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Microbial Profile and Antibiotic Susceptibility Patterns of Microbes Isolated from Wash Hand Basins in Covenant University

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ABSTRACT

Although, wash-hand basins aid in the disposal of washouts from contaminated hands, unsanitised basins can be a source of cross-contamination. The aim of this study was to analyse the microbial profiles and antibiotic susceptibility patterns of isolates from wash-hand basins in selected areas in Covenant University. A total of 70 swab samples were collected from wash-hand basins in the laboratories, cafeteria, and lavatories around the hostels and classroom areas. The isolates were identified based on their colonial, microscopic, and biochemical characteristics. The antibiotic susceptibility of selected isolates was determined using the Kirby-Bauer disk diffusion method. A total of seven bacteria and nine fungal isolates were obtained from the swab samples. The most occurring bacteria and fungi were *Staphylococcus aureus* (100%) and *Aspergillus sp.* (88%), respectively. *Escherichia coli* and *Salmonella sp.* were isolated from the wash-basins in the lavatories in the students' hostel. A 100% sensitivity to amoxicillin and amoxicillin-clavulanic acid was observed in the enteric bacteria. Pathogenic *Staphylococcus sp.* was sensitive to Amoxicillin-clavulanic acid, cefuroxime, and cloxacillin. Wash-hand basins could serve as a reservoir for the potential transmission of antibiotic-resistant organisms and may be of public health concern.

Keywords: Wash-basin, Hand-hygiene, Antibiotic-resistance, Bacteria, Fungi.

Introduction

Globally, there is a lot of attention to hand washing as a significant method of preventing several infections and diseases such as diarrhoea and respiratory diseases. Frequent hand washing is also one of the approved control measures of the current COVID-19 pandemic.[1] Furthermore, there is a growing awareness of the role of hand washing not only as a preventive measure but also as a means of reducing the burden of these diseases by eliminating the pathogens that are responsible for their etiologies.[2]

Many washroom users take special care to wash their hands properly. Still, little attention is paid to the potential risks of re-contamination of hands from contaminated surfaces of wash-hand basins. Contaminated surfaces of wash-hand basins may also serve as reservoirs for the acquisition and transmission of bacterial, fungal and viral pathogens to the nose, eyes or mouth of washroom users via indirect contact.[3,4] These pathogens may survive on surfaces for extended periods. Wash-hand basins may also be potential sites for the transmission of pathogenic organisms, especially antibiotic-resistant bacteria.[5,6] The aim of this study was to analyse the microbial profiles and antibiotic susceptibility patterns of isolates from wash-hand basins in selected areas in Covenant University. Given the high population density and the rate of daily use of these wash hand basins, the findings from this study may have substantial public health implications.

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Materials and Methods

Sample collection

This research study was carried out at Covenant University, Ota, Ogun State, Nigeria. It comprises of fully operative campus and lecturing facilities which also includes laboratories. With the use of sterile moistened swab sticks, triplicate samples were obtained from 5 cm^2 of the wash-hand basins in the offices and laboratories in the College of Science and Technology, College of Engineering and College of Leadership and Development Studies, as well as in the cafeteria and the lavatories in the student hostels. The swab samples were conveyed aseptically in a sterile container to the laboratory for further analysis.

Sample preparation

The swab samples collected from 5 cm^2 of each wash-hand basin were rinsed out into test tubes with 5 mL of sterile peptone water.[7] A 1ml rinse-out from each swab sample was dispensed into the appropriately labelled sterile disposable Petri dishes for microbiological assessment.

Microbiological assessment of samples

Pour plate method was carried out with Nutrient Agar, MacConkey Agar, Mannitol Salt Agar, Salmonella-Shigella Agar and Potato Dextrose Agar for 1 mL of each sample. Screening and selection were based on morphology, microscopy (Gram staining) and cultural characteristics. The distinct colonies were stored on agar slants at 4°C for further identification using biochemical tests.[8]

Identification of fungal isolates

The identification and classification of fungal isolates were carried out according to methods as previously described.[9,10]

Antimicrobial susceptibility testing

The Kirby-Bauer method was used for the disk diffusion test, and results were interpreted according to the Clinical and Laboratory Standards Institute.[11]

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Results and Discussion

A total of seven fungi and eight bacteria were isolated from the wash-hand basins in lavatories, offices, cafeteria, and laboratories in the study site. Predominant fungal species were *Aspergillus niger, A. flavus, Neurospora sp., Fusarium spp, Penicillium* and yeast, with *Aspergillus* being the most frequently isolated (Table 1). Many of the fungi isolated are ubiquitous in the environment and are commonly isolated (Table 1).

The isolated bacteria were identified as *Staphylococcus aureus, S. epidermidis, Bacillus sp., Pseudomonas spp. Escherichia coli, Salmonella spp., Micrococcus spp,* and *Proteus sp.* (Table 2). These microorganisms were also isolated and reported in similar studies.[12–14] Although *Staphylococcus spp.* is a normal flora of the skin and organisms such as *Bacillus spp. and Pseudomonas spp.* are ubiquitous and frequently isolated in the environment, others such as *P. mirabilis and E. coli* are pathogenic and are commonly associated with urinary tract infections.[15] *Staphylococcus spp* was the most frequently occurring organism isolated at all the sample sites (Table 3).

The incidence of *S. aureus* and *S. epidermidis* is not surprising as they are normal flora of the skin and their presence may be attributed to the normal shedding of the skin. On the other hand, these bacteria can carry multiple antibiotic-resistance genes which can be a source for community transmission. A study by Mkrtchyan et al. revealed that more than one-third of *Staphylococcus spp*. isolated from toilets in public buildings in the United Kingdom carried multiple antibiotic resistance determinants.[16] Strains of community-associated Methicillin-Resistant *Staphylococcus aureus* (MRSA) have also been increasingly implicated in skin and soft tissue infections nationwide in the United States and pose a significant public health challenge.[17-18] *Staphylococcus spp*. isolates were resistant to Gentamicin, Ofloxacin and Erythromycin (Table 4), which are first-line antimicrobial agents, and the observed antibiotics resistance rate was 37.5%, similar with results from a previous study on the antibiotic resistance patterns of isolates from public washrooms.[13] Similar antibiotic resistance rate of 37.8% was also observed in *Staphylococcus spp*. isolates, as reported by Mkrtchyan *et al*.[16]

Table 1: Fungal contamination of the wash-hand basins at the different locations sampled

Organism	Lavatories N = 13	Offices N = 7	Cafeteria N = 3	Laboratories N = 9	Total	Total %
Aspergillus spp.	12	5	2	8	27	88
Geotrichum spp.	5	1	1	8	15	47
Yeast	4	1	1	1	7	22
Penicillium spp.	1	0	2	4	7	22
Fusarium spp.	4	1	1	1	7	22
Alternaria spp.	3	0	2	1	6	19
Neurospora spp.	3	0	0	1	4	13
Cladosporium spp.	1	0	0	0	1	3

N = total number of sample sites per location

Table 2: Biochemical characteristics of bacterial isolates

	Isolates							
Tests	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
Colour	Grey	White	Grey-white	Green brown	Grey-white	Green with black centre	Yellow	Pink
Shape	Cocci	Cocci	Rods	Rods	Rods	Rods	Cocci	Rods
Gram stain	+	+	+	-	-	-	+	-
H_2S Gas	-	-	-	-	-	+	-	-
Indole	-	-[a]	-	-	+	-	-[a]	-
Motility	-	-	+	+	-[a]	-	-	+
Oxidase	-	-	-	+	-[a]	+	+	-
Catalase	+	+	+	+	+	NT	+	+
Coagulase	+	-	NT	-	-[a]	NT	-	NT
MR	+	-	-	-	+	+	-	+
VP	+	+	+	-	-	-	+	-
Citrate	+	-	+	+	-	-	+	+
Lactose	+	+	-	-	+	-	+	-
Maltose	+	+	+	-	-[a]	+	-[a]	-
Glucose	+	+	+	-	-[a]	+	+	+
Most probable organism	*S. aureus*	*S. epidermidis*	*Bacillus sp.*	*Pseudomonas sp.*	*Escherichia coli*	*Salmonella sp.*	*Micrococcus sp.*	*Proteus sp.*

NT = Not Tested

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Table 3: Bacteria isolated from the wash-hand basins at the different locations sampled

Organism	Lavatories N = 13	Offices N = 7	Cafeteria N = 3	Laboratories N = 9	Total	Total %
Staphylococcus spp.	13	7	3	9	32	100
Escherichia coli	13	6	0	5	24	75
Micrococcus spp.	9	3	1	4	17	53
Bacillus cereus	3	3	2	3	11	34
Pseudomonas spp.	3	0	1	0	4	12.5
Proteus sp.	2	0	0	2	4	12.5
Bacillus spp.	2	1	0	1	4	12.5
Salmonella spp.	2	0	0	0	2	6

N = total number of sample sites per location

Table 4: Antibiogram of bacterial isolates

Organism	Number of resistant Antibiotic	% resistance N = 8
Staphylococcus aureus	GEN, OFL, ERY (3)	37.5
Staphylococcus epidermidis	GEN, OFL, ERY (3)	37.5
Bacillus cereus	GEN, OFL, ERY (3)	37.5
Micrococcus sp.	GEN, OFL, ERY (3)	37.5
Pseudomonas sp.	NIT, GEN, OFL (3)	37.5
Escherichia coli	NIT, GEN, NAL, OFL, TET (5)	62.5
Salmonella	COT, NIT, GEN, NAL, OFL, TET (6)	75
Proteus mirabilis	COT, NIT, GEN, NAL, OFL, TET (6)	75

Key: Trimethoprim-sulfamethoxazole – COT, Nitrofurantoin – NIT, Gentamicin – GEN, Nalidixic acid– NAL, Ofloxacin – OFL, Tetracycline – TET, Erythromycin – ERY, N – total number of antibiotics tested

Our results also revealed that pathogenic organisms such as *Salmonella sp., Escherichia coli* and *Proteus mirabilis* (Table 2) were isolated from the sampled wash-hand basins. These organisms are usually associated with faecal contamination and/or urinary tract infection. The presence of these organisms also highlights a potential risk of disease, which can arise from direct contact with contaminated wash hand surfaces or by person-to-person contact or via the faecal-oral route. These organisms also had the highest rate of antibiotic resistance, with *Salmonella sp.,* and *Proteus mirabilis* showing resistance to 75% of the tested antibiotics, and *E. coli* to 62.5% (Table 4). A 100% sensitivity to Amoxicillin and Amoxicillin-clavulanic acid, however, was observed in these enteric bacteria (Figure 1).

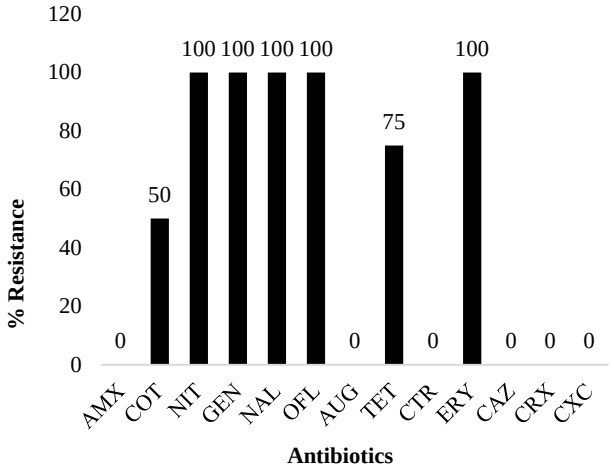

Figure 1: Resistance patterns of the isolates to the tested antibiotics. Trimethoprim-sulfamethoxazole – COT, Nitrofurantoin – NIT, Gentamicin – GEN, Nalidixic – NAL, Ofloxacin – OFL, Tetracycline – TET, Erythromycin – ERY.

Conclusion

This research highlights the need for good hygiene practices during the use of wash-hand basins in public places, as these facilities may be a reservoir for pathogens. Regular cleaning and disinfection of shared wash-basins are also recommended to prevent community spread of pathogens.

Conflict of interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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