Enterobacter aerogines</i>	R	R	R	S	R	R	S	R	S	R
<i>Klebsiella pneumoniae</i>	R	R	S	S	R	R	S	R	S	R
<i>Proteus vulgaris</i>	R	R	S	S	R	R	S	R	S	R
<i>Acinetobacter haemolyticus</i>	R	R	S	S	R	R	S	R	S	R
<i>E.coli</i>	R	R	S	S	R	R	S	R	S	R

Key: SXT- Septrin (30µg), CH- Chloraphenicol (30µg), SP- Sparfloxacin (10µg), CPX- Ciprofloxacin (10µg), AM- Amoxicillin (30µg), AU- Augmentin, CN-Gentamycin (10µg), PEF- Perloxacin (10µg), S – Streptomycin (30µg), Z- Zinnacef (20µg), R- Rocephin (25µg), E- Erythromycin (10µg), OFX- Ofloxacin (10µg), APX- Ampiclox (30µg), S- Susceptible (15- 23) R-Resistant (≤15)

The plasmids analyses revealed detectable plasmid fragments in *Escherichia coli*, *Pseudomonas aeruginosa*, *Corynebacterium* sp, *Staphylococcus epidermidis*, *Staphylococcus aureus*, *Proteus vulgaris*, *Enterobacter* sp, *Klebsiella pneumonia* (Plate1). These observations are similar to the report of Nwankwo and Afuruobi (2015), who reported antibiotic resistant bacteria on door handles in a tertiary institution in Abia State. This also suggest that resistance to these antibiotics, that some of the isolates were plasmid mediated Drug resistance and could been / attributed to the misuse and overuse (of antibiotics) as well as the possession of drug resistance plasmids (Madhavan and Sowmiyan, 2011).

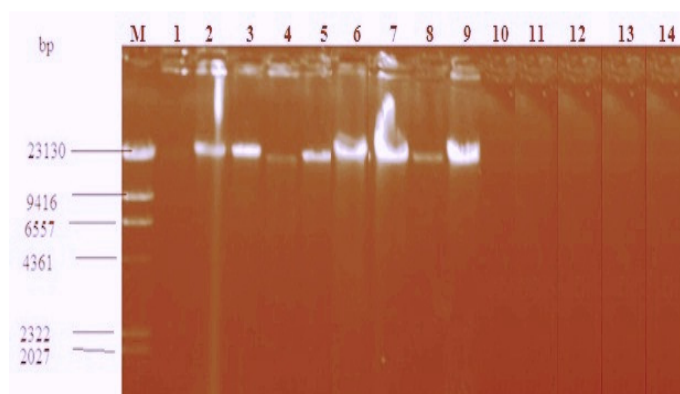


Plate 1: Plasmid DNA Profile of Bacteria Isolates from Door Handle

Lane 1: *Bacillus subtilis*, Lane 2= *Escherichia coli*, Lane 3= *Pseudomonas aeruginosa*, Lane 4= *Corynebacterium* sp., Lane 5 *Staphylococcus epidermidis*, Lane 6= *Staphylococcus aureus*, Lane 7= *Proteus vulgaris*, Lane 8 = *Enterobacter* sp., Lane 9= *Klebsiella pneumonia*, Lane 10= *Streptococcus pyogenes*, lane 11= *Micrococcus* sp., Lane 12= *Acinetobacter haemolyticus*, Lane 13= *Serratia mercesens*, Lane 14= *Aerococcus viridians*, Lane M= λ Hind III digest molecular weight marker

4. CONCLUSION

The results of this study showed the presence of bacterial contaminants on door handles of three student hostels of University of Benin. The presence of drug resistant bacteria on the door handles is of significant health concern on account of its capability to bring about epidemics. Preventing this will require health education, community health superintendents, sanitary officers and Environmental Protection Board and in addition private organizations to enlighten the populace on personal and environmental hygiene. Likewise, to reduce the frequency of drug resistance, the carelessness usage of antibiotics by individuals whose health are impaired needs serious action for health education of the consequence of drug abuse. It is recommended that, more researches be carried out in this area to prevent infection outbreak.

5. CONFLICT OF INTEREST

There is no conflict of interest associated with this work.

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