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Research Article

Isolation and Identification of Fungi and Bacteria Associated With Contamination of Tap and Door Handles In Faculty of Allied Health Sciences, Bayero University, Kano-Nigeria

H. Sule and A. I. Abdulkadir

¹Department of Medical Laboratory Science, Faculty of Allied Health Sciences, Bayero University, Kano-Nigeria

*Corresponding author's Email: sule.hamza@yahoo.com, doi.org/10.55639/607.02010064

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ABSTRACT

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The research aimed to isolate and identify possible fungal and bacterial contaminants associated with these surfaces in Faculty of Allied Health Sciences, Bayero University, Kano. In the study protocol, tap and door handles surfaces were swabbed using sterile swab stick and inoculated onto Potatose Dextrose Argar (PDA), Nutrient gar and chocolate agar and incubated at 37°C for yeast and bacteria and 28 °C for moulds. Positivity rate of the different sample sources showed that tap handles (for toilet had 17.5%, laboratory had 16.7%) while for the door handles (that of toilet had 21.1% while offices had only 7% as the least); also for the door handles, interior had 57.3% while exterior of the door handles had 42.7%. With regards to the isolates, bacteria had 74.7% while fungi had 25.3%. Distribution of the isolate shows: *Candida* spp had 30.4%, *A. fumigatus* 21.7%, *A. flavus* 10.9%, *A. niger* 6.5%, *Penicillium* 21.7%, *Rhizopus* spp 6.5% and *Mucors* spp had 2.2% isolation rate. We can therefore conclude that tap and door handles harbours a variety of organisms including fungal and bacterial species.

Corresponding author: H. Sule, Email: sule.hamza@yahoo.com

Department of Medical Laboratory Science, Faculty of Allied Health Sciences, Bayero University, Kano-Nigeria

INTRODUCTION

Door handles particularly of public offices and restrooms where many people utilise, serves as vehicle that potentially transfer pathogens to one another. Although people spread infections through everyday interactions, but fomites play a major role in that respect, especially in community-acquired infections (Akinjogula, and Divine 2017). Normally, faecal matters acts as source of human pathogens, and usually leads to outbreaks of infection in the event of unhygienic utilization of toilet facilities which is of great public health concern (Akinjogula, and Divine 2017). Some of the risks factors of fomites lead transmission include the frequency of contamination of the surface or object with infectious agents; the number of infectious agent involved; the virulence of the pathogen and the susceptibility of the host exposed to it and good personal hygiene status (Nguyen *et al.*, 2007).

When in contact with surfaces like tables, furniture or even computer keyboards, people can easily pick up pathogens from those surfaces, as it also happens with door handles and that of toilets and office facilities, which are frequently been touched in public spaces. They serve as inanimate sources of infection, if they are previously contaminated with infectious agents by any means (Amala *et al.*, 2015). For facilities toilets, offices and bathrooms, their door handles remain among the cheap sources of infections due to contamination (Kawo *et al.*, 2012; Opere *et al.*, 2013).

Naturally, our hands are the primarily used to interact with and also help us to manipulate the physical world around our environments. The hands carries a variety of micro-organisms, pathogenic or non-pathogenic ones; bacteria or fungi and can spread to other people and surfaces with ease, potentially leading to diseases (Reynolds and Hurst, 2008). In northern parts of Nigeria, Kano State inclusive, many if not most, people care less about what surfaces carries in terms of pathogens, therefore mingles with them anyhow. More so, there are few researches that has to do with some of these surfaces especially the taps and door handles.

MATERIALS AND METHODS

Study Area

This study was conducted at Faculty of Allied Health Sciences, Bayero University, Kano.

The Faculty is one of the four faculties that make up the College of Health Sciences, Bayero University Kano, located at Aminu Kano Teaching Hospital (AKTH) along Zaria road, Kano State, Nigeria. The Faculty currently has five departments offering both academic and professional training in various health care specialties. The Departments include; Medical Laboratory Science, Physiotherapy, Nursing Sciences, Medical Radiography and Optometry.

Study Population/Objects

The study targeted regularly contacted tap and door handles of classrooms, laboratories, library, offices and toilets in different departments of the Faculty of Allied Health Sciences. Stratified sampling technique was used to divide the population of tap and door handles into subgroups or strata within the faculty.

Sample Size Determination

The sample size was calculated and determined using the formula by Naing *et al.* (2006) which is as follows: $n = z^2 p (1 - p) / d^2$

Where;

n =Sample size

z =Statistic for a level of confidence, in this case the level of confidence is 95% (1.96)

p =Expected prevalence which in this case will be 12.5% (0.125) (Otu *et al.*, 2022)

d =Precision at 5% (0.05)

Thus,

$$n = 1.96^2 \times 0.125 (1 - 0.125) / 0.05^2$$

$$n = 168$$

With attrition of 5% the sample size become 176.

Sampling Technique

Convenient sampling technique was used in this study.

Data Collection

Characteristics data was collected using an observational methods of data collection. Detail information about the number of tap and door handles in each department in Faculty of Allied Health Science was obtained.

Sample Collection

A solution of 1% glucose peptone in bijou bottle was used to moistened the swab and swabbed the tap and door handles (inner and outer handles), and the cotton end were then placed into the bijou bottle containing 10ml of 1% glucose peptone. The samples were labelled accordingly (Satyada and Sandle, 2016).

Sample Transportation

The specimen were immediately transported to the laboratory and incubated for 24 hours after which a ten-fold serial dilution were carried out on each sample before been inoculated (FAO, 2006).

Sample Preparation

One part of each incubated sample was transferred to nine part of another fresh 1% glucose peptone water before inoculated on the media (FAO, 2006).

Sample Analysis

As part of the methodology, Direct Gram's Staining Technique, Potassium Hydroxide (KOH) Preparation were done directly on the sample (Ochei and Kohlhatkar, 2013)

Culture and Isolation

A serial dilution of the samples were made before inoculation onto freshly prepared Potatoes Dextrose Agar (PDA) supplemented with antibiotics and incubated at 28°C for 4-7 days (for mould growth) and 37°C (for yeast growth); and onto Nutrient Agar and Chocolate agar were incubated at 37°C for 24 hours (for bacterial growth). All fungal culture are incubated for minimum of 30 days before discarding as negative (FAO, 2006).

Purity Plate

From the primary plates, well separated colonies of both fungi and bacteria were sub-cultured. For fungi, onto separated PDA plate by picking minute number of hyphae or spores onto a centre of non-inoculated solidified agar plate, so as to support best colonial growth and spore formation of fungi. The plates were then incubated at 28°C for 72 hours (for mould) and at 37°C (for yeast and bacteria). Fungal growth (pure culture) was usually seen from three (3) days after incubation (Al-mohanna, 2016).

Characterization of the isolates

Several tests were carried out to further characterize and identify the isolates after growth on culture media, and this depend on the colonies obtained.

For fungi: include observing the colonial morphology, smell of the cony and Gram reaction (to check for oval budding yeast) in case of *Candida* species and the use of Lactophenol Cotton Blue (LPCB) for *Aspergillus* and other molds species (Cheesebrough, 2006).

For the bacteria:

Catalase and Citrate Utilization Test (Cheesebrough, 2006); Indole, and Coagulase Test (Baker and Silverton, 2014); Methyl Red Test, and Urease Test (Ochei and Kohlhatkar, 2013), Oxidase Test, Triple Sugar Iron (TSI) Test, and Voges Proskauer (VP) Test (Cheesebrough, 2006) were done to further identify them.

Statistical Analysis

The data generated was analyzed using statistical package for social sciences SPSS version 20

RESULT

A total of one hundred and seventy-six (176) samples were collected from both tap and door handles of offices, lecture rooms, laboratories, library and toilets in Faculty of Allied Health Science, Bayero University, Kano. The following fungi and bacteria were isolated *Candida* species, *Aspergillus fumigatus*, *Aspergillus flavus*, *Aspergillus niger*, *Penicillium* species, *Rhizopus* species, *Mucor* species, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Staphylococcus saprophiticus*, *Streptococcus* species, *Micrococcus* species, *Bacillus* species, *Escherichia coli*, *Salmonella* species, *Klebsiella* species, *Pseudomonas* species and *Proteus* species.

The Distribution of positive and negative samples sources from different sample sites, tap (laboratory and toilets); door handles (offices, lecture rooms, laboratories, library and toilets) are presented (Table 1). Distribution of positive samples based on interior and exterior door handles were also presented (Table 2). Distribution of the isolates according to the type of organisms isolated in the study are shown in (Table 3) some samples had both fungi and bacterial isolates. Distribution of isolates according to Gram's staining reaction and morphology are also presented in (Table 4). Distribution of the different fungal species isolated are also presented in (Table 5). The different bacterial species identified in the study were also presented (Table 6). The distribution of the different fungi and bacteria isolated based on their sources were presented (Table 7).

Table 1: Rate of positive and negative results from the different sample sources

Fomites	Sample Site	NE(25)	NP	Percentage (%)
Tap handles	Laboratories	„	19	16.7
	Toilets	„	20	17.5
Door handles	Offices	„	8	7.0
	Lectures room	„	14	12.3
	Laboratories	„	18	15.8
	Libraries	„	11	9.6
	Toilets	„	24	21.1
Total		176	114	100

Key: NE =number examine, NP= number positive

Table 2: Distribution of positive samples of interior and exterior door handles

Type of door handle	NE(63)	NP	Percentage (%)
Interior	„	43	57.3%
Exterior	„	32	42.7%
Total	126	75	100%

Key: NE =number examine, NP= number positive

Table 3: Distribution of isolates according to type of organisms isolated

Type of organism	Number of isolate	Percentage (%)
Fungi	46	25.3%
Bacteria	136	74.7%
Total	182	100%

Table 4: Distribution of isolates according to grams staining reaction

Gram reaction	No. of isolates	Percentage (%)
Gram positive cocci	66	48.5
Gram positive bacilli	11	8.1
Gram negative cocci	0	0.0
Gram negative bacilli	55	43.4
Total	136	100

Table 5: Distribution of the different fungal species isolated

Fungi	Tap handle	Door handle	Frequency	Percentage (%)
<i>Candida</i> spp	6	8	14	30.4
<i>A. fumigatus</i>	6	4	10	21.7
<i>A. flavus</i>	2	3	5	10.9
<i>A. niger</i>	2	1	3	6.5
<i>Penicillium</i>	5	5	10	21.7
<i>Rhizopus</i> spp	2	1	3	6.5
<i>Mucors</i> spp	1	0	1	2.2
Total	24	22	46	100

Key: A = *Aspergillus*, spp = species

Table 6: Distribution of the different bacterial species isolated

Bacteria	Tap handle	Door handle	Frequency	Percentage (%)
<i>S. aureus</i>	7	31	38	27.9%
<i>E. coli</i>	7	14	21	15.4%
<i>S. epidermidis</i>	3	9	12	8.8%
<i>Bacillus spp</i>	2	9	11	8.1%
<i>Klebsiella spp</i>	1	10	11	8.1%
<i>Salmonella spp</i>	5	5	10	7.4%
<i>Pseudomonas spp</i>	5	4	9	6.6%
<i>Micrococcus spp</i>	2	6	8	5.9%
<i>Proteus spp</i>	4	4	8	5.9%
<i>Streptococcus spp</i>	1	5	6	4.4%
<i>S. saprophyticus</i>	0	2	2	1.5%
Total	37	99	136	100%

Key: *S* = *Staphylococcus*, *E* = *Escherichia*, *spp* = species

Table 7: Percentage distribution of the different fungi and bacteria isolated by type of fomite

Sample source	Number of fungal isolates	Percentage (%)	Number of bacterial isolate	Percentage (%)
Tap handles	24	52.2%	37	27.2%
Door handles	22	47.8%	99	72.8%
Total	46	100%	136	100%

DISCUSSION

Micro-organisms such as fungi and bacteria are found everywhere, even on objects surfaces such as tap and door handles. Many researches have shown that these surfaces are better at transmitting pathogens than other types of surfaces, leading to easy spread of pathogens and causing diseases to the affected individuals (Kawo *et al.*, 2012).

In this research, interior door handles had 43(57.3%) while the exterior had 32(42.7%) of the positive samples. This observation is in conformity with the finding of Shade and Mark, 2023, who also discovered that interior part of handles are more contaminated 47% compared to the exterior part 38% (Shade and Mark, 2023), this is largely due to the fact that interior handles has less contact with air and sunlight and are more often touched with contaminated hands especially for toilets.

From the positive samples, bacteria were the predominant 136(74.7%) while fungi had 46(25.3%). This agrees with the findings of Nworie and colleagues, in their work on handles/knobs in selected public conveniences (Nworie *et al.*, 2012). The distribution of isolates according to Gram's staining reaction and morphology revealed that Gram positive cocci had 66(48.5%) while Gram negative bacilli with 59(43.4%). This observation aligned with findings of (Nworie *et al.*, 2012)

who reported to be 46% for gram positive cocci as against 32% for gram negative bacilli. The findings among the fungi revealed that, *Candida spp* had the highest frequency with 14(30.4%) followed by *Aspergillus fumigatus* and *Penicillium spp* with 10(21.7%) each. While for bacterial isolates *Staphylococcus aureus* showed the highest frequency with 38(27.9%) followed by *Escherichia coli* 21(15.4%), *Staphylococcus epidermidis* 12(8.8%), *Bacillus spp* 11(8.1%), *Klebsiella spp* 11(8.1%), *Salmonella spp* 10(7.4%), *Pseudomonas spp* 9(6.6%), *Micrococcus spp* 8(5.9%), *Proteus spp* 8(5.9%), *Streptococcus spp* 6(4.4%) and *Staphylococcus saprophyticus* 2(1.5%). A similar findings was also reported by Oluduro *et al.* (2011).

From the finding, fungal isolates on tap handles had higher frequency with 24(52.2%), and door handles had 22(47.8%). While for bacteria, door handles had 99(72.8%) while tap handles had 37(27.2%). This is in line with report from other researchers Al-harmoosh *et al.* (2018) and Jabber *et al.* (2016), who reported higher fungal presence on tap than bacteria but more bacteria than fungi on door handles. The presence of these varieties of pathogens associated with these surfaces is of serious public health concern.

CONCLUSION

The study shows that, tap and door handles of offices, lecture rooms, laboratories, library and toilets can become reservoirs of infection when contaminated by users. Hence, one can easily get infected through these surfaces with either fungi, bacteria or both.

RECOMMENDATION

Based on the finding of this research, we want to recommend for frequent cleaning of taps and door handles to help (for Laboratories offices and toilets among others) limit the level spread of the pathogen via hand contact.

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