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A Survey of microbial contamination of door handles in various locations in Lokoja metropolis, Kogi state, Nigeria

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Abstract

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. The use of Door handles/knobs daily is inevitable, and it is used by many people. Door handles of selected places in Lokoja Metropolis were investigated for bacterial contamination. The door handles were swabbed and cultured on Blood agar and MacConkey agar, and then various biochemical tests were carried out for confirmation of the isolates. Out of the 170 swab samples cultured, 130/170 (76.5%) were positive and 40/170 (23.5%) were negative, of the Bacteria growth, 50.2% were gram positive and 49.8% were gram negative. The study also found that toilet door handles had highest rate of contamination and the door handles of the reception and Medical laboratory had the least contamination rate. The isolated bacterial contaminants were *Staphylococcus aureus* (29.6%), *Escherichia coli* (25.5%), *Pseudomonas aeruginosa* (24.3%), and Coagulase negative Staphylococcus (20.6%). This shows that the door handles harbors highly pathogenic bacteria which have the potentials of causing epidemics like in the case of Covid-19. These microorganisms can lead to serious health problems. Therefore, it is necessary to practice good personal hygiene through hand washing and use of hand sanitizer as well as daily washing and cleaning of toilets to reduce the incidence of microbial transmission.

Keywords: Microbial contamination, Door handles/knobs, pathogenic bacteria.

Introduction

Fomites when on constant contact with humans or natural habitats of pathogenic organism constitute a major source of spread of infectious diseases. Micro-organisms are found everywhere bacteria contaminate our body and whole environment. Fortunately many billions of bacteria can be dangerous for our health, causing different disease such as pneumonia or skin infection [1]

The areas where microorganisms stay the most are usually the ones that come in direct contact with human beings. When most people think of places where microorganisms congregate, they think of money, phones, keyboards and door handles. Door handles, in home and in public, are the most commonly touched surfaces, and bacteria can easily be transferred from person to person this way [2]. The use of door handles each day is inevitable with little knowledge about the usefulness of these handles in the transmission of diseases. Fomites when on constant contact with humans or natural habitats of pathogenic organism constitute a major source of spread of infectious diseases [3] The fomites include door handle of conveniences, hand lockers, showers, and toilet, including those found in public offices, hospitals, hotels, restaurants and restrooms. [4]

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 such as door handles. Microorganisms constitute a major part of every ecosystem. In these environments, they live either freely or as parasites. 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 [5] Human hands usually harbor microorganisms both as part of body normal flora as well as transient microbes contacted from the environment [6] In a city like Lokoja, people move around from place to place and make use of doors 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 [7]

Materials and Methods

Door handles, Petri dishes, Swab sticks, Autoclave, Incubator, Microscope, Weighing balance, Bunsen burner , Wire loop, Hand gloves, Slides and Cover slips, Measuring cylinder, Conical flasks, MacConkey agar, Nutrient agar, Required stains: Gram stain; Crystal violet, Lugol's Iodine, Acid acetone, Neutral red.

Collection of samples

A total of 170 samples were collected from 11 door handles type in 5 locations in Lokoja metropolis, the locations included Gadumo, Adankolo Market, Lokongoma Phase 2, Ganaja, and Barracks. The specimens were collected from door handles by means of sterile cotton swabs moistened in sterile nutrient broth. The swab was wiped firmly on the entire surface of the door handle and was soaked with Normal Saline. Each swab was placed in small tube and labeled, before being placed in sterile polythene bag and transported the Laboratory.

Examination of the samples

The swabs were inoculated aseptically on Chocolate agar, and MacConkey agar plates, the inoculum were properly done and the plates were incubated aerobically at 37°C. Bacterial growth was checked after 24-48 hours of incubation. At the end of incubation period, the MacConkey and Chocolate agar were examined for their colonial morphology. Each colony represented a different organism and was picked up for identification and characterization.

Identification of Bacteria

Colonial morphology

The diverse colonies observed in the plate were differentiated based on the morphological feature of the yielded colonies including color, size, shape, elevation and pigmentation

Gram stains

Gram stain is an essential step for the next experimental work to identify Gram positive or negative isolates. The procedure was carried out according to [8] as follows;

The smear was prepared from overnight culture on a clean and dry slide. The smear was left to air dry. Fixation was done by rapidly passing the slide three times through the flame of a Bunsen burner, it was allowed to cool before staining. Crystal violet stain was added to smear for 60 seconds, and rinsed with water. Lugol's iodine was added for 60 seconds and rinsed with water and It was decolorized rapidly (few seconds) with acetone alcohol and rinsed immediately with water.

Finally, the smear was covered with Neutral red stain for 2 minutes and rinsed with water. The back of the slide was then blotted with a cotton wool and was placed in a draining rack for smear to air dry. A drop of Immersion oil was added to the dried smear and it was examined under the light microscope (Carl Zeiss, Germany) by oil lens 100X.

Biochemical Tests

Biochemical tests were carried out for the confirmation of the isolates. Tests carried out are;

Citrate utilization test is important in identification of Entetobacteriaceae. Utilization of citrate by tested bacteria was detected in Simmons citrate medium by the production of alkaline by-products. Bacteria that can use citrate can also extract nitrogen from ammonium salt with production of ammonia. [8]

Indole production, Tryptophan broth (peptone water) was inoculated with the test organism and incubated at 37°C for 18 to 24 hour. At the end of incubation period few drops of Kovac reagent was added, and the development of red color at the interface of the reagent and broth in seconds after adding the reagent is an indicator of presence of indole [8]

Oxidase is used to determine bacteria that produce oxidase enzyme which oxidized the oxidase reagent (tetramethyl-ρ-phenylenediaminedihydrochloride) to give a dark-blue color. The test was performed by filter paper impregnated with the oxidase reagent; a pure colony was smeared on the disc by sterile wooden stick. A positive reaction was indicated by developing deep blue color within 10 seconds [8] Coagulase test was used to identify *Staphylococcus aureus* which produces the enzyme coagulase. Catalase test was used to differentiate Bacteria that produce the enzyme catalase from non-catalase producing bacteria.

Statistical Analysis

The data obtained were analyzed using one way ANOVA and descriptive analysis and presented in form of frequency tables and charts.

Results

Frequency and percentage of growth from door handles in Lokoja

The study revealed that a total of 170 door handle swabs were collected from five locations in Lokoja metropolis in Kogi State which yielded both positive and negative culture outcome. The frequency and percentages of these swabs were presented in table 1 Cultivation of these swabs yielded bacterial growth on 130/170 (76.5%) plates. The rest of 40/170 (23.5%) swabs showed no bacterial growth. Of the bacterial growths, 50.2% were gram-positive bacteria and 49.8% were gram- negative.

Frequency of growth in relation to location

Barracks had the highest rate of contamination with 25.3%, followed by Lokongoma phase 2 with 20% then Adankolo market and Ganaja with 18.5% respectively and the lowest is Gadumo with 17.7%. As seen in Table 2

Bacteria species from door handles in Lokoja metropolis

The study shows that, *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus epidermidis*. Biochemical tests adopted for identification of the Organisms were tabulated in Table 3

The study revealed that colony with gram positive cocci reactions, catalases, coagulase positive, citrate, indole and and oxidase negative were identified as *Staphylococcus aureus*. Colonies with gram positive cocci, catalase positive, coagulase, citrate, indole and oxidase negative were identified as *Staphylococcus epidermidis* Table 3

Also, colony with gram negative rods, catalase, citrate and oxidase positive and coagulase and indole negative were identified as Pseudomonas aeruginosa. Colonies with gram negative rods, indole positive, catalase, coagulase, citrate and oxidase negative were identified as *Escherichiacoli* Table 3

Frequency of contamination of door handles in relation to location

The door handles with the highest contamination are those of the toilet especially toilets that are located in public areas such as hospitals, general toilets and toilets in market places while the door handles in areas with the least contaminated door handle is that of the Reception and the Medical laboratory which is probably as a result of door sterilization and observation of proper and adequate hand washing with medicated soaps.

Frequency of Bacteria isolates on door handles

In this study, *Staphylococcus aureus* is the most frequently occurring organism with 29.6%, followed by *Escherichia coli* 25.5%, then *Pseudomonas aeruginosa* and 24.3% lastly *Staphylococcusepidermidis* 20.6%. As in Table 5

Table 1 Sample collection site, frequency of growth and no growth

Numbers examined	Handles that yielded growth	Handles that yielded no growth
DOOR HANDLES		
Refrigerator	15	12 (80%)	3 (20%)
Shops	15	10 (66.7%)	5 (33.3%)
Cell	5	4 (80%)	1 (20%)
Stations	10	7 (70%)	3 (30%)
Laboratory	15	9 (60%)	6 (40%)
Hospital	15	13 (86.7%)	2 (13.3%)
Toilet	20	19 (95%)	1 (5%)
Offices	15	11 (73.3%)	4 (26.7%)
Reception	15	9 (60%)	6 (40 %)
Kitchen	15	12 (80%)	3 (20%)
Hotel	15	11 (73.3%)	4 (26.7%)
Vehicles	15	13 (86.7%)	2 (13.3%)
Total	170	130 (76.5%)	40 (23.5%)

This table shows the output of the ANOVA analysis and whether there is a statistically significant difference between doors handles that yielded growth and doors handles that yielded no growth. It can be seen that the significant value is 0.010 (i.e., *p-value* = .010), which is below 0.05 and, therefore, there is a statistically significant difference between doors handles that yielded growth and doors handles that yielded no growth.

Table 2: Location of collection of samples and frequency of growth

		LOCATION				
	Number of +ve samples	Gadumo	Lok. Phase 2	Barracks	Adankolo market	Ganaja
DOOR HANDLES						
Refrigerator	12	1	3	3	2	3
Shops	10	x 2	3	1	3	1
Cell	41	-	3	-	-	
Stations	7	2	-	5	-	-
Laboratory	9 -	1	2	2	4	
Hospital	13 3	3	2	3	2	
Toilet	19 4	3	4	4	4	
Office	11 2	2	3	3	1	
Reception	9	1	3	2	2	1
Kitchen	12	1	3	3	2	3
Hotel	11	3	3	2	1	2
Vehicles	13	3	2	3	2	3
Total	130	23(17.7%)	26(20%)	33(25.3%)	24(18.5%)	24(18.5%)

KEY : +ve = positive
- = no growth

Table 3 Identification of the probable organisms

CHARACTERISTICS	1	2	3	4
CELL MORPHOLOGY	Cocci	Cocci	Bacilli	Bacilli
GRAM REACTION	+	+	-	-
CATALASE	+	+	-	+
COAGULASE	-	+	-	-
CITRATE	-	-	-	+
INDOLE	--	+	-	
OXIDASE	--	-	+	
PROBABLE ORGANISM	*S. epidermidis*	*S. aureus*	*E. coli*	*P. aeruginosa*

KEY: + = positive
- = negative

Table 4: Frequencies of contamination of door handles in different areas

Door handles	ORGANISMS			
	S. epidermidis	*S. aureus*	*E. coli*	*P. aeruginosa*
Refrigerator	4 (18.2%)	5 (22.7%)	7 (31.8%)	6 (27.3%)
Shop	5 (31.2%)	4 (25%)	4 (25%)	3 (18.8%)
Cell	2 (20%)	1 (10%)	3 (30%)	4 (40%)
Station	5 (22.7%)	4 (18.2%)	6 (27.3%)	7 (31.8%)
Hospital	8 (30.8%)	6 (23.1%)	5 (19.2%)	7 (26.9%)
Laboratory	4 (30.8%)	2 (15.4%)	4 (30.8%)	3 (23.0%)
Toilet	12 (26.7%)	14 (31.1%)	9 (20%)	10 (22.2%)
Offices	7 (33.3%)	6 (28.6%)	5 (23.8%)	3 (14.3%)
Reception	8 (40%)	2 (10%)	4 (20%)	6 (30%)
Hotel	9 (52.9%)	3 (17.7%)	5 (29.4%)	0
Restaurant	6 (20%)	5 (16.7%)	10 (33.3%)	9 (30%)
Vehicles	9 (36%)	3 (12%)	6 (24%)	7 (28%)

Table 5: Frequency of Bacteria isolates from door handles

Bacteria isolates	Frequency	%
Staphylococcus epidermidis	7929.6	
Staphylococcus aureus	55	20.6
Escherichia coli	68	25.5
Pseudomonas aeruginosa	65	24.3

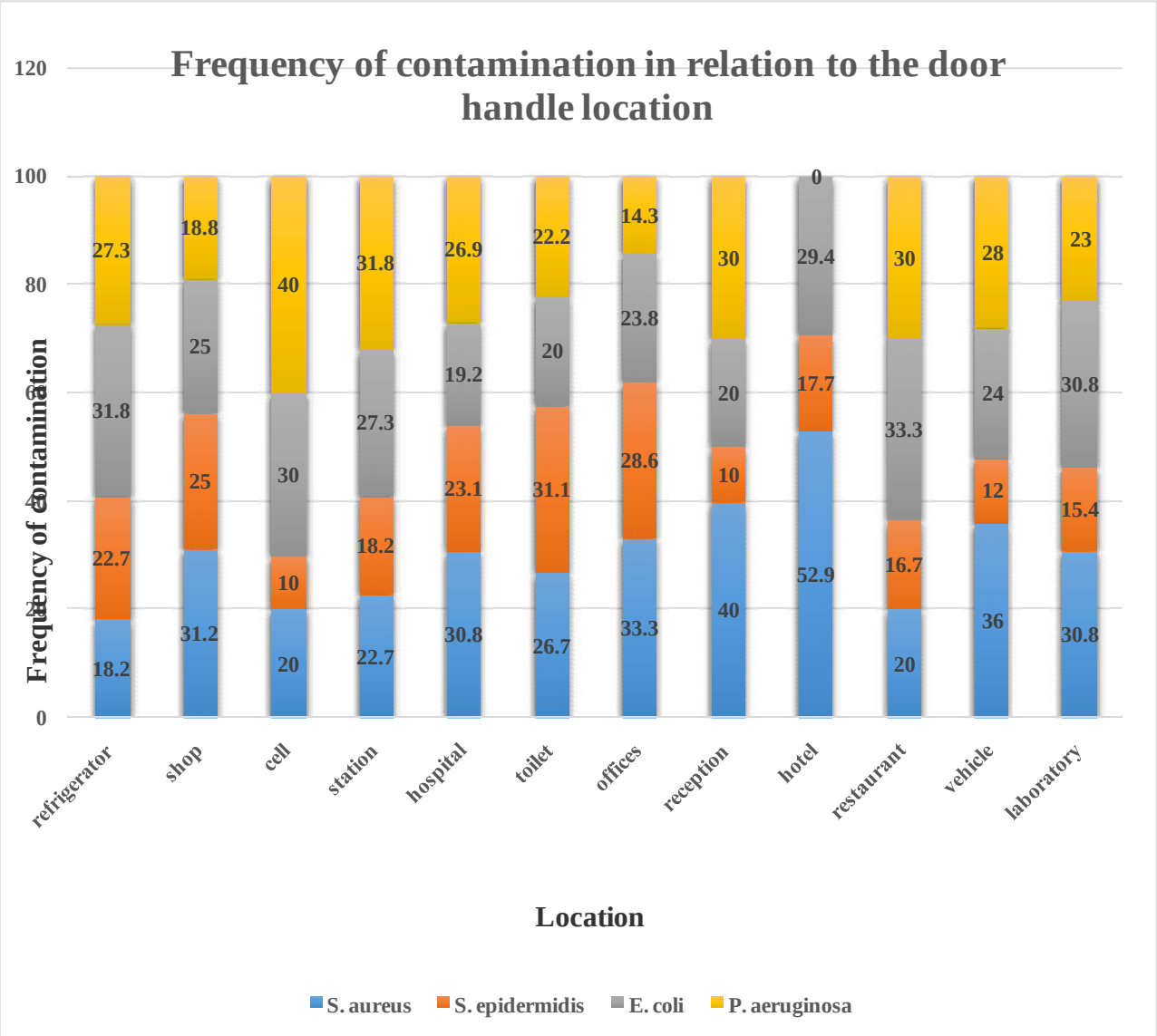

Figure 1 showing the Frequency of contamination in relation to the door handle location.

Discussion

Inanimate environmental surfaces such as door handles can become directly contaminated with microorganisms after frequent exposure to different people. They acquire these pathogens through contact with contaminated surfaces such as of door handles [9] The microbiological analyses of samples from the different door handles in different locations in the study showed that door locations played significant role in the distribution of microorganisms. Generally, it was found that samples analyzed from toilet door handles especially toilet in public areas recorded the highest bacterial load. This could be as a result of high rate of exposure of the door handles to large traffic users who crowd in and out without proper hand hygiene, thereby disseminating their flora to the door handles. Toilet environments usually contain higher microbial loads than other facilities within any public center [7]

Environmental factors such as relatively high humidity and moisture content can play crucial role in influencing microbial transfer rates on fomites or hands. It was found that greater microbial carriage and dissemination occurred typically at relative humidity. Also previous studies have shown that moist fomites or hands influence microbial transfer more than dry hands or surfaces [9].

Hence, the relatively high microbial load of the bacterial pathogens from the samples collected from the Toilet door handles could have been facilitated by the relatively high humid nature of the toilet environment that probably encourages microbial replication and development. It also suggests poor hand hygiene practices after making use of the toilet as well as lack of adequate cleaning and sanitation of facilities [10] This study also revealed a significant difference in the levels of contamination levels of door handles of different doors at locations between Shops, Refrigerators, Cell, Stations, Laboratory, Hospital, Toilet, Offices, Receptions, Kitchens, Hotel and Vehicles. A high level of contamination was also observed at door handles of Hospitals which is an indication of high inflow and outflow of patients and health care-givers that normally use the routes for routine work. It has been reported that several nosocomial pathogens were shed by patients and health-care workers during interactions as well as during uncontrolled movement of visitors in and out of different sections in the hospital. Another door handle with a high level of contamination is the handles of vehicles which were mostly public vehicles which are as a result of its usage by many people daily. However, relatively lower rate of microbial contaminations was observed from handles of Receptions and the Medical laboratory. This could be attributed to a relatively higher level of compliance to environmental and hand hygiene practices which encompasses hand washing with antimicrobial soap and clean water, as well as appropriate use of gloves in the Laboratory and the use of a Door men at the doors of the receptions which reduces the amount of people that makes direct contact with the door handles. A similar submission was made by an earlier reporter, who recommended strict personal and environmental sanitation within and around hospital wards to prevent horizontal dissemination of pathogens [11]

In this study, it was observed that *Staphylococcus aureus* (29.6%) had the mean highest frequency of occurrence on the door handles, and this was followed by Escherichia coli (25.5%) *Pseudomonas aeruginosa* (24.3%), and *Staphylococcus epidermisis* (20.6%), are the least. The finding of *Staphylococcus aureus* as the most frequent bacterial contaminant in the study is in accordance with previous works in Nigeria that reported *Staphylococcus aureus* as the most prevalent contaminants of door handles. In their study, [12] reported *Staphylococcus aureus* 33(25.0%) as the most frequently isolated bacteria from door handles of a tertiary Institution at Umuahia, Abia State, Nigeria.

Another on The Role of Door Handles in the Spread of Microorganisms of Public Health Consequences in University of Benin Teaching Hospital (UBTH), Benin City, Edo State, isolates they reported, in decreasing order of frequencies were coagulase negative Staphylococcus (CoNS), 28(21.2%), Streptococcus species 22(16.6%), Bacillus species 22(16.6%), *Enterococcus feacalis* 6(4.8%), Klebsiella species 3(2.2%), *Escherichia coli* 4(3.0%), *Proteus mirabilis* 4(3.0%), *Proteus vulgaris* 6(4.6%), and *Pseudomonas aeruginosa* 2(1.5%) being the least. Similarly, Staphylococcus species (43.3%) were reported as the most prevalent bacterial contaminants of doors handles of Secondary Schools in Bokkos Local Government of Jos Plateau State, Nigeria [13]
The other microbial contaminants reported, in decreasing order of prevalence, were Candida species (10%), *Escherichia coli* (16.7%), Citrobacter species (1.7%), Klebsiella species (20%), Proteus species (6.7%) and Salmonella species (1.7%). In addition, Staphylococcus species were also reported as the most frequent contaminants of door handles of public conveniences, hospital equipment and surfaces [14]
Escherichia coli being the highest gram negative organism in this study is of great public health concern. Apart from the fact *Escherichia coli* are the major indicators of fecal contamination and poor hand hygiene; they possess diverse strains with potent virulence and toxic factors. They are mainly responsible for urinary, gastrointestinal and urogenital ailments of humans [15] Therefore, *Escherichia coli* could be transferred to people who come in contact with such door handles without strict personal hygiene. Also, the significance of Staphylococcus species contamination observed in this study is of lower risk, because they were reportedly carried by 30-50% of healthy humans as normal flora and one of the frequently implicated bacteria in hospital-acquired infections [16]

Staphylococcus species are also known to cause diverse human ailments ranging from minor skin infection such as pimples and boil to a number of life-threatening diseases like bacteremia and sepsis, toxic shock syndrome (TSS), pneumonia, meningitis, osteomyelitis, endocarditis [17]

The presence of *Pseudomonas aeruginosa*, isolates is also of public health significance. *Pseudomonas aeruginosa* has been found to be a major opportunistic pathogen and reportedly identified as one of the vital causes of infection-related mortality among seriously ill and immunocompromised people [18]

These pathogens were found to be the leading causes of wound infection and diarrhea especially in developing countries. They are best described as classic opportunistic nosocomial pathogens which can cause a wide spectrum of infection and morbidity in immune compromised patient. It is therefore surprising that inanimate surfaces, such as door handles in unexpected place like the kitchen, refrigerators, homes and likewise the Hospitals where patients seek medical attention have an important influence on the risk of acquiring infection that may further complicates their health conditions.

The presence of these bacterial isolates from door handles used on daily basis by different people is worrisome, in the light of the fact that some of the isolated bacteria have been reported to demonstrate multi-drug resistance to currently available chemotherapeutic agents [19]

Conclusion

This study has revealed that door handles of different places even the most unexpected places are contaminated by a variety of pathogenic and non-pathogenic microorganisms. This most frequent bacterial isolates was *Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa and Staphylococcus epidermidis* being the least. Hence, door handle surfaces within could therefore act as potential fomites for communicable diseases dissemination. People are encouraged to pay strict attention to personal hygiene practices to avoid the incidence and spread of infections through door handles

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