



## **Exploring Multidrug-Resistant Staphylococci on Community Surfaces: A MALDI-TOF MS Analysis from Nigeria**

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### **Abstract**

Recently, there is an apprehension of the increasing likelihood of contaminated fomites to mediate the transmission of infectious diseases, especially caused by staphylococci, to and from humans. The present study sought to estimate the prevalence of staphylococci and their multidrug-resistant species on fomites collected from Okada town situated in Edo State, Nigeria. A total of 250 swab samples were randomly collected from fomites (tables, chairs, door handles, keys, equipment, switches and handrails). Isolation of staphylococcal colonies was done by streak plate technique. Presumptive staphylococci colonies were initially identified by standard phenotypic tests, with the suspected staphylococcal isolates subsequently confirmed by Matrix-assisted Laser Desorption/Ionization Time of Flight (MALDI-TOF) mass spectrometry. Confirmed isolates were tested for multidrug resistance by Kirby Bauer disc diffusion test. The overall estimated prevalence of fomites contaminated with *Staphylococcus* species was estimated at 15.60% (39/250), with door handles accounting for the most contaminated fomites. Speciation by MALDI-TOF MS identified *S. haemolyticus*, *S. kloosii*, *S. nepalensis*, *S. saprophyticus*, *S. sciuri*, *S. simulans* and *S. xylosus*, with *S. sciuri* occurring most frequently on the fomites. All staphylococcal colonies were resistant to all beta-lactam antibiotics tested, but were generally most sensitive to fluoroquinolone (ciprofloxacin) and aminoglycoside (gentamicin) classes of antibiotics. The likelihood of contamination of fomites with multidrug-resistant staphylococci was estimated at 11.20% (28/250). The contaminated fomites were found to be a potential source of multidrug-resistant staphylococci since the reported MAR (0.79) exceeded the recommended limit (0.2). Appropriate hygienic measures to mitigate potential staphylococcal cross-contamination mediated by the contaminated fomites are recommended to avoid potential significant health risks in the future.

**KEYWORDS:** Antibiotics, Fomite, MALDI-TOF MS, Multidrug-resistant staphylococci, Multiple antibiotic resistance index (MAR).

### **Introduction**

Fomites refer to inanimate objects that are capable of spreading infections upon contamination with pathogenic microbes (Barrie *et al.*, 1994; Nwankwo, 2012). Several epidemiological studies have revealed that fomites are important vehicles for transmission of

human pathogens in environments such as sports facilities, child-care facilities, hospitals, as well as other outdoor and indoor environments (Bloomfield & Scott, 1997; Bures *et al.*, 2000; Al-Harbi *et al.*, 2017). Myriads of microbes such as *Pseudomonas*, *Streptococcus*, *Staphylococcus*, *Serratia*, *Campylobacter*, *Aspergillus*, *Penicillium* and viruses have been reported as



contaminants of fomites, and are widely distributed in outdoor and indoor environments (Al-Harbi *et al.*, 2017). Some of the fomites where contamination has been reported include telephones (Missri *et al.*, 2019), computer keyboards (Smibert *et al.*, 2018), healthcare personnel attire and devices (Haun *et al.*, 2016), as well as towels (Harrison *et al.*, 2003) and handrails (Harrison *et al.*, 2003). The spread of pathogenic microorganisms from fomites to humans is driven by factors such as the type of surfaces, type of microorganisms, as well as the hydrophobicity and moisture content of the contact surfaces (Rusin *et al.*, 2002; Stephens *et al.*, 2019). The cross-contaminations that is associated with fomites-to-human routes have been reported to be significantly ameliorated by physical cleaning (Cogan *et al.*, 2002; De Jong *et al.*, 2008), sanitization (Rutala *et al.*, 2007), inclusive of the use of antimicrobial additives and surface coatings to manufacturing fomites such as door knobs, fabrics and telephones (Kalyon & Olgun, 2001). *Staphylococcus* species are known worldwide as a cause of human and animal infections such as bacteremia, wound infections and mastitis (Scott & Bloomfield, 1990; Jones, 2017). They can be differentiated by their ability to produce coagulase, with coagulase-positive staphylococci regarded as more pathogenic than coagulase-negative species (Al-Ghamdi *et al.*, 2011). However, the carriage of antibiotic resistance genes amongst strains of the *Staphylococcus* species has been reported to be responsible for increased morbidity and mortality, higher healthcare costs and prolonged hospitalization (Simoes *et al.*, 2011; Osei-Sekyere & Mensah, 2020). Even though the transmission of *Staphylococcus* species is mainly via direct human-to-human skin contact route, fomites are also important reservoirs for the spread of *Staphylococcus* species because of their ability to survive on fomites for long periods (Neely & Maley, 2000).

In recent times, matrix-assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF MS) is increasingly being used for microbial identification (Reynolds *et al.*, 2005; Tsuchida *et al.*, 2020). This is because an infinitesimal quantity of samples is often needed by the equipment for analysis, as well as the ease of use of the equipment and the ability to identify either culturable or non-culturable microorganisms. MALDI-TOF MS technique produces microbial peptides fingerprint spectrum that ensures accurate identification of microbial species. Unlike molecular biology, MALDI-TOF MS exhibit no genomic features of the analyzed microbes, thus making this technique a taxonomic tool that does not have direct phylogenetic components (Moore *et al.*, 2002).

### **Materials and methods**

A total of 250 swab samples were randomly collected from fomites located at the Igbinedion University Teaching Hospital (IUTH), the Microbiology Laboratory at the Department of Biological Sciences, Igbinedion University, as well as from other areas in Okada town such as the grocery stores and the central market. The fomites that were screened at the sampling locations included the tables, chairs, door handles, keys, switches, handrails and equipment (computers, television, shopping carts and baskets, incubators and microscopes). Sterile cotton swab sticks moistened in 3 ml tryptic soy broth were used to swab the surfaces of the fomites and subsequently transported in ice packs to the Microbiology Laboratory at the Igbinedion University for microbiological analysis.

### **Bacterial enrichment**

Contents of each of the swab sticks used for sample collection were rinsed into the 3 ml tryptic soy broth and the broth incubated at 37°C overnight.

### **Bacterial isolation and phenotypic identification**



A loopful of each of the enriched samples was inoculated onto duplicate mannitol salt agar (MSA) plates by the streak plate technique (Public Health England, 2014). The inoculated Petri dishes were then incubated at 37°C for 48 hours. After incubation, presumptive staphylococcal colonies on the agar Petri plates were identified by phenotypic tests according to standard methods (Krieg & Holt, 1984). The phenotypic tests that were performed included Gram staining, coagulase, catalase and haemolysis tests.

#### **Bacterial identification by Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS)**

Bacterial isolates that were suspected to be staphylococci by phenotypic tests were prepared for mass spectrometry analysis using the formic acid extraction protocol as previously described (Chen *et al.*, 2015; Wireko, *et al.*, 2021). Pure colonies of suspected staphylococci cultures were suspended in 300 microlitres distilled water, and suspension was subsequently mixed with ethanol (90% v/v) followed by centrifugation at  $13000 \times g$  for 2 minutes. Twenty-five microlitres of formic acid (70% v/v) was used to re-suspend the pellets obtained from centrifugation, followed by the addition of 25 microlitres of pure acetonitrile. The mixture was further centrifuged for 2 minutes at  $13000 \times g$ . Aliquots of the supernatant (0.5 µl) containing the bacterial proteins were spotted in duplicate onto the MALDI Ground Steel target (Bruker Daltonics, Coventry, UK) and air-dried at room temperature for 5 minutes, followed by overlaying of 1 µl  $\alpha$ -Cyano-4-hydroxycinnamic acid matrix solution on each target spot. Analysis of the bacterial isolates was performed by matrix-assisted laser desorption/ionization-time of flight mass spectrometry (MALDI-TOF-MS) (Microflex LT, MALDI-TOF MS, Bruker Daltonics, Coventry, UK) in a positive linear

mode (2000 to 20000  $m/z$  range). The spectra that were obtained for each of the bacterial cultures were analyzed by MALDI-Biotyper 2.0 software (Bruker Daltonics, Coventry, UK) that compared each spectrum to the reference spectrum in the Bruker Taxonomy Database which identified the best match from the database records. The results were expressed as scores (IS) from 0 – 3. Scores that were less than 1.7 ( $> 1.7$ ) were not a reliable identification parameter. Genus's identification corresponded to scores that were between 1.70 and 1.99 (1.70 – 1.99), while scores that were greater than or equal to 2.0 ( $\geq 2.0$ ) corresponded to acceptable species-level identification.

#### **Antibiotic susceptibility test**

Suspected staphylococci colonies that were confirmed to be *Staphylococcus* species by MALDI-TOF MS were tested for multidrug resistance by the Kirby Bauer disc diffusion technique according to the guidelines prescribed by the Clinical and Laboratory Standards Institute (Clinical and Laboratory Standards Institute, 2014). Saline suspension of each pure bacterial colony was incubated and subsequently adjusted to 0.5 McFarland turbidity standards, followed by inoculation on Mueller–Hinton agar plates and the addition of antibiotic discs on the agar surface. The inoculated Petri dishes were incubated at 35°C for 16 hours. After incubation, the inhibitory zone diameter around each of the bacterial colonies was interpreted as resistant, intermediate, or sensitive based on zone diameter interpretive standards (breakpoints) recommended by the Clinical and Laboratory Standards Institute. *Staphylococcus aureus* ATCC 25923 was used as a reference strain for the disc diffusion test. The bacterial isolates were tested against antibiotic discs that included erythromycin (15 µg), gentamicin (10 µg), ampicillin (10 µg), ciprofloxacin (5 µg), pefloxacin (5 µg), ceftriaxone (30 µg), cotrimoxazole (30 µg), ceftiofur (30 µg) and



cefuroxime (30 µg). All the antibiotics employed in this study belonged to five antibiotic classes. Bacterial colonies that exhibited resistance to at least three antibiotics from three different antibiotic classes out of the five different antibiotic classes examined were regarded as multidrug-resistant.

### Estimation of multiple antibiotic resistance indices

Multiple antibiotic resistance indices (MAR) of the staphylococci colonies were determined according to the method of Krumperman (Krumperman, 1983). The MAR estimated the risk of acquiring multidrug-resistant staphylococci from the fomites, and was calculated as follows:

$$MAR = \frac{\sum(AR)}{A \times B} \quad (2)$$

MAR is the mean multiple antibiotic resistance index. AR is the antibiotic resistance scores of each *Staphylococcus*

isolate. AR represents the sum of antibiotic classes to which a particular *Staphylococcus* colony exhibited resistance. A is the total number of antibiotic classes tested. B is the total count of *Staphylococcus* isolates examined. A MAR value that was greater than 0.2 (> 0.2) indicated a high-risk source of acquiring multidrug-resistant staphylococci.

### Statistical analysis

The prevalence data were estimated as a percentage, with 95% confidence intervals. A Chi-square test was used to compare the presence of fomites in Okada town and their probable contamination with staphylococci. A p-value of less than 0.05 was considered significant in all circumstances. NCSS (version 12) was used to analyze the data.

## Results

### Characterization of staphylococcal isolates on fomites

Table 1 shows the characterization of suspected staphylococci by MALDI-TOF MS. Each of the presumptive staphylococcal colonies that were subjected to phenotypic tests was suspected to be *Staphylococcus* genus if it was shown to be Gram-positive cocci in clusters, coagulase-variable and catalase-positive. All the 39 staphylococcal isolates obtained from the 250 fomites that were suspected to be staphylococci by the phenotypic tests were all confirmed to be staphylococcal species by the MALDI-TOF MS, corresponding to an overall estimated prevalence of 15.60% (39/250). Speciation by the MALDI-TOF MS indicated that the identified *Staphylococcus* species included *S. haemolyticus*, *S. kloosii*, *S. nepalensis*, *S. saprophyticus*, *S. sciuri*, *S. simulans* and *S. xylosus*. All the confirmed species of

staphylococci found on the fomites were negative for the coagulase test and were regarded as coagulase-negative staphylococci. *S. sciuri* were the species that occurred most frequently on the fomites. *S. haemolyticus* and *S. sciuri* were the species that were isolated from switches. *S. kloosii*, *S. nepalensis*, *S. sciuri* and *S. xylosus* were found on door handles. *S. kloosii*, *S. sciuri* and *S. xylosus* were isolated from chairs; while on the handrails; *S. saprophyticus* and *S. sciuri* were the species that were isolated. *S. sciuri* were the only species that were found on the tables, keys, mobile phones and equipment that were examined.

### Prevalence of contaminated fomites

The prevalence or likelihood of contamination of the fomites by *Staphylococcus* species is presented in Table 2. The likelihood of contamination of the different types of fomites with *Staphylococcus* species



ranged from 7.81% to 57.14%, with the door handles accounting for the most contaminated fomites and the tables accounting for the least staphylococcal-contaminated fomites. Overall, 15.60% of all the fomites examined in this study were contaminated with *Staphylococcus* species.



Table 1: Characterization of confirmed staphylococci on fomites by MALDI-TOF MS

Identity of confirmed staphylococci on fomites	Prevalence of the identified staphylococci			MALDI-TOF-MS identification scores for the tested staphylococcal isolates	Implicated fomites
	H/N	M (%)	95% CI (%)		
<i>Staphylococcus haemolyticus</i>	1/250	0.40	0.00 – 1.19	2.1	Switch
<i>Staphylococcus kloosii</i>	2/250	0.80	0.00 – 1.91	2.1, 2.2	Door handle and chair
<i>Staphylococcus nepalensis</i>	1/250	0.40	0.00 – 1.19	2.0	Door handle
<i>Staphylococcus saprophyticus</i>	1/250	0.40	0.00 – 1.19	2.0	Handrail
<i>Staphylococcus sciuri</i>	11/250	4.40	1.85 – 6.95	2.0, 2.1, 2.1, 2.2, 2.1, 2.0, 2.1, 2.2, 2.1	Chair, table, handrail, switch, mobile phone, door handle, key, and equipment
<i>Staphylococcus simulans</i>	1/250	0.40	0.00 – 1.19	2.3	Handrail
<i>Staphylococcus xylosus</i>	2/250	0.80	0.00 – 1.91	2.0, 2.2	Chair and door handle
Other confirmed staphylococci	20/250	8.00	4.63 – 11.37	1.7 – 1.9	Door handle, keys, chair, table, handrail, switch, keys, mobile phones and equipment

H: count(s) of confirmed staphylococci on fomites; N: total number of fomites examined; M: mean prevalence; CI: confidence interval of mean

Table 2: Prevalence of fomites contaminated by *Staphylococcus* species

Fomites	Prevalence of staphylococci-contaminated fomites		
	F/N	M (%)	95% CI (%)
Table	5/64	7.81	1.18 – 14.44
Chair	6/61	9.84	2.30 – 17.38
Door handle	8/14	57.14	50.77 – 63.51
Handrail	7/46	15.22	4.72 – 25.72
Key	3/14	21.43	0.00 – 43.73
Mobile phone	6/25	24.00	6.91 – 41.09
Switch	1/9	11.11	0.00 – 32.89
Equipment	3/17	17.65	0.00 – 36.33
All fomites	39/250	15.60	10.39 – 19.21

F: count(s) of confirmed staphylococci on the fomites; N: number of fomites examined; CI: confidence interval of mean; M: mean prevalence



### **Prevalence of multidrug-resistant staphylococci on the fomites**

Tables 3 and 4 show the antibiotic resistance patterns and resistance profile of confirmed *Staphylococcus* species isolated from all the fomites examined in this study. All the staphylococcal colonies obtained from the fomites were resistant to all beta-lactam antibiotics tested. Twenty-three staphylococcal colonies were resistant to all the classes of antibiotics tested out of the 39 tested staphylococcal colonies. The staphylococcal colonies were generally most sensitive to the fluoroquinolone (ciprofloxacin) and aminoglycoside (gentamicin) classes of antibiotics. Twenty-eight staphylococcal colonies were detected to be multidrug-resistant. The 28 multidrug-resistant staphylococci were distributed across 28 different fomites examined in this study. Hence, the likelihood of contamination of all the examined fomites with multidrug-resistant staphylococci was estimated at 11.20% (28/250).

### **Estimates of MAR associated with contaminated fomites**

Overall, the mean multiple antibiotic resistance index (MAR) associated with the contaminated fomites was estimated at 0.79. As indicated by MAR, the contaminated fomites were found to be a potential source of multidrug-resistant staphylococci, with a significant health risk, since the reported MAR exceeded the recommended limit of 0.2.

### **Association between the fomites and presence of staphylococci**

Table 5 shows the contingency table for estimating the association between the fomites and the presence of staphylococci and their multidrug-resistant strains. The chi-square test of independence showed that no significant association was found between the fomites examined in Okada and the presence of either staphylococci ( $p = 0.44$ ) or multidrug-resistant staphylococci ( $p = 0.37$ ).



Table 3: Antibiotic resistance patterns of confirmed *Staphylococcus* species isolated from the fomites

Confirmed staphylococci	Tested colonies	Resistance patterns of the tested antibiotics
<i>Staphylococcus haemolyticus</i>	A1	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus kloosii</i>	B1	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus kloosii</i>	B2	CXM, AM, FOX, CRO, E
<i>Staphylococcus nepalensis</i>	C1	CXM, AM, FOX, CRO, E
<i>Staphylococcus saprophyticus</i>	D1	CXM, AM, FOX, CRO, E
<i>Staphylococcus sciuri</i>	E1	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus sciuri</i>	E2	CXM, AM, FOX, CRO, E
<i>Staphylococcus sciuri</i>	E3	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus sciuri</i>	E4	CXM, AM, FOX, CRO, PEF, E
<i>Staphylococcus sciuri</i>	E5	CXM, AM, FOX, CRO, E
<i>Staphylococcus sciuri</i>	E6	CXM, AM, FOX, CRO, E
<i>Staphylococcus sciuri</i>	E7	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus sciuri</i>	E8	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus sciuri</i>	E9	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus sciuri</i>	E10	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus sciuri</i>	E11	CXM, AM, FOX, CRO, E
<i>Staphylococcus simulans</i>	F1	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus xylosus</i>	G1	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus xylosus</i>	G2	CXM, AM, FOX, CRO, PEF, C
<i>Staphylococcus</i> sp.	H1	CXM, AM, FOX, CRO, E
<i>Staphylococcus</i> sp.	H2	CXM, AM, FOX, E
<i>Staphylococcus</i> sp.	H3	CXM, AM, FOX, CRO, C, GM, E
<i>Staphylococcus</i> sp.	H4	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H5	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H6	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H7	CXM, AM, FOX, CRO, PEF, E
<i>Staphylococcus</i> sp.	H8	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H9	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H10	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H11	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H12	CXM, AM, FOX, CRO, E
<i>Staphylococcus</i> sp.	H13	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H14	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H15	CXM, AM, FOX, CRO, PEF, E
<i>Staphylococcus</i> sp.	H16	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H17	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H18	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H19	CXM, AM, FOX, CRO, PEF, CIP, C, GM, E
<i>Staphylococcus</i> sp.	H20	CXM, AM, FOX, CRO, C

CXM: cefuroxime; AM: ampicillin; FOX: ceftioxin; CRO: ceftriaxone; PEF: pefloxacin; CIP: ciprofloxacin; C: cotrimoxazole; GM: gentamicin; E: erythromycin

Table 4: Antibiotic resistance profile of staphylococci colonies obtained from the fomites

Confirmed staphylococci	K	Prevalence of antibiotic resistance									MR	$\Sigma$ (AR1)	$\Sigma$ (AR2)
		CXM	AM	FOX	CRO	PEF	CIP	C	GM	E			
		30 µg	10 µg	30 µg	30 µg	5 µg	5 µg	30 µg	10 µg	15 µg			
		F (%)	F (%)	F (%)	F (%)	F (%)	F (%)	F (%)	F (%)	F (%)			
<i>Staphylococcus haemolyticus</i>	1	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1	5	153
<i>Staphylococcus kloosii</i>	2	2 (100.00)	2 (100.00)	2 (100.00)	2 (100.00)	1 (50.00)	1 (50.00)	1 (50.00)	1 (50.00)	2 (100.00)	1	7	
<i>Staphylococcus nepalensis</i>	1	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (100.00)	0	2	
<i>Staphylococcus saprophyticus</i>	1	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (100.00)	0	2	
<i>Staphylococcus sciuri</i>	11	11 (100.00)	11 (100.00)	11 (100.00)	11 (100.00)	7 (63.64)	6 (54.55)	6 (54.55)	6 (54.55)	11 (100.00)	7	41	
<i>Staphylococcus simulans</i>	1	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1 (100.00)	1	5	
<i>Staphylococcus xylosus</i>	2	2 (100.00)	2 (100.00)	2 (100.00)	2 (100.00)	1 (50.00)	1 (50.00)	2 (100.00)	1 (50.00)	2 (100.00)	2	8	
Other confirmed staphylococci	20	20 (100.00)	20 (100.00)	20 (100.00)	19 (95.00)	15 (75.00)	13 (65.00)	15 (75.00)	14 (70.00)	19 (95.00)	16	83	

K: relative count of confirmed staphylococci colonies; MR: relative count of multidrug-resistant staphylococci colonies;  $\Sigma$ (AR1): relative sum of antibiotic resistance scores;  $\Sigma$ (AR2): overall sum of antibiotic resistance scores; C: cotrimoxazole; GM: gentamicin; E: erythromycin; CXM: cefuroxime; AM: ampicillin; FOX: cefoxitin; CRO: ceftriaxone; PEF: pefloxacin; CIP: ciprofloxacin



Table 5: Contingency table for estimating the association between the fomites and a probable contamination by staphylococci and their multidrug-resistant strains

Sampling locations in Okada metropolis	Number of fomite samples	Presence of staphylococci		Chi square test for staphylococci		Presence of multidrug- resistant staphylococci		Chi square test for MRS	
		Yes	No			Yes	No		
				X <sup>2</sup>	p-value			X <sup>2</sup>	p-value
				2.70	0.44			3.16	0.37
IUTH	55	9	46			8	47		
Microbiology Laboratory	51	11	40			8	43		
Grocery stores	48	8	40			5	43		
Central market	96	11	85			7	89		

IUTH: Igbinedion University Teaching Hospital. MRS: multidrug-resistant staphylococci



## Discussion

Door handles accounted for most of the contaminated fomites (Tables 1 and 2). Hence the door handles may be the most implicated probable source of acquiring infections in Okada metropolis. In consistence with the current study, Eze *et al.* (2012) and Moore *et al.* (2002) also reported huge bacterial contaminations in door handle swab samples collected from Abuja, Nigeria and United Kingdom, respectively. The high prevalence of contaminated door handles in this study may be due to frequent contact by the human inhabitants and inappropriate hygienic conditions of the environment. Handrails, keys, telephones, as well as equipment such as computers, television, shopping carts and baskets, incubators and microscopes were also found to be significantly contaminated with staphylococci (Table 2), and may also play a substantial role in the spread of infections. Reynolds *et al.* (2005), Al-Ghamdi *et al.* (2011) and Jones (2017) have reported significant biological contamination of shopping carts and baskets with staphylococci. The frequent use of shopping carts and baskets by customers of the grocery stores could be the main reason for contamination since the customers have different hygienic statuses, and upon the handling of shopping carts and baskets, they may transmit pathogenic microbes from their hands to the handles of this shopping equipment, and vice versa.

The survival of the isolated bacterial species on the surfaces of the examined fomites may be ascribed to several factors, some of which may be the nature of the surrounding environment and characteristics of the bacterial species. Many bacteria can use their structures, such as their glycocalyx and flagella, to adhere and survive on fomites and on the hands for several hours (Scott & Bloomfield, 1990).

Transmission of bacterial pathogens may either be through direct or indirect routes. The transmission of bacterial pathogens from contaminated fomites to the hands of humans, and ultimately into the human body

by ingestion or inhalation, is a typical example of the indirect pathway. Hence the contaminated fomites examined in this study could indirectly cause severe health problems in humans if adequate hygienic practices are not applied. Typical direct pathways of bacterial pathogen transmission include human to human transmissions. *S. sciuri*, *S. xylosus*, *S. simulans*, *S. haemolyticus*, *S. kloosii* were the main multidrug-resistant *Staphylococcus* species that were found on the examined fomites. Some staphylococci that were identified to the genus level were also found to be multidrug-resistant (Tables 3 and 4). The findings from this study were consistent with those of Smibert *et al.* (2018) who found multidrug-resistant organisms on mobile phones and computer keyboards. The isolated multidrug-resistant bacteria derived from this study exhibited an extremely high prevalence of resistance to all the tested beta-lactam antibiotics (100%). High bacterial resistance to these beta-lactam antibiotics that are frequently used to treat humans calls for concern. However, the bacterial isolates obtained from the present study were most susceptible to the fluoroquinolones (ciprofloxacin) and aminoglycosides (gentamicin) classes of antibiotics. Hence these classes of antibiotics may still provide treatment options for humans that become indirectly infected with multidrug-resistant bacteria mediated by contaminated fomites.

## Conclusions

This study revealed that fomites in Okada metropolis were contaminated with staphylococci that have the potential of causing health risks, though not presently at a significant level. Nevertheless, appropriate hygienic measures to mitigate potential staphylococcal cross-contaminations mediated by the contaminated fomites are recommended to avoid potential significant health risks in the future.

## Conflicts of interest



There is no conflict of interest in this study.

Supplement, CLSI document M100-S24. Wayne, PA: CLSI.

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