

INFLUENCE OF DOOR HANDLES ON THE PROLIFERATION OF MICROORGANISMS OF PUBLIC HEALTH IMPORTANCE IN UNIVERSITY OF BENIN STAFF OFFICES.

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ABSTRACT

The aim of this research was to determine the role of door handles in the proliferation of microorganisms in staff offices within University of Benin, Benin City. Isolation and characterization of isolates was carried out using standard methods. The results of this study showed that surfaces of door handles of Plant Biology and Biotechnology, Microbiology and Chemistry Departments had high bacterial count, which ranged from $6.17 \pm 0.75 \times 10^4$ cfu/cm² - $7.45 \pm 0.67 \times 10^4$ cfu/cm², while Geology, Physics and Adult Education had low counts ($3.31 \pm 0.64 \times 10^4$ cfu/cm² - $3.55 \pm 0.80 \times 10^4$ cfu/cm²). The bacterial isolates were: *Staphylococcus epidermidis*, *Citrobacter* sp., *Escherichia coli*, *Enterobacter* sp., *Staphylococcus aureus*, *Klebsiella* sp., *Streptococcus pyogenes*, *Corynebacterium* sp., *Bacillus subtilis*, *Pseudomonas aeruginosa* and *Proteus vulgaris*, while the fungal isolates were *Rhizopus* sp., *Aspergillus niger*, *Fusarium* sp., *Penicillium* sp. and *Mucor* sp. The most predominant bacterial isolate was *Staphylococcus aureus* (25.22%), while *Citrobacter* sp. (1.74%) was the least. Plasmid profile revealed plasmid fragments in all the bacterial isolates, with most of the isolates, being multi-drug resistant. Results showed that door handles harbor pathogenic organisms. Therefore, everyone is encouraged to have good personal hygiene, use of hand-sanitizers, hand-washing before and after the use of door handles and routine surface disinfection of door handles in order to prevent cross contamination.

Keywords: ADHD, Conner's Teachers' Rating Scale, Primary School

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Fomites, when in constant contact with humans or natural habitats of pathogenic organism constitute a major source of spread of infectious diseases (Osterholm et al., 1995). The fomites include door handle of conveniences, showers, toilet, hand lockers especially those found in public offices, hospitals, hotels, restaurants and restrooms (Bright et al., 2010). Beside the day to day interaction of people, which constitute one way of spreading disease, the major source of spread of community acquired infections are fomites (Prescott et al., 1993). Microorganisms are found everywhere, bacteria and fungi contaminate our body, our houses, work places, and whole environment. Microorganisms such as bacterial and fungal spores are almost always present in the air. The quality of indoor environment, however, is not easily defined or readily controlled, and can potentially place human occupants at risk (Aladenika et al., 2014). Airborne transmission is one of the routes of spreading diseases responsible for a number of nosocomial infections (Ekhaise and Ogboghodo, 2011). Microorganisms constitute a major part of every ecosystem. In these environments, they live either freely or as parasites (Sleigh and Timbury, 1998). The study of airborne microorganisms in indoor environments is important to understand the dissemination of airborne microbes particularly the pathogenic ones (Ekhaise and Ogboghodo, 2011). The number and type of airborne microorganisms can be used to determine the degree of cleanliness. The hand serves as a medium for the propagation of microorganisms from place to place and from person to person. Although it is nearly impossible for the hand to be free of microorganisms, the presence of pathogenic bacteria may lead to chronic or acute illness (Oranusi et al., 2013). Human hands usually harbor microorganisms both as part of body normal flora as well as transient microbes contacted from the environment. In the university environment, students have access to service offices regularly for different purposes. Given that the door handles are not routinely disinfected, the opportunity for the transmission of contaminating microorganisms is great. Although it is accepted that the infection risk in general community is less than that associated with patients in hospital. The increasing incidence of epidemic outbreaks of certain diseases and its rate of spread from one community to the other has become a major public health concern (Oranusi et al., 2013).

This study was therefore aimed at investigating the Influence of door handles on the proliferation of microorganisms in University of Benin staff offices, Ugbowo Campus, Benin City.

MATERIALS AND METHODS

Study Area

The study area, University of Benin, Benin-City, Nigeria, lies within longitude 5.62 o E and latitude 6.4 ON, 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. SAMPLE COLLECTION Ten (10) sampling units were used for the study. They included: Microbiology Department, Animal and Environmental Biology Department, Plant Biology and Biotechnology Department, Biochemistry Department, Chemistry Department, Physics Department, Geology Department, Mathematics Department, Faculty of Education and Faculty of Agricultural Sciences. A total of three hundred (300) samples were randomly collected with the aid of sterile swab sticks moistened with normal saline for a period of six months (March 2017 – August 2017). The swab sticks were inserted into 10 ml of sterile peptone water contained in a sterile test tube and transported to the Laboratory.

ENUMERATION OF BACTERIA

The enumeration of the microorganisms was carried out in accordance to the methods described by Chesebrough (2000) using standard plate count procedures. Serial dilution of the sample was aseptically carried out by pipetting 1ml 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. Aliquot of the 10-3 dilution was plated in nutrient agar amended with nystatin for isolation of bacteria and potato dextrose agar amended with streptomycin for isolation of fungi. The plates were inoculated in duplicates. The inoculated nutrient agar plates were incubated at 28 OC for 24 hrs while the potato dextrose agar plates were incubated at 28 OC for 72 hrs. After incubation, the colonies of the isolates were counted and expressed in cfu/g and sfu/g for bacteria and fungi respectively. Isolated colonies were further purified by sub-culturing and identified using biochemical tests.

CHARACTERISATION OF BACTERIAL ISOLATES

Bacterial isolates were characterized and identified after studying their Gram reaction as well as morphological characteristics. Other tests performed were spore formation, motility, and catalase production. Citrate utilization, oxidative/fermentative utilization of glucose, indole production, methyl red - Voges Proskauer reaction, urease and coagulase production, starch hydrolysis, production of H₂S from triple sugar iron (TSI) agar and sugar fermentation. Characterisation of bacterial isolates were carried out according to Cheesbrough (2000) and Holt et al. (1994). CHARACTERIZATION OF FUNGAL ISOLATES The fungal species were identified and characterized based on their morphological characteristics and microscopic analysis by using taxonomic guides and standard procedures (Barnett and Hunter, 1972; Domsch et al., 1980; Ellis, 1976; Gilman, 1944). ANTIBIOTIC SUSCEPTIBILITY TEST 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 (Bauer 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).

PLASMID PROFILING

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 0.04M Tris-acetate, (pH 8.0) and 2 mM EDTA. Also, a volume of 2ml lyses buffer (0.05M Tris, 3% SDS, pH 12.5, adjusted with 2N NaOH and mixed gently, the suspension was incubate at 60-68°C for 30-45 min. An amount 6ml of phenol/chloroform (1:1) was added to the hot samples and mixed gently to complete emulsification. Phases were separated by centrifugation at 10000 rev/s for 15-20 mins, and the upper aqueous phase was transferred carefully to new microfuge tube containing 400 µl 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).

PLASMID CURING

The isolates that showed resistance to the antibiotics used (Septrin (30µg), Chloraphenicol (30µg), Sparfloxacin (10µg), Ciprofloxacin (10µg), Amoxicilin (30µg), Augmentin, Gentamycin (10µg), Perfloxacin (10µg), Streptomycin (30µg), Zinnacef (20µg), Rocephin (25µg), Erythromycin (10µg), Ofloxacin (10µg), Ampiclox (30µg), were subjected to standard plasmid curing method (Sijhary et al., 1984). Overnight broth culture was inoculated into 4.5ml nutrient broth and 0.5ml of sodium dodecyl sulphate was added and incubated at 37°C for 48 hrs. An aliquot, 0.5ml of the broth was added to a freshly prepared 4.5ml nutrient broth, incubated for another 24 hrs at 37 °C after which post-plasmid curing 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.

STATISTICAL ANALYSIS

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).

RESULTS

The results showed positive for bacterial and fungal contamination for all samples. The bacterial count ranged from $1.35 \pm 0.46 \times 10^4$ cfu/cm² - $6.97 \pm 0.77 \times 10^4$ cfu/cm² for Microbiology, $1.08 \pm 0.49 \times 10^4$ cfu/cm² - $6.12 \pm 0.91 \times 10^4$ cfu/cm² for Animal and Environmental Biology, $1.69 \pm 0.33 \times 10^4$ cfu/cm² - $7.45 \pm 0.67 \times 10^4$ cfu/cm² for Plant Biology and Biotechnology, $1.02 \pm 0.83 \times 10^4$ cfu/cm² - $5.79 \pm 0.76 \times 10^4$ cfu/cm² for Biochemistry, $1.02 \pm 0.82 \times 10^4$ cfu/cm² - $6.17 \pm 0.75 \times 10^4$ cfu/cm² for Chemistry, $1.86 \pm 0.33 \times 10^4$ cfu/cm² - $3.47 \pm 0.64 \times 10^4$ cfu/cm² for Physics, $1.10 \pm 0.77 \times 10^4$ cfu/cm²

TABLE 1: TOTAL VIABLE BACTERIAL COUNTS ($\times 10^4$ cfu/cm²)

Departments	March	April	May	June	July	August	X \pm S.E	pValue
Microbiology	6.97 \pm 0.77	1.35 \pm 0.46	2.08 \pm 0.91	6.77 \pm 0.26	2.54 \pm 0.50	3.61 \pm 0.78	3.89 \pm 0.99	0.511
AEB	6.12 \pm 0.91	2.37 \pm 0.75	1.08 \pm 0.49	3.70 \pm 0.41	2.36 \pm 0.37	2.97 \pm 0.94	3.10 \pm 0.70	0.511
PBB	7.45 \pm 0.67	1.69 \pm 0.28	3.47 \pm 0.17	1.69 \pm 0.33	1.83 \pm 0.43	3.44 \pm 0.67	3.26 \pm 0.91	0.511
Biochemistry	5.79 \pm 0.76	1.72 \pm 0.14	1.53 \pm 0.35	1.18 \pm 0.56	1.36 \pm 0.95	1.02 \pm 0.83	2.10 \pm 0.74	0.511
Chemistry	6.17 \pm 0.75	2.80 \pm 0.37	1.02 \pm 0.82	3.47 \pm 0.21	1.15 \pm 0.34	1.77 \pm 0.62	2.73 \pm 0.79	0.511
Physics	3.47 \pm 0.64	1.94 \pm 0.43	2.20 \pm 0.76	1.86 \pm 0.33	2.30 \pm 0.53	3.08 \pm 0.29	2.48 \pm 0.27	0.511
Geology	3.55 \pm 0.80	1.10 \pm 0.77	1.47 \pm 0.62	1.62 \pm 0.48	1.94 \pm 0.38	2.46 \pm 0.41	2.02 \pm 0.36	0.511
Mathematics	4.34 \pm 0.77	3.13 \pm 0.51	1.31 \pm 0.67	2.77 \pm 0.74	1.38 \pm 0.82	1.79 \pm 0.75	2.45 \pm 0.48	0.511
Adult Edu.	3.31 \pm 0.64	1.80 \pm 0.47	2.79 \pm 0.55	1.01 \pm 0.82	1.19 \pm 0.57	1.25 \pm 0.87	1.89 \pm 0.39	0.511
FW	3.64 \pm 0.56	1.79 \pm 0.39	1.62 \pm 0.42	2.03 \pm 0.29	3.46 \pm 0.32	2.35 \pm 0.50	2.48 \pm 0.35	0.511

Note: No significant difference between bacterial counts in the different months ($P > 0.05$).

Key: AEB = Animal and Environmental Biology PBB = Plant Biology and Biotechnology FW = Forestry and Wildlife

TABLE 2: TOTAL FUNGAL COUNTS $\times 10^3$ (sfu/cm²)

Microbiology		4.0 \pm 0.80	1.7 \pm 0.80	5.2 \pm 0.50	2.1 \pm 0.25	2.7 \pm 0.17	3.23 \pm 0.54	0.071
AEB		3.0 \pm 0.12	4.3 \pm 0.20	5.7 \pm 0.66	1.1 \pm 0.51	3.2 \pm 0.32	3.30 \pm 0.64	0.071
PBB		1.1 \pm 0.30	6.0 \pm 0.13	1.1 \pm 0.42	1.8 \pm 0.57	2.0 \pm 0.12	2.48 \pm 0.75	0.071
Biochemistry		8.0 \pm 0.11	2.0 \pm 0.43	4.2 \pm 0.21	2.0 \pm 0.20	3.6 \pm 0.42	3.57 \pm 0.98	0.071
Chemistry		5.0 \pm 0.12	3.8 \pm 0.60	2.5 \pm 0.57	3.2 \pm 0.15	2.8 \pm 0.36	3.62 \pm 0.39	0.071
Physics		1.1 \pm 0.80	5.0 \pm 0.19	5.1 \pm 0.11	1.2 \pm 0.11	1.9 \pm 0.27	3.30 \pm 0.86	0.071
Geology		1.2 \pm 0.40	1.9 \pm 0.66	4.1 \pm 0.41	2.8 \pm 0.18	1.1 \pm 0.32	2.37 \pm 0.48	0.071
Mathematics		6.0 \pm 0.60	1.6 \pm 0.17	2.9 \pm 0.31	1.7 \pm 0.28	3.3 \pm 0.20	3.23 \pm 0.66	0.071
Adult Education		8.0 \pm 0.50	6.6 \pm 0.16	3.1 \pm 0.52	4.2 \pm 0.42	5.9 \pm 0.11	5.22 \pm 0.79	0.071
Forestry and Wildlife		1.2 \pm 0.11	1.3 \pm 0.38	2.1 \pm 0.29	1.5 \pm 0.21	2.7 \pm 0.59	1.75 \pm 0.23	0.071

Note: No significant difference between fungal counts in the different months ($P > 0.05$).

Key: AEB = Animal and Environmental Biology PBB = Plant Biology and Biotechnology

The fungal count ranged from 1.7 \pm 0.80 $\times 10^3$ cfu/cm² - 5.2 \pm 0.50 $\times 10^3$ cfu/cm², 1.1 \pm 0.51 $\times 10^3$ cfu/cm² - 5.7 \pm 0.66 $\times 10^3$ cfu/cm², 1.1 \pm 0.30 $\times 10^3$ cfu/cm² - 6.0 \pm 0.13 $\times 10^3$ cfu/cm², 1.6 \pm 0.19 $\times 10^3$ cfu/cm² - 8.0 \pm 0.11 $\times 10^3$ cfu/cm², 2.5 \pm 0.57 $\times 10^3$ cfu/cm² - 5.0 \pm 0.12 $\times 10^3$ cfu/cm², 1.1 \pm 0.80 $\times 10^3$ cfu/cm² - 5.5 \pm 0.01 $\times 10^3$ cfu/cm² 1.1 \pm 0.32 $\times 10^3$ cfu/cm² - 4.1 \pm 0.41 $\times 10^3$ cfu/cm², 1.6 \pm 0.17 $\times 10^3$ cfu/cm² - 6.0 \pm 0.6 $\times 10^3$ cfu/cm² 3.1 \pm 0.52 $\times 10^3$ cfu/cm² - 8.0 \pm 0.50 $\times 10^3$ cfu/cm² and 1.2 \pm 0.11 $\times 10^3$ cfu/cm² - 2.7 \pm 0.59 $\times 10^3$ cfu/cm² for the,,

door handles of Microbiology, Animal and Environmental Biology, Plant Biology and Biotechnology, Biochemistry, Chemistry, Physics, Geology, Mathematics, Adult Education and Forestry and Wildlife Departments. There was no significant difference between fungal counts in the different months ($P > 0.05$) (Table 2). The results of this study showed that surfaces of door handles of Plant Biology and Biotechnology Department, Microbiology Department and Chemistry Department had very high viable bacterial counts, which ranged from $6.17 \pm 0.75 \times 10^4$ cfu/cm² - $7.45 \pm 0.67 \times 10^4$ cfu/cm², while Geology Department, Physics Department and Faculty of Education recorded the least bacterial load that ranged from $3.31 \pm 0.64 \times 10^4$ cfu/cm² - $3.55 \pm 0.80 \times 10^4$ cfu/cm². The bacterial isolates recovered from the door handles were Staphylococcus epidermidis, Citrobacter sp., Escherichia coli, Enterobacter

TABLE 3: CULTURAL, MORPHOLOGICAL AND BIOCHEMICAL CHARACTERISTICS

	1	2	3	4	5	6	7	8	9	10	11
Shape	Circular	Circular	Circular	Circular	Circular	Circular	Circular	Circular	Irregular	Circular	Irregular
Elevation	Convex	Convex	Convex	Convex	Convex	Convex	Convex	Convex	Flat	Low convex	Flat
Margin	Entire	Entire	Entire	Entire	Entire	Entire	Entire	Entire	Undulate	Entire	Serrated
Wetness/dryness	Wet	Wet	Wet	Wet	Wet	Wet	Wet	Wet	Dry	Wet	Wet
Transparency	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque
Colour	Cream-white	Cream	Cream	Cream	Yellow	Cream	Whitish	Dull cream	Cream	Green	Cream
Size	Medium	Small	Small	Medium	Small	Medium	Small	Medium	Large	Medium	Large
Gram staining	+	-	-	-	+	-	+	+	+	-	-
Cell type	Cocci	Rod	Rod	Rod	Cocci	Rod	Cocci	Rod	Rod	Rod	Rod
Cell arrangement	Cluster	Single	Single	Single	Cluster	Single	Chain	Single	Chain	Single	Single
Catalase	+	+	+	+	+	+	-	+	+	+	+
Oxidase	-	-	-	-	-	-	-	-	-	+	-
Coagulase	-	-	-	-	+	-	-	-	-	-	-
Urease	+	-	-	-	+	+	+	+	+	-	+
Indole	-	-	+	-	-	-	-	-	-	-	+
Citrate	+	+	-	+	+	+	+	+	+	+	+
Glucose	A	AG	AG	AG	A	AG	A	A	A	A	A
Lactose	-	+	+	+	-	+	-	-	-	-	-
Possible isolates	Staphylococcus Epidermidis	Citrobacter sp.	Escherichia Coli	Enterobacter sp.	Streptococcus Aureus	Klebsiella sp.	Staphylococcus Pyogenes	Corynebacterium sp.	Bacillus Subtilis	Pseudomonas aeruginosa	Proteus vulgaris

sp., Staphylococcus aureus, Klebsiella sp., Streptococcus pyogenes, Corynebacterium sp., Bacillus subtilis, Pseudomonas aeruginosa and Proteus vulgaris (Table 3), while the fungal isolates were Rhizopus sp., Aspergillus niger, Fusarium sp., Penicillium sp. and Mucor sp (Table 4). Nine of the ten bacterial isolates (Staphylococcus epidermidis, Escherichia coli, Enterobacter sp., Staphylococcus aureus, Klebsiella sp., Streptococcus pyogenes, Corynebacterium sp., Bacillus subtilis, Pseudomonas aeruginosa and Proteus vulgaris) were identified to show significant multi-drug resistance (Tables 5 and 6). The most predominant bacterial isolate was Staphylococcus aureus (25.22%), while Citrobacter sp. (1.74%) was the least (Table 7). The plasmid profile of the isolates revealed detectable plasmid fragments in all the bacterial isolates (Plate 1

Table 4: Cultural and microscopic characteristics of the fungal isolates

Characteristics	1	2	3	4	5
Cultural	White fluffy	Black fluffy	Cottony	Greenish colony	White flat colony
	colony with reverse side	colony with reverse side	white colony with reverse	with white periphery	with reverse cream
	cream	yellow	side cream		
Microscopic					
Nature of hyphae	Non-septate	Septate	Septate	Septate	Non-septate
Colour of spore	Cream	Brown	Cream	Green	Cream
Type of spore	Sporangiospore	Conidiospore	Conidiospore	Conidiospore	Conidiospore
Appearance of special structure	Rhizoid	Foot cells	Micro and macro	Brush-like conidi	Sporangium
Possible Isolates	Rhizopus sp.	Aspergillus	Fusarium sp.	Penicillium sp.	Mucor sp.

TABLE 5: ZONE OF INHIBITION (MM) FOR ANTIBIOTIC SENSITIVITY TEST BEFORE CURING

	PEF	CN	APX	Z	AM	R	CPX	S	SXT	E
Gram +ve										
Staphylococcus epidermidis	R	I	R	S	R	S	S	R	R	I
Staphylococcus aureus	R	R	R	I	R	R	S	R	R	I
Streptococcus pyogenes	R	R	R	S	R	I	S	R	R	S
Corynebacterium sp.	R	I	R	R	R	R	S	I	R	I
Gram -ve	SXT	CH	SP	CPX	AM	AU	CN	PEF	OF	XS
Citrobacter sp.	S	S	I	S	R	R	S	S	S	I
Escherichia coli	R	R	R	S	R	R	S	R	S	R
Enterobacter sp	R	R	I	S	R	R	S	R	S	R
Klebsiella sp.	R	R	R	S	R	R	S	R	S	R
Pseudomonas aeruginosa	R	R	R	I	R	R	I	R	S	R
Proteus vulgaris	R	R	S	S	R	R	S	R	S	R

KEY: SXT= Septrin (30µg), CH= Chloranphenicol(30µg), SP= Sparfloxacin (10µg), CPX= Ciprofloxacin(10µg), AM= Amoxicilin (30µg), CN= Gentamycin (10µg), PEF= Pefloxacin (10µg), OFX= Ofloxacin (10µg), S= Streptomycin (30µg), E= Erthromycin (10µg), APX= Ampiclox (30µg), R= Rocephin (25µg), Z= Zinaceff (20µg).

TABLE 6: ZONE OF INHIBITION (MM) FOR ANTIBIOTIC SENSITIVITY TEST AFTER CURING

Gram +ve	P E F	C N	A P X	Z	A M	R	C P X	S	S X T	E
Staphylococcus epidermidis	I	S	R	S	I	S	S	I	R	I
Staphylococcus aureus	I	R	I	I	R	S	S	R	I	S
Streptococcus pyogenes	R	I	R	S	I	S	S	I	I	S
Corynebacterium sp.	I	S	I	I	I	I	S	I	R	S
Gram -ve	S X T	C H	S P	C P X	A M	A U	C N	P E F	O F X	S
Citrobacter sp.	S	S	S	S	I	I	S	S	S	I
Escherichia coli	I	R	I	S	I	R	S	R	S	R
Enterobacter sp	I	I	I	S	R	I	S	R	S	R
Klebsiella sp.	R	I	R	S	I	R	S	R	S	I
Pseudomonas aeruginosa	R	I	R	S	R	R	S	R	S	R
Proteus vulgaris	I	I	S	S	I	R	S	I	S	I

KEY: SXT= Septrin (30µg), CH= Chloranphenicol (30µg), SP= Sparfloxacin (10µg), CPX= Ciprofloxacin(10µg), AM= Amoxicilin (30µg), CN= Gentamycin (10µg), PEF= Pefloxacin (10µg), OFX= Ofloxacin (10µg), S= Streptomycin (30µg), E= Erthromycin (10µg), APX= Ampiclox (30µg), R= Rocephin (25µg), Z= Zinaceff(20µg)

TABLE 7: FREQUENCY OF OCCURRENCE OF THE BACTERIAL ISOLATES

Isolates	No of isolates	Frequency (%)
Staphylococcus epidermidis	13	11.3
Staphylococcus aureus	29	25.22
Streptococcus pyogenes	8	6.96
Corynebacterium sp.	5	4.35
Citrobacter sp.	2	1.74
Escherichia coli	16	13.91
Enterobacter sp	10	8.7
Klebsiella sp.	7	6.09
Pseudomonas aeruginosa	17	14.78
Proteus vulgaris	8	6.96

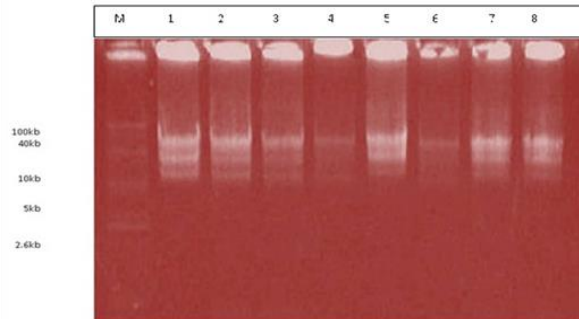


Plate 1: Agarose gel electrophoresis of plasmids harbored by the bacterial isolates. Lane 1: *Staphylococcus epidermidis*, Lane 2: *Citrobacter* sp., Lane 3: *Escherichia coli*, Lane 4: *Enterobacter* sp., Lane 5: *Staphylococcus aureus*, Lane 6:

DISCUSSION

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 various offices at the time of sampling (Tables 1 and 2). This result is 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. There was no significant difference between bacterial counts in the different months ($P > 0.05$) (Table 1) and there was no significant difference between fungal counts in the different months ($P > 0.05$) (Table 2). 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). The high bacterial count in some months, specifically during examination period in the months of March, 2017 and June, 2017 could as well be attributed to increased activities of students visiting the various offices, 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. The microbial

isolates characterized in this research (Tables 3 and 4) have been very much reported, according to Monarca et al., 2000; Otter and French, 2009; Bright et al., 2010, and these fomites in turn serve as vehicles for cross-infections and recontamination of washed hands. 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 poorhygienic conditions in the various Departments. It could also be attributed to the unhygienic handling of the facilities by lecturers and 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 pathogenic and non-pathogenic microbiology. The results of the pre and post-curing susceptibility test of the isolates revealed that some of the bacterial isolates were discovered to show multi-drug resistance to the antibiotics used (Tables 5 and 6). This result is in agreement with those of Singh et al., 2004. Plasmid profile revealed that all the bacterial isolates possessed resistance plasmids (Plate 1). It has been suggested that the development of resistant population of microorganisms can result from gene transfer (Pelczar and Reid, 1998). Plasmids are the major mechanism for the spread of antibiotic resistant genes in bacterial populations. It was revealed that all the isolates subjected to plasmid profiling, had plasmids. 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 were plasmid-mediated in these isolates. Drug resistance and could be attributed to the misuse and overuse of antibiotics in humans and animals as well as the possession of drug resistance plasmids (Madhavan and Sowmiyan, 2011).

CONCLUSION

The results of this study showed that there was high level of bacterial and fungal contamination on the door surfaces of the ten (10) different sampling sites in University of Benin staff offices.

The presence of bacterial and fungal isolates on the door surfaces is of major health concern for students, staff and visitors of the University, because of their potential to cause disease outbreak.

Therefore, everyone is encouraged to adopt excellent personal hygiene practices such as the

use of hand-sanitizers, hand-washing before and after the use of door handles and routine surface disinfection of door handles in order to prevent cross-contamination.

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REFERENCES

Aladenika SST and Matthew FO (2014). Indoor airborne microflora in various section of a tertiary healthcare centre in rural area of ovia northeast, Edo State. *Am J Infect Dis Microbiol* 2(4):86-90.

Bannett HL and Hunter BB (1972). *Illustrated genera of Imperfect Fungi*, Burgess Publishing Company, Minneapolis, Minnesota. Pp: 241.

Bauer AW, Kirby WM, Sherris JC and Jurck M (1966). Antibiotic susceptibility testing by a

standard single disc method. *Am J Clin Pathol* 45(1): 493-496.

Boone SA and Gerba CP (2010). The Prevalence of human parainfluenza virus on indoor office formite. *Food Environ Virol* 2(1): 41-46.

Bright KR, Boone SA and Gerba CP (2010). Occurrence of bacteria and viruses on Elementary classroom surfaces and the potential

role of Elementary Classroom hygiene in the spread of infectious diseases. *The J Sch Nursing* 26(1): 33-41

Chesebrough M (2000). *Medical Laboratory Manual for Tropical Countries Volume 11*. Second edition, University Press, Cambridge, Great Britain, Pp: 377

Clinical and Laboratory Standards Institute (2012). Performance for antimicrobial susceptibility testing. *Twenty- Second Inform Suppl* 32(2): 45-188.

Domsch KH, Gams W and Anderson TH (1980). *Compendium of soil fungi*, Academic press, A subsidiary of Harcourt Brace Jovanovich, publisher. Pp: 383.

Ekhaise FO and Ogboghodo BI (2011). Microbiological indoor and outdoor air quality of two major hospitals in Benin City, Nigeria. *Sierra Leone J Biomed Res* 3(3): 169-174.

Ellis MB (1976). Dermatacious Hyphomycetes, Commonwealth Mycological Institute, Kew, Surrey, UK. Pp: 507 Gilman JC (1944). A manual of soil fungi, Revised 2nd edition, Oxford and IBH publishing Co.

Holt JG, Krieg NR and Sneath PHA (1989). Bergey's Manual of Determinative Bacteriology (Vol. 4). London: Cambridge University Press Pp: 2493.

Kado CL and Liu ST (1981). Rapid procedure for detection and isolation of large and small plasmids. *J Bacteriol* 145: 1365-1373

Kennedy DI, Enriquez CE and Gerba CP (2005). Enteric bacterial contamination of public restrooms. *Cleaning Ind Res Inst*, 1: 12-20.

Li S, Eiseberg JNS, Sicknall IH and Koopman JS (2009). Dynamics and control of infections transmitted from person to person through the environment. *Am J Epidemiol* 170(2): 257-265.

Madhavan HN and Sowmiyan M. (2011). Mechanism of development of antibiotic resistance in bacteria among clinical specimens. *J clin Biomed Sci* 1(2): 42-48.

Monarca S, Grottole, M., Renzi, D, Paganelli, PS, Zerbini, I and Nardi G (2000). Evaluation of

environmental bacterial contamination and Procedures to control cross infection in a sample of Italian Dental Surgeries. *Occup Environ Med* 57: 721-726.

Nwankwo, E. O. and Afuruobi, H. C. (2015). Isolation and Identification of Bacterial Contaminants from Door Handles in a tertiary Institution in Umuahia, Abia State, Nigeria.

Nigerian Journal of Microbiology, 29, pp.3139-3147

Ogbeibu AE (2015). *Biostatistic: a practical approach to research and data handling*, 2nd ed.,

Mindex Publishing Company, Lagos, Nigeria. pp. 17-22.

Oranusi S, Dahunsi SO, Owoso OO and Olatile TN (2013). Microbial profiles of hands, foods, easy contact surfaces and food contact surfaces: A case study of a University campus. *Int J Biotech Biosci* 2(1): 30-38.

Osterholm MT, Hederg CW and MacDonald KL (1995). *Epidemiology of Infectious Diseases. Bennett's principles and Practice of Infectious Diseases*, vol. 1, 4th ed., Churchill-Livingstone, New York. Pp: 165

Otter J and French G (2009). Bacterial contamination in touch surfaces in the public transport system and in public areas of a hospital in London. *Lett Appl Microbiol* 49: 803-805.

Pelczar MJ, and Reid RD (1998). Activities of antimicrobial agents in Microbiology. *Antibiot Drugs* 87(6): 74-83.

Prescott LM, Harvey JP and Klein DA (1993). *Microbiology*, 2nd ed. W.M.C. Brown, England. Pp: 805.

Sijhary TJ, Bermann ML and Enquist LW (1984). *Experiments with Gene Fusions*. Cold Spring Harbour Laboratory Press, New York. Pp: 55.

Singh K, Brajesh K, Walker A, Alum J, Morgan W and Wright DJ (2004). Biodegradation of chloropyrifos by *Enterobacter* strain B-14 and its use in biodegradation of contaminated soils. *Applied Environmental Microbiology* 70: 4855 – 4863. Sleight DJ and Timbury MC. (1998). Notes on Medical Microbiology, 5th ed., ChurchillLivingstone, New York. Pp: 173