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Bacterial Assessment of Electronic Hardware User Interfaces in Ile-Ife, Nigeria

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ABSTRACT

The study was undertaken to quantify and identify bacterial contaminants associated with private and open access user interfaces in various establishments in the town of Ile-Ife, Nigeria. The study was conducted in selected offices, business centres, banks and cybercafés within Ile-Ife. Swab samples were aseptically collected from each user interface (keyboard, mouse, ATM) and users' hands and cultured on nutrient and MacConkey agar, to determine the total bacterial load and coliform count, respectively, by the pour-plate technique. Bacterial loads present on different types of interface (keyboard, mouse and ATM) were found to be significantly different ($p < 0.01$). A total of 669 isolates comprising 11 distinct bacterial species were recovered from 313 randomly sampled user interfaces. The frequencies of occurrence of the species were *Aerococcus viridans* (9.4%), *Bacillus* spp. (8.4%), *Enterobacter aerogenes* (4.9%), *Gaffkya tetragena* (2.1%), *Klebsiella pneumoniae* (11.1%), *Micrococcus luteus* (10.9%), *Moraxella catarrhalis* (1.6%), *Proteus* spp. (10.6%), *Pseudomonas aeruginosa* (16.0%), *Staphylococcus aureus* (16.7%) and *Staphylococcus epidermidis* (8.2%). All the interfaces examined were contaminated. Contamination on interfaces in educational institutions differed significantly from that found in banks and cybercafés, but was comparable to that in commercial centres. Most isolates were resistant to amoxicillin, augmentin, nitrofurantoin and ceftriaxone, while resistance to ciprofloxacin and ofloxacin was the least frequent. Multiple antibiotic resistance was observed in 89.1% of bacterial isolates, with a total of 68 resistance patterns, resistance to three antibiotics being the most frequent (31.9%). About 74% of multiple antibiotic resistant isolates profiled for plasmid DNA contained either single or multiple plasmids. It was concluded that user interfaces were contaminated with potentially pathogenic bacteria, highly resistant to some commonly used antibiotics. These interfaces are therefore potential vehicles for the transmission of clinically important pathogens.

Keywords: Bacterial. Contaminants. Antibiotics. Resistance. Plasmid.

INTRODUCTION

Contamination of environmental objects and surfaces is a common phenomenon. The presence of viable pathogenic bacteria on inanimate objects has been reported by earlier investigators. Several studies of the human environment have demonstrated colonization and contamination of objects such as door handles, faucets, phones, money, fabrics and plastics (Bures et al., 2001; Michael et al., 2001; Despina et al., 2008; Famurewa & David, 2009). People come into daily contact with all sorts of fomites, with an increasing rate of bacterial infection (Eguia & Chambers, 2003). Human beings have a marked tendency to pick up microorganisms from environmental objects and the hand has been shown to play a role in the transmission of organisms. Colonization of objects by pathogenic organisms has been reported as a potential vehicle for their transmission (Neely & Maley, 2000; Gerba, 2005; Famurewa & David, 2009; Fatma et al., 2009, Fraser & Girling, 2009; Gholamreza et al., 2009). Furthermore, microorganisms found to contaminate fomites have also been shown to persist on environmental surfaces for varying periods of time ranging from hours to months and it has also been illustrated that they can still be detected and recovered from surfaces after routine conventional cleaning (French et al., 2004). In addition, cross infection of microorganisms between environmental surfaces and a host has equally been established (Hardy et al., 2006). The ability of plastics and other inanimate objects to support viable microorganisms for a prolonged period of time is well documented (Stuart et al., 2006) and such environmental surfaces and objects, especially those in

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close proximity with persons and frequently touched, pose a threat to human health and are a cause for concern. One such inanimate object in the environment that is currently in frequent contact with the hands is the interface of a computer system or an automated teller machine (ATM).

The use of hardware interfaces such as the keyboard, mouse and ATM keypad has greatly expanded over the past few years with the development of various forms of computer-based management applications. Personal computers are now ubiquitous and this is due to the fact that computers have become widely available and affordable, as well as easy to use, with the introduction of the graphical user interface (GUI) (Onibere et al., 2001). Nowadays, the implementation and use of computer systems and consequently interfaces is growing in schools, offices, cybercafés and hospitals, so that interfaces continue to have an increased presence in almost every occupational, recreational and residential environment. This upsurge has consequently led to regular and unrestricted sharing of interfaces among users. With the harboring of microorganisms acquired from the human microflora or as transient organisms from the environment, and previous accounts of cross contamination of microorganisms (Lindberg et al., 2004; Hardy et al., 2006), it is readily conceivable that pathogens could be transferred among users who share interfaces.

Several investigations have assessed the degree of microbial contamination and the types of contaminating organisms on computer keyboards (Schultz et al., 2003; Issmat et al., 2007; Anderson & Palombo, 2009). Some authors have demonstrated such contamination on the computer keyboard and mouse (Steffen et al., 2008). Concern has been raised that contact with contaminated computer keyboards might serve as a mechanism for contaminating the hands with potential pathogens, leading to cross-contamination of users (Steffen et al., 2008; Anderson & Palombo 2009). One study conducted in a hospital established the fact that the colonization rate of computer user interfaces was greater than that of other fomites tested in the hospital (Schultz et al., 2003). Accordingly, these may be additional reservoirs for the transmission of microorganisms and become vehicles for cross contamination. While the contamination of user interfaces has thus been established, most of the above studies were single-centred, having a narrow perspective as they either focused on hospital and health care facilities or were specific to the isolation of a particular microorganism, species or strain or specific to only one type of interface. In view of these findings: the growth and detection of opportunistic pathogens on computer user interfaces, survival of bacteria on surfaces and a low rate of compliance with good hygiene practice, it is imperative to examine the extent of bacterial contamination on interfaces used by different people under everyday conditions and in various types of institution or organization and to investigate probable sources of high contamination rates.

MATERIALS AND METHODS

Study area and study population

The study was conducted in selected offices, business centres, banks and cybercafés within and outside the Obafemi Awolowo University (OAU), Ile-Ife, Nigeria. It was a multi-perspective microbiological assessment of three user interface types (keyboard, mouse and ATM). This cross-sectional study was conducted in 2010 and a total of 313 swab samples, from 141 keyboard, 140 mouse and 20 ATM surfaces, and 12 samples from interface users' hands, were collected in various departments of 2 educational institutions, 6 cybercafés, 4 banks and 4 commercial outfits in Ile-Ife.

Sources and collection of samples

Samples were collected from the keyboard and mouse of computer systems that were either privately owned or used and from those in open access areas, shared by an array of different users or open to the general public. Sterile moistened cotton-tipped swabs were methodically moved several times over the surfaces of some selected frequently-used keys on the keyboard and ATM keypad and over the left and right click buttons on the computer mouse; swabs were also rubbed over the entire surface of the hands (including the thumb and the fingers) of interface users. Swabs were processed within an hour of collection. Gender, profession, duration of interface use and routine cleaning of the interfaces were recorded.

Processing and identification of isolates

All samples were identified with a laboratory number, which was a combined code for the place of sampling, interface type and ID number. All samples were cultured by the pour-plate method on MacConkey agar (Oxoid) for coliform enumeration and Nutrient agar (Oxoid) for total bacterial count. Plates were incubated at 37^{o}C for 24 hours, after which the colonies grown were counted. Pure cultures of the colonies were obtained by subculturing on fresh nutrient agar plates. Isolated bacteria were identified by extensive phenotypic testing, Gram stain and conventional biochemical identification procedures (Barrow & Felthan, 2004).

Antibiogram of isolates

An antibiogram of the 18-hour pure cultures of the isolates was obtained by the disk diffusion method, carried out on Diagnostic Sensitivity Test (DST) agar, according to CLSI (2006). The antibiotic dose employed included: 25µg amoxicillin, 5µg ofloxacin, 10µg streptomycin, 30µg chloramphenicol, 10µg gentamicin, 5µg pefloxacin, 25µg cotrimoxazole, 10µg ciprofloxacin, 10µg erythromycin, 30µg ceftriaxone, 30µg augmentin, 200 µg nitrofurantoin and 30µg tetracycline. The antibiotic discs were firmly placed on the DST agar plates previously lawn seeded with the standardized inocula. After 24h incubation at 37^{o}C, zones of inhibition were measured with a ruler calibrated in millimeters.

Plasmid extraction

Forty-five multiple antibiotic resistant isolates, randomly selected from keyboard, mouse and ATM surfaces and users' hands at 2 sampling sites (a cybercafé

and a departmental office) were profiled for the presence of plasmid DNA. The purified bacterial isolates profiled for plasmid DNA were identified as *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Staphylococcus aureus, Micrococcus luteus* and *Enterobacter aerogenes.* Plasmid extraction was carried out by the TENS method described by Zhou et al., *(*1990). The extracted plasmids were then separated by horizontal 1% agarose gel electrophoresis and viewed on a UV transilluminator after staining with ethidium bromide.

RESULTS

The bacterial loads on the user interfaces examined are presented in Table 1. Of the keyboard samples, 59.6% had total bacterial counts within the range 31 to 299 cfu/mL, while 37.9% had bacterial counts of <= 30, this range being more frequent on mouse interfaces than on other types. Meanwhile, there were more ATM interface samples with bacterial contamination loads exceeding 300 cfu/mL than in any lower range. Bacterial loads on the three interface types (keyboard, mouse and ATM) were found to be significantly different ($p < 0.01$). Analysis of variance revealed that, for both the total bacterial and coliform counts, the rate of contamination on the hand was significantly higher than that on the keyboard ($p < 0.01$) and mouse ($p < 0.001$) and comparable with that on the ATM ($p > 0.05$) (Table 1).

Table 1: Total bacterial and coliform counts on the different interface types

	Bacterial Load (cfu/mL of 25cm^2)								
Sources	≤ 30		31-150		150-299		≥ 300		Total
	TBC	CFC	TBC	CFC	TBC	CFC	TBC	CFC	
Keyboard	31(22.0%)	55(39.0%)	56(39.7%)	54(38.3%)	28(19.9%)	21(14.9%)	26(18.4%)	11(7.8%)	141
Mouse	53(37.9%)	71(50.7%)	49(35.0%)	50(35.7%)	21(15.0%)	12(8.6%)	17(12.1%)	7(5.0%)	140
Hand	1(8.3%)	1(8.3%)	2(16.7%)	5(41.7%)	3(25.0%)	2(16.7%)	6(50.0%)	4(33.3%)	12
ATM	6(30.0%)	7(35.0%)	4(20.0%)	5(25.0%)	2(10.0%)	3(15.0%)	8(40.0%)	5(25.0%)	20
Total	91(29.1%)	134(42.8%)	111(35.5%)	114(36.4%)	54(17.3%)	38(12.1%)	57(18.2%)	27(8.6%)	313
χ^2	TBC: 27.906				CFC: 26.738				
P-value	TBC: P < 0.001				CFC: P < 0.01				

Key: TFTC: Too few to count TNTC: Too numerous to count TBC: Total bacteria count CFC: Coliform count cfu/mL: Colony-forming unit per millilitre χ^2: Chi-squared P: Probability

The bacterial load on the interfaces is presented in relation to interface users' age, occupation and organization in Table 2. Contamination on interfaces in educational institutions differed significantly from contamination on interfaces in banks and cybercafés, but was comparable to that on interfaces in commercial centres.

Table 2. Total bacterial and coliform counts on interfaces in relation to interface users' characteristics

User Attribute	Number Sampled	Bacterial Load (cfu/mL)							
		≤ 30 (TFTC)		31 – 150		151 – 299		≥ 300	
		TBC (%)	CFC (%)	TBC (%)	CFC (%)	TBC (%)	CFC (%)	TBC (%)	CFC (%)
Age									
10-19	9	5 (35.7)	5 (35.7)	3 (21.4)	5 (35.7	2 (14.3)	2 (14.3)	4 (28.6)	2 (14.3)
20-29	129	36 (27.9)	59 (45.7)	54 (41.9)	48 (37.2)	17 (13.2)	13 (10.1)	22 (17.1)	9 (7.0)
30-39	84	27 (32.1)	32 (38.1)	26 (31.0)	31 (36.9)	15 (17.9)	15 (17.9)	16 (19.0)	6 (7.1)
40-49	43	12 (27.9)	21 (48.8)	18 (41.9)	15 (34.9)	8 (18.6)	3 (7.0)	5 (11.6)	4 (9.3)
≥ 50	23	5 (21.7)	10 (43.5)	6 (26.1)	10 (43.5)	10 (43.5)	2 (8.7)	2 (8.7)	1 (4.3)
Total	293	85(29.0)	126(43.0)	107(36.5)	109(37.2)	52(17.7)	35(11.9)	49(16.7)	22 (7.5)
χ^2 (df=4)								17.821	6.909
P								0.121 (ns)	0.864 (ns)
Sex									
Male	156	41 (26.3)	65 (41.7)	58 (37.2)	58 (37.2)	27 17.3)	21 (13.5)	30 (19.2)	12 (7.7)
Female	137	44 (32.1)	61 (44.5)	49 (35.8)	52 (38.0)	25 18.2)	14(10.2)	19 (13.9)	10 (7.3)
Total	293	85(29.0)	126(43.0)	107(36.5)	109(37.2)	52(17.7)	35(11.9)	49(16.7)	22(7.5)
χ^2 (df=1)								2.186	0.792
P								0.535 (ns)	0.851 (ns)
Occupation									
Teaching	49	10 (20.4)	15 (30.6)	16 (32.7)	21 (42.9)	12 (24.5)	7 (14.3)	11 (22.4)	6 (12.2)
Office Staff	93	24 (25.8)	39 (41.9)	37 (39.8)	39 (41.9)	24 (25.8)	11 (11.8)	8 (8.6)	4 (4.3)
Student	101	27 (26.7)	41 (40.6)	39 (38.6)	37 (36.6)	11 (10.9)	13 (12.9)	24 (23.8)	10 (9.9)
Bank Cashier	22	15 (68.2)	20 (90.9)	6 (27.3)	2 (9.1)	1 (4.5)	0 (0)	0 (0)	0 (0)
Business	24	7 (29.2))	9 (37.5)	8 (33.3)	10 (41.7)	4 (16.7)	4 (16.7)	5 (20.8)	1 (4.2)
Lawyer	4	2 (50.0)	3 (75.0)	1 (25.0)	0 (0)	0 (0)	0 (0)	1 (25.0)	1 (25.0)
Total	293	85(29.0)	126(43.0)	107(36.5)	109(37.2)	52(17.7)	35(11.9)	49(16.7)	22(7.5)
χ^2 (df=5)								37.250	32.927
P								0.001	0.005
Organization									
Educational	184	41 (22.3)	68 (37.0)	77 (41.8)	80 (43.5)	35 (19.0)	22 (12.0)	31 (16.8)	14 (7.6)
Bank	42	21 (50.0)	27 (64.3)	10 (23.8)	7 (16.7)	3 (7.1)	3 (7.1)	8 (19.0)	5 (11.9)
Cybercafe	66	23 (34.8)	28 (42.4)	16 (24.2)	19 (28.8)	11 (16.7)	11 (16.7)	16 (24.2)	8 (12.1)
Commercial	21	6 (28.6)	10 (47.6)	8 (38.1)	8 (38.1)	5 (23.8)	2 (10.0)	2 (9.5)	1 (4.8)
Total	313	91(29.1)	133(42.6)	111(35.5)	114(36.5)	54(17.3)	38(12.2)	57(18.2)	27(8.7)
χ^2 (df=4)								21.862	8.664
P								0.009	0.028

KEY: χ^2 = Chi square P value= Probability ns = Not significant TBC = Total bacterial count CBC = Coliform bacteria count cfu/mL = colony forming unit per milliliter TFTC = Too few to count TNTC = Too numerous to count % = percentage df = degree of freedom

Table 3 summarizes the frequencies of isolated bacterial species on the various interface types and hands of users examined. A total of 669 bacterial isolates, comprising 376 Gram-positive and 293 Gram-negative organisms belonging to eleven distinct species, were recovered from electronic hardware user interfaces. The number of Gram-positive bacteria recovered from each interface ranged from 11 from ATMs to 175 from keyboards, while Gram-negative organisms ranged from 10 on hands to 170 on keyboards. However, five control samples, comprising unused keyboard, mouse and ATM, showed no bacterial contamination.

The prevalence of bacterial species on user interfaces (Table 3) ranged from 11 (2.1%) for *Moraxella catarrhalis* to 112 (16.7%) for *Staphylococcus aureus*. [These organisms were thus the least and most prevalent bacterial contaminants overall.] *Pseudomonas aeruginosa* (39%) was the most prevalent bacterial contaminant recovered from keyboard, *S. aureus* (36%) from mouse, *Bacillus* spp. (50%) from ATM and *Micrococcus luteus* (75%) from hands. ATMs showed the lowest diversity of contaminating bacterial species. Prevalence of bacterial contaminants on ATMs ranged from 1 (5.0%) for *Aerococcus viridans* to 10 (50.0%) for *Bacillus* spp. Users' hands were found to be highly contaminated with most of the bacterial contaminants. However, the diversity of bacterial species present on hands was lower than on keyboard and mouse but higher than on ATM. All the bacterial isolates except *E. aerogenes* and *M. catarrhalis* were recovered from the hands of interface users (Table 3). *Bacillus* spp. was more prevalent on ATM than any other interface and differed significantly from hand ($p < 0.05$), keyboard ($p < 0.01$) and mouse ($p < 0.001$) (Table 3).

Table 3: Distribution of bacterial contaminants on the different interface types

	Keyboard (N=141)				ATM (N=20)				Hand (N=12)			Total (N=313)		χ^2 (df=1)	P
Isolated species	N	%	n/N	%	n/N	n	%	n/N	n	%	n/N	n	%		
GPB	174	50.4	1.23	59.8	1.09	18	52.9	0.90	30	75.0	2.5	376	55.8		
GNB	170	49.3	1.21	39.8	0.72	13	38.2	0.65	10	25.0	0.83	293	43.5		
None (Control)	1	0.3	0.01	0.4	0.01	3	8.8	0.15	0	0.0	0.0	5	0.7	45.984	<0.001
Total	345	100	2.45	100	1.82	34	100	1.7	40	100	3.33	674	100		
Identified Bacterial Species														χ^2 (df=3)	P
Aerococcus viridans	37	26.2		13.6	1		5.0	6		50.0	63		20.1	16.529	<0.001
Bacillus spp.	26	18.4		13.6	10		50.0	1		8.3	56		17.9	16.462	<0.001
Enterobacter aerogenes	23	16.3		7.1	0		0.0	0		0.0	33		10.5	10.463	<0.05
Gaffkya tetragena	7	5.0		4.3	0		0.0	1		8.3	14		4.5	1.446	ns
Klebsiella pneumonia	42	29.8		17.1	4		20.0	4		33.3	74		23.6	6.996	ns
Micrococcus luteus	34	24.1		20.0	2		10.0	9		75.0	73		23.3	20.819	<0.001
Moraxella catarrhalis	8	5.7		2.1	0		0.0	0		0.0	11		3.5	3.881	ns
Proteus spp.	43	30.5		16.4	4		20.0	1		8.3	71		22.7	9.522	<0.05
Pseudomonas. aeruginosa	55	39.0		30.0	5		25.0	5		41.7	107		34.2	3.596	ns
Staphylococcus aureus	49	34.8		36.4	5		25.0	7		58.3	112		35.8	2.154	ns
Staphylococcus. epidermidis	20	14.2		20.7	0		0.0	6		50.0	55		17.6	14.610	<0.01

Key: GPB=Gram-positive bacteria, GNB= Gram-negative bacteria, N=Number of samples, n=Number of isolates

With regard to organization and occupation of interface users, the prevalence of multiple contaminants on interfaces was higher among teachers and students in educational institutions (Table 4). Multiple contamination occurred less frequently on interfaces used in banks by bank personnel. Among the various age groups examined, interfaces used by people aged from 20 to 39 years supported a greater variety of bacterial species than interfaces used by any other age group. Multiple contamination of interfaces by 6 bacterial species was recorded solely on interfaces whose users were within this age range. Co-contamination of interfaces with diverse bacterial species varied significantly among the three interface types: keyboard, mouse and ATM ($F = 51.527$, $P < 0.001$). Co-contamination by bacterial species was commoner on hands than on interfaces. Also, the degree of diversity of bacterial species on keyboard was significantly different from that on mouse and ATMs. However mouse and ATM were found to be comparable ($P > 0.05$) (Table 4).

The number of bacterial isolates from interfaces used by different occupational groups varied significantly ($F = 32.478$; $P < 0.001$). Also, the rate of bacterial contamination of interfaces used by office staff differed significantly from those used by academics ($p < 0.05$).

However, the prevalence of multiple bacterial contamination of interfaces was found to be similar for both male and female users. Regarding the establishments where these interfaces were sampled, the presence of multiple bacterial contaminants on user interfaces was also found to be significantly different among the various types of organization studied ($F = 29.182$, $P < 0.001$). Thus, contamination of interfaces in the educational institutions was significantly different from that in the banks and cybercafés, but comparable to interface contamination in commercial centres (Table 4).

Table 4. Distribution of bacterial contaminants on interfaces from selected organizations and occupational group

Interface users	Number of samples	*Aerococcus viridans*		*Bacillus* spp.		*Enterobacter aerogenes*		*Gaffkya tetragena*		*Klebsiella pneumoniae*		*Micrococcus luteus*		*Moraxella catarrhalis*		*Proteus* spp.		*Pseudomonas aeruginosa*		*Staphylococcus aureus*		*Staphylococcus epidermidis*	
		N	(%)	N	(%)	N	(%)	N	(%)	N	(%)	N	(%)	N	(%)	N	(%)	N	(%)	N	(%)	N	(%)
Organization																							
Educational	184	36	(19.6)	15	(8.2)	32	(17.4)	12	(6.5)	45	(24.5)	56	(30.4)	5	(2.7)	54	(29.3)	69	(37.5)	76	(41.3)	37	(20.1)
Bank	42	8	(19.0)	14	(33.3)	0	(0.0)	0	(0.0)	6	(14.3)	4	(9.5)	1	(2.4)	7	(16.7)	10	(23.8)	15	(35.7)	0	(0.0)
Cybercafe	66	14	(21.2)	24	(36.4)	0	(0.0)	2	(3.0)	17	(25.8)	11	(16.7)	3	(4.5)	5	(7.6)	23	(34.8)	19	(28.8)	10	(15.2)
Commercial	21	5	(23.8)	5	(23.8)	1	(4.8)	0	(0.0)	6	(28.6)	2	(9.5)	2	(9.5)	5	(23.8)	5	(23.8)	2	(9.5)	8	(38.1)
Total	313	63	(20.1)	56	(17.9)	33	(10.5)	14	(4.5)	74	(23.6)	73	(23.3)	11	(3.5)	71	(22.7)	107	(34.2)	112	(35.8)	55	(17.6)
χ^2 (df=3)		0.292		31.655		22.622		5.079		2.551		13.547		2.947		14.131		3.926		9.716		15.768	
P		Ns		<0.001		<0.001		Ns		Ns		<0.01		ns		<0.01		ns		<0.05		<0.001	
Occupation of interface users surveyed																							
Teaching	49	10	(20.4)	3	(6.1)	7	(14.3)	7	(14.3)	19	(38.8)	25	(51.0)	1	(2.0)	17	(34.7)	22	(44.9)	18	(36.7)	7	(14.3)
Office Staff	93	17	(18.3)	11	(11.8)	10	(10.8)	4	(4.3)	19	(20.4)	15	(15.1)	2	(2.2)	26	(28.0)	29	(31.2)	30	(32.3)	21	(22.6)
Students	101	22	(21.8)	22	(21.8)	13	(12.9)	1	(1.0)	20	(19.8)	21	(20.8)	4	(4.0)	18	(17.8)	41	(40.6)	39	(38.6)	20	(19.8)
Bank Cashier	22	7	(31.8)	4	(18.2)	0	(0.0)	0	(0.0)	2	(9.1)	2	(9.1)	1	(4.5)	3	(13.6)	5	(22.7)	10	(45.5)	0	(0.0)
Businessman	24	6	(25.0)	6	(25.0)	3	(12.5)	2	(8.3)	8	(33.3)	9	(37.5)	3	(12.5)	3	(12.5)	3	(12.5)	7	(29.2)	7	(29.2)
Lawyer	4	0	(0.0)	1	(25.0)	0	(0.0)	0	(0.0)	2	(50.0)	0	(0.0)	0	(0.0)	0	(0.0)	2	(50.0)	2	(50.0)	0	(0.0)
Total	293	62	(21.2)	47	(16.0)	33	(11.3)	14	(4.8)	70	(23.9)	71	(24.2)	11	(3.8)	67	2(2.9)	102	(34.8)	106	(36.2)	55	(18.8)
X^2 (df=3)		3.286		9.020		4.070		14.938		12.838		30.397		6.347		10.421		11.312		2.547		10.221	
P		Ns		Ns		Ns		<0.05		<0.05		<0.001		Ns		Ns		<0.05		Ns		Ns	

Ns-not significant; N-number of isolates df=degrees of freedom

Bacterial isolates recovered from user interfaces varied in their patterns of resistance to the antibiotics used (Table 5). Resistance to these antibiotics ranged from 1.1% (ciprofloxacin and ofloxacin) to 93.8% (amoxicillin).

A high percentage of isolates were also resistant to amoxicillin, augmentin, nitrofurantoin and ceftriaxone, though resistance to amoxicillin was slightly higher. *K. pneumoniae* showed its highest resistance to augmentin (92.1%) and *M. luteus* to amoxicillin (93.8%). Similarly *Proteus* spp. (91.6%) showed its highest resistance to augmentin. The least resistance of all was that of *Pseudomonas aeruginosa* to ciprofloxacin and ofloxacin (1.1%).

Multiple antibiotic resistance ranged from resistance to 2 antibiotics to 9 antibiotics (Table 6). A total of 68 multiple antibiotic resistance patterns were observed among the 595 bacterial isolates tested for their susceptibility, of which 530 (89.1%) isolates exhibited multiple antibiotic resistance. Resistance to three antibiotic groups (31.9%) was the commonest multiple resistance pattern recorded among the isolates, while resistance to eight antibiotic groups (0.2%) was the least prevalent (Table 6).

Most of the bacterial isolates of one species had the same multiple antibiotic resistance pattern, irrespective of the source and location of the sample. Resistant patterns of isolates from males and females were comparable (χ^2 =68.591, p>0.05). Among the occupational groups studied, resistant patterns of isolates were similarly found to be comparable (χ^2 =383.667, p>0.05).

Plasmid molecular weights of selected multiple antibiotic resistant isolates are shown in Table 7, while the gel electrophoretic plasmid profiles are depicted by Figures 1a and b. Plasmid profile analyses revealed the presence of one or more detectable plasmids in 73.3% of the isolates. In isolates that showed a single plasmid, its size was frequently 23.130kb.

Each of the isolates bearing multiple plasmids was resistant to at least four antibiotics. Forty percent of multiple antibiotic resistant bacterial isolates recovered from educational institutions showed a similar plasmid pattern and size. Gel electrophoresis revealed that isolates from similar sampling sites had related banding patterns (Figure 1a, lanes 4-2b, 10-12, 25-26; Figure 1b lanes 34-39). However, isolates selected from one departmental office in O.A.U. University showed greater relatedness, because they harboured a similar plasmid band profile with a band of 23.13 kb.

Table 5. Susceptibility of bacterial isolates from user interfaces to commonly used antibiotics

	Aerococcus viridans	*Bacillus* spp.	*Enterobacter aerogenes*	*Gaffkya tetragena*	*Klebsiella pneumoniae*	*Micrococcus luteus*	*Moraxella catarrhalis*	*Proteus* spp.	*Pseudomonas aeruginosa*	*Staphylococcus aureus*	*Staphylococcus epidermidis*
Augmentin	86.7%	72.7%	89.7%	55.6%	92.1%	61.5%	62.5%	91.6%	85.7%	63.4%	90.9%
Pefloxacin	1.8%	0%	0%	0%	3.1%	0%	0%	0%	1.1%	2.0%	3.9%
Tetracycline	10.0%	18.2%	24.1%	11.1%	28.6%	11.5%	0.0%	25.9%	27.4%	12.2%	0.0%
Ciprofloxacin	0.0%	0.0%	3.2%	0.0%	1.5%	0.0%	0.0%	0.0%	1.1%	0.0%	0.0%
Amoxicillin	83.6%	90.6%	83.9%	92.3%	89.2%	93.8%	90.0%	81.7%	91.4%	87.0%	70.6%
Ofloxacin	0.0%	0.0%	0.0%	0.0%	3.1%	0.0%	0.0%	0.0%	1.1%	1.0%	0.0%
Cotrimoxazole	36.4%	30.2%	38.7%	23.1%	29.2%	24.6%	40.0%	40.0%	39.8%	25.0%	9.8%
Gentamycin	7.3%	5.7%	9.7%	0.0%	0.0%	1.5%	0.0%	1.7%	6.5%	11.0%	2.0%
Nitrofurantoin	53.3%	72.7%	75.9%	55.6%	88.9%	80.8%	75.0%	79.2%	82.1%	65.9%	63.6%
Ceftriaxone	60.0%	75.5%	1.6%	23.1%	53.8%	69.2%	40.0%	45.0%	65.6%	59.0%	49.0%
Streptomycin	8.0%	2.4%	0.0%	25.0%	50.0%	5.1%	50.0%	15.4%	22.2%	11.9%	2.5%
Chloramphenicol	40.0%	19.0%	0.0%	50.0%	50.0%	30.8%	50.0%	30.8%	22.2%	20.3%	10.0%
Erythromycin	12.0%	19.0%	0.0%	25.0%	0.0%	15.4%	0.0%	15.4%	0.0%	16.9%	0.0%

Table 6. Multiple antibiotic resistance among the bacterial isolates from user interfaces

Number of Antibiotics	Antibiotic Resistance Pattern	Frequency	Total
2	AUG/AMX	35 (5.8%)	
	AMX/CHL	5 (0.8%)	
	AMX/COT	4 (0.7%)	
	AMX/CRO	77 (12.9%)	
	AMX/GEN	5 (0.8%)	
	AMX/NIT	6 (1.0%)	160 (30.2%)
	AUG/COT	1 (0.2%)	
	AUG/CRO	2 (0.3%)	
	AUG/NIT	9 (1.5%)	
	COT/CRO	14 (2.4%)	
	NIT/CRO	2 (0.3%)	
3	AMX/AUG/COT	5 (0.8%)	
	AMX/AUG/CRO	13 (2.2%)	
	AMX/AUG/NIT	54 (9.1%)	
	AMX/AUG/TET	1 (0.2%)	
	AMX/CHL/CRO	13 (2.2%)	
	AMX/CHL/STR	1 (0.2%)	
	AMX/COT/CRO	24 (4.0%)	
	AMX/COT/ERY	6 (1.0%)	
	AMX/COT/NIT	4 (0.7%)	169 (31.9%)
	AMX/CRO/ERY	3 (0.5%)	
	AMX/CRO/GEN	2 (0.3%)	
	AMX/CRO/NIT	23 (3.9%)	
	AMX/ERY/GEN	1 (0.2%)	
	AMX/NIT/TET	3 (0.5%)	
	AUG/COT/NIT	3 (0.5%)	
	AUG/CRO/NIT	7 (1.2%)	
	CHL/COT/CRO	1 (0.2%)	
	CRO/NIT/TET	5 (0.8%)	
4	AMX/AUG/COT/CRO	1(0.2%)	
	AMX/AUG/COT/NIT	6 (1.0%)	
	AMX/AUG/CRO/NIT	50 (8.4%)	
	AMX/AUG/NIT/TET	11 (1.8%)	101 (19.1%)
	AMX/CHL/COT/CRO	5 (0.3%)	
	AMX/CHL/COT/STR	10 (1.7%)	
	AMX/CHL/CRO/ ERY	2 (0.3%)	
	AMX/COT/CRO/ERY	3 (0.5%)	
	AMX/COT/CRO/GEN	3 (0.5%)	
	AMX/COT/CRO/NIT	5 (0.8%)	
	AMX/CRO/NIT/TET	1 (0.2%)	
	AUG/COT/CRO/NIT	2 (0.3%)	
	AUG/CRO/NIT/TET	2 (0.3%)	
5	AMX/AUG/COT/CPX/NIT	1 (0.2%)	
	AMX/AUG/COT/CRO/GEN	1 (0.2%)	
	AMX/AUG/COT/CRO/NIT	39 (6.6%)	
	AMX/AUG/COT/NIT/TET	9 (1.5%)	
	AMX/AUG/CRO/GEN/NIT	2 (0.3%)	
	AMX/AUG/CRO/NIT/TET	8 (1.3%)	
	AMX/AUG/GEN/NIT/TET	1 (0.2%)	76 (14.3%)
	AMX/CHL/COT/CRO/ERY	8 (1.3%)	
	AMX/CHL/COT/CRO/GEN	1 (0.2%)	
	AMX/CHL/COT/CRO/STR	2 (0.3%)	
	AMX/CHL/COT/ERY/STR	1 (0.2%)	
	AMX/CHL/CRO/ ERY/GEN	1 (0.2%)	

Table 7: Molecular Weight of Transmissible Plasmid DNA from Multiple Antibiotic Resistant Isolates from User Interfaces

Slot No.	SID	NOA	Antibiotics	NOAG	Ant. Groups	MW (in kb)
1	ED/1K	6	Ag,Am,Co,Cr,Nit,Tet	5	Pen,Sul,Cep,Nit,Tet	23.130
2	ED/1M	6	Ag,Am,Co,Cr,Nit,Tet	5	Pen,Sul,Cep,Nit,Tet	23.130
3	ED/1H	9	Ag,Px,Tet,Cx,Am,Ox,Co,Nit,Cr	6	Pen, Fq,Tet,Sul,Nit,Cep	23.130
4	ED/7K	6	Am,Cr,St,Chl,Er,Co	6	Pen,Cep,Chl,Agl,Mac,Sul	23.130
6	ED/3M	5	Ag,Am,Co,Cr,Nit	4	Pen,Sul,Cep,Nit	23.130
8	ED/1H	6	Am,Cr,St,Chl,Er,Co	6	Pen,Cep,Chl,Agl,Mac,Sul	23.130
9	ED/2K	5	Ag,Am,Co,Nit,Tet	4	Pen,Sul,Nit,Tet	23.130
10	ED/3M	5	Am,Co,Cr,Nit,Tet	4	Pen,Cep,Sul,Nit,Tet	23.130
11	ED/1H	5	Am,Cr,Chl,Co,St	5	Pen,Cep,Chl,Sul,Agl	23.130
12	ED/7K	7	Am,Cr,St,Chl,Er,Co,Gen	6	Pen,Cep,Chl,Agl,Mac,Sul	23.130
14	ED/3K	5	Ag,Am,Co,Cr,Nit	4	Pen,Sul,Cep,Nit	23.130
15	ED/3M	5	Ag,Am,Co,Cr,Nit	4	Pen,Sul,Cep,Nit	23.130
17	ED/2H	7	Am,Cr,St,Chl,Er,Co,Gen	6	Pen,Cep,Chl,Agl,Mac,Sul	23.130
18	I/1/1	6	Ag,Am,St,Chl,Er,Co	6	Pen,Chl,Ag,Mac,Sul	23.130
19	I/2/1	5	Ag,Am,Co,Nit,Tet	4	Pen,Sul,Nit,Tet	23.130
20	I/1/1	7	Ag,Am,Cr,Co,Gen,Nit,Tet	6	Pen,Cep,Sul,Mac,Nit,Tet	23.130
24	EC/O/1K	6	Ag,Am,Co,Cr,Nit,Tet	5	Pen,Sul,Cep,Nit,Tet	19.123
25	EC/O/1M	5	Ag,Am,Co,Nit,Tet	4	Pen,Sul,Nit,Tet	23.130
26	EC/O/1H	5	Ag,Am,Co,Nit,Tet	4	Pen,Sul,Nit,Tet	23.130
30	EC/O/2K	5	Am,Cr,Chl,Er,Co	5	Pen,Cep,Chl,Mac,Sul	23.130
32	EC/O/2M	2	Ag,Am,Co,Cr,Nit	4	Pen,Sul,Cep,Nit	23.130
34	EC/O/2K	5	Ag,Am,Co,Cr,Nit	4	Pen,Sul,Cep,Nit	23.130
35	EC/O/2M	6	Ag,Am,Co,Cr,Nit,Tet	5	Pen,Sul,Cep,Nit,Tet	23.130
36	EC/O/5M	6	Ag,Am,Co,Cr,Nit,Tet	5	Pen,Sul,Cep,Nit,Tet	23.130
37	EC/O/4K	5	Ag,Am,Co,Cr,Nit	4	Pen,Sul,Cep,Nit	23.130
38	EC/O/4K	5	Ag,Am,Co,Cr,Nit	4	Pen,Sul,Cep,Nit	23.130
39	EC/O/6M	5	Ag,Am,Co,Cr,Nit	4	Pen,Sul,Cep,Nit	23.130
42	EC/O/5K	5	Ag,Am,Cr,Nit,Tet	4	Pen,Cep,Nit,Tet	23.130

KEY: SID: Sample identity code; NOA: Number of antibiotics; NOAG: Number of antibiotic groups; MW: Molecular weight; kb: Kilo-basepair. Am= Amoxicillin Ag= Augmentin Chl=Chloramphenicol Co=Cotrimoxazole Cr= Ceftriaxone Er= Erythromycin Gen= Gentamicin Nit= Nitrofurantoin Ox= Ofloxacin Cx=Ciprofloxacin Px=Pefloxacin St= Streptomycin Tet= Tetracycline Pen=Penicillin Agl=Aminoglycoside Sul=Sulphonamide Mac=Macrolide Cep=Cephalosporin Fq=Fluoroquinolones

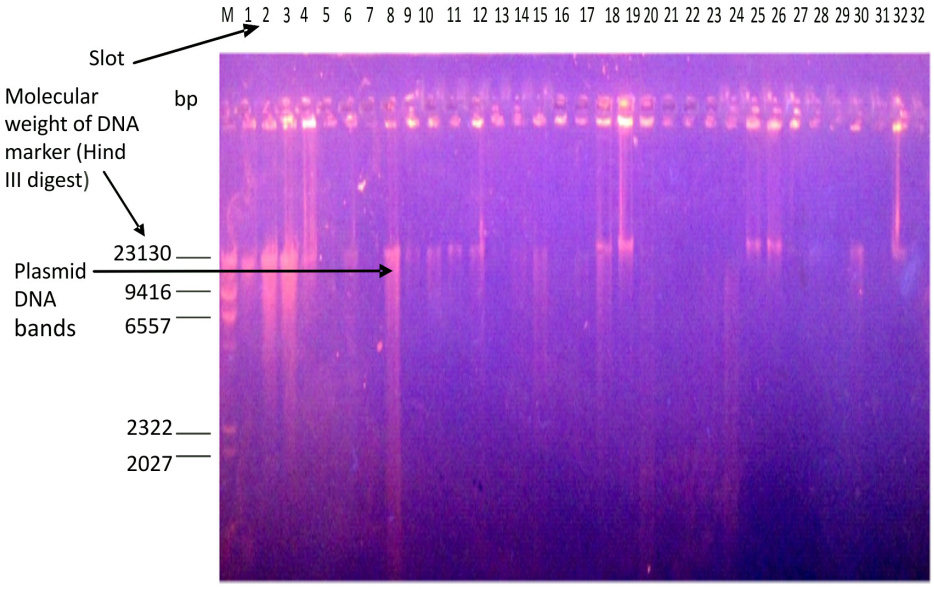

Figure 1a: Plasmid profiles of multiple antibiotic resistant isolates from user interfaces. Lane M: 23.1 kb DNA marker (Hind III digest); Lanes 1-17: bacteria isolated from keyboard, mouse and users' hands in a cybercafé, arranged in this order for each bacterial type: Lanes 1-3: *K. pneumoniae* (23.13kb), Lanes 4-6: *M. luteus* (23.13kb, lane 5 had no plasmid), Lanes 7-10: *P. aeruginosa* (lane 7 had no plasmid), Lanes 11-14: *Staphylococcus aureus* (lanes 13-14 had no plasmid), Lanes 15-17: *E. aerogenes*; Lane 18: *K. pneumoniae* from ATM (23.13kb); Lane 19: *P. aeruginosa* from ATM (23.13kb); Lane 20: *S. aureus* from ATM (no plasmid). Lanes 21-33 are bacterial isolates recovered from a selected departmental office in an educational institution.

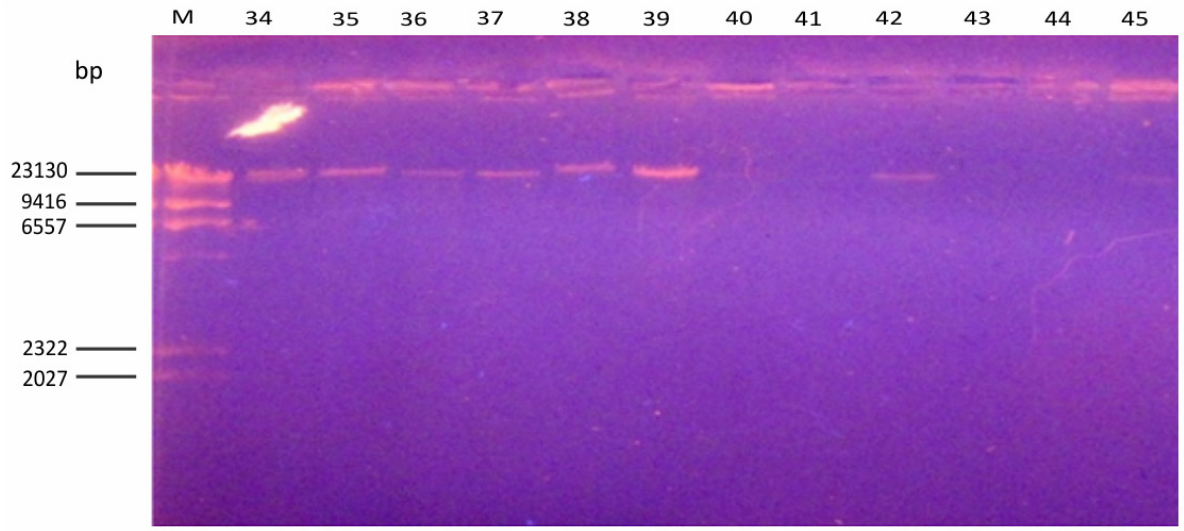

Figure1b. Lanes 34-42 are bacterial isolates recovered from selected offices in a department of Obafemi Awolowo University. Control: Lanes 43-45. Lanes 34 to 39 showed similar plasmid DNA patterns

DISCUSSION

The study revealed that high levels of bacterial contamination were detected on electronic hardware user interfaces examined in Ile-Ife. High bacterial loads were detected on interfaces in all the types of organization surveyed. ATMs and keyboards harboured more bacterial contaminants than mouse devices; this can be attributed to their structural design and large surface area, but a greater number of bacterial contaminants was recorded on ATMs than on keyboards and this may be due to the fact that ATMs are usually located in the open, exposed to wind and rain. Mouse interfaces were contaminated with lower bacterial loads than the others. This could be due to its small and smooth surface area, as a result of which the bacteria present on the mouse are exposed to conditions that may not favour their survival. Most investigations on bacterial contamination of interfaces have been centred on the keyboard and mouse, within hospital and university settings. Little or no work has been reported on bacterial contamination on ATMs or interfaces used in banks and cybercafés; therefore, the findings from this study may be pioneering in this regard. The similarity in the bacterial loads recorded on interfaces used by the various occupational groups studied can be attributed to frequent dermal contact and sharing by numerous users with differing hygiene practices and health conditions. The number of microorganisms present on a surface is amongst the microbe-associated factors that determine whether an infection will occur or not. The bacterial load on a fomite also determines the survival of bacteria on that fomite; the higher the concentration of a microorganism on a fomite the longer it survives and this invariably increases the chances of picking up the microbe from the environment (Neely & Sittig, 2002). Furthermore, Neely & Maley (2000) demonstrated that microorganisms can survive for longer on plastics, the main material of which most accessible components of user interfaces are composed, than on other surfaces such as fabrics or steel. Thus, the ability of microorganisms to survive long on plastic user interfaces suggests the possibility of their serving as reservoirs for microorganisms and as a vehicle for their transfer. Rutala & David (2004) reported similar bacterial counts on keyboards at a university health-care system.

Users' hands were more contaminated than interfaces. The magnitude of the bacterial load on hands shows that users' hands are probably a major source of bacterial contamination on the interfaces, since on a daily basis, hands typically touch a continuous sequence of surfaces, substances, objects, skin, food and body fluid.

Apart from the quantity of bacteria, the type and quality of microorganism present on a surface is also an important determinant of whether an infection will occur or not. In this study, the electronic hardware user interfaces examined were contaminated with considerable numbers of both Gram-positive and Gram-negative bacteria; however, Gram-positive bacteria were found to occur more than Gram-negative bacteria. Most skin flora bacteria are Gram-positive, which would account for their predominance on the interfaces. A total of 11 bacterial species were recovered from interfaces in this study, which included skin commensals, environmental bacteria and enteric bacteria (Table 3). The health risks associated with the majority of these bacteria are well documented (Prescott et al., 2002). The enteric bacteria encountered in this work are opportunistic human pathogens and have been associated with nosocomial infections (Ducel et al., 2002).

The bacterial contaminants cultured from electronic hardware user interfaces are similar to bacteria that have been recovered from surfaces and objects in both hospital and non-hospital settings. Other investigators have cultured similar organisms from other environmental surfaces and objects such as mobile phones (Fatma et al, 2009; Gholamreza et al., 2009), currency notes (Onibere et al., 2001), daycare centres (Itah & Ben, 2004), stethoscope covers (Michael et al., 2001) and computer keyboard and mouse interfaces (Anderson & Palombo, 2009; Eltablawy & Elhifnawi, 2009; Fraser & Girling, 2009).

Staphylococcus aureus, with 112 isolates (35.8%), was the most frequent bacterial contaminant of the electronic hardware user interfaces in Ile-Ife. This result is similar to the report by Anderson & Palombo (2009) that *S. aureus* was the commonest isolate found to contaminate keyboards in a university setting. *Staphylococcus aureus* is a major component of the normal flora of the skin and nostrils, which probably explains its high prevalence as a contaminant, as it can easily be discharged by several human activities, including sneezing, talking and contact with moist skin (Itah & Ben, 2004). It has also been associated with numerous infectious disease conditions and nosocomial infections. It follows that since users constantly touch interfaces and often sneeze, there is every chance of introducing *S. aureus* on to the interface in use. Also, airborne organisms can be transported from users or passers-by.

Pseudomonas aeruginosa, K. pneumoniae, M. luteus, Proteus spp., *A. viridans*, *Bacillus* spp. and *S. epidermidis* were other major contaminants on electronic hardware user interfaces in Ile-Ife. The presence of these organisms on electronic hardware user interfaces is a cause for some alarm, because they have been shown to possess the potential to cause infections, especially in a hospital setting (Ducel et al., 2002). In different studies, each of these organisms has been implicated either as a major contaminant or as the most prevalent pathogenic bacteria recovered (Fraser & Girling, 2009; Rutala & David, 2004).

Enterobacter aerogenes, Moraxella catarrhalis and *Gaffkya tetragena* were the least frequent bacterial contaminants on electronic hardware user interfaces in Ile-Ife. Other studies have isolated *Enterobacter aerogenes* and *Moraxella catarrhalis* from environmental objects. Fraser & Girling (2009) recovered *Moraxella* spp. from keyboards in a veterinary practice. It appears that this is the first report of *Gaffkya tetragena* being isolated from keyboard or mouse interfaces.

A high rate of contamination of user interfaces by bacteria was recorded in this study, as all interfaces sampled yielded bacterial isolates. This result was comparable to reported culture rates of over 70% in previous works (Schultz et al., 2003; Eltablawy & Elhifnawi, 2009; Fraser & Girling, 2009). Such a high level of contamination on user interfaces is worrisome because a relationship can be demonstrated between environmental contamination and the acquisition of bacteria by people (Hardy et al., 2006; Bures et al., 2001; Yuhuan et al., 2001)

Various bacterial species were found to coexist on an interface and on the hands of users. Interfaces harbor a community of bacteria with varying virulence and pathogenicity, thereby increasing the risk of infection and also the severity of infections. The conducive environment provided by interface users as a result of their unhygienic practices may account for this problem. Multiple contamination of interfaces was common to all the organizations studied, among the various occupational groups, the male and female interface users and all age groups (Table 4). Diversity of species on user interfaces was highest for interfaces used by people aged from 20 to 39 years, in educational institutions, as teachers and students. This could be related to the fact that multiple contamination is influenced by the level of personal hygiene exhibited by users, since these groups display a poor level of hygienic practice during interface usage. Multiple contamination was higher on keyboards and users' hands than on ATMs and mouse devices; the fact that keyboards are more frequently used than the other interfaces could explain the great diversity of bacteria found on them. Also, multiple contamination of hands is higher than that of the interfaces. Hands touch an array of different surfaces and objects, regularly picking up different types of bacteria from different fomites. Although the ATM had the highest contaminant loads of bacteria, the number of bacterial species was reduced; this can be attributed to the fact that users spend very little time on the ATM. Multiple contamination differs among different occupational groups and organizations; this could be attributed to differences in hygiene level among these occupational groups and organizational types. Fraser & Girling (2009) demonstrated a positive correlation between poor hygiene and high levels of bacterial contamination.

The isolation of a mixed assemblage of bacterial contaminants, especially multi-drug resistant bacteria, from public interfaces has been reported by Issmat et al., (2007), who isolated a mixed assemblage of methicillin-resistant *Staphylococcus* species from public computers in a university setting. Schultz et al., (2003) equally reported co-contamination of 95% of the keyboards examined in a Teaching hospital by skin flora and Gram-negative bacteria.

Resistance to 2 antibiotics was the commonest multiple antibiotic resistance pattern observed in bacterial isolates in this study, while resistance to a combination of 3 antibiotics was also prevalent. Infection with antibiotic-resistant bacteria has a negative impact on public health, owing to an increased incidence of treatment failure and severity of disease (Walsh and Fanning, 2008). Treatment of infections is hampered worldwide by the emergence of bacteria that are resistant to multiple antibiotics (Aleshin & Levy, 2007).

In addition, 73.3% of the multiple antibiotic resistant isolates profiled for plasmid DNA revealed the presence of such DNA. Bacterial isolates from the same sampling sites revealed similarities in the patterns of plasmid DNA isolated from them. Although resistance to multiple antibiotics can be attributed to chromosomal mutations, it is commonly associated with extrachromosomal elements (such as plasmids, transposons and integrons) acquired from other bacteria in the environment (Aleshin & Levy, 2007; Walsh & Fanning, 2008). In addition to the reports of earlier studies, the similarity in size of plasmid in isolates from the same sampling site recorded in this study suggests the possibility of the transfer of resistance among isolates. The presence of plasmids in multiple antibiotic resistant isolates raises concerns about the potential for interspecies transfer of genes conferring antibiotic resistance and could effectively increase the diversity and number of antibiotic-resistant pathogens on public interfaces.

In conclusion, user interfaces were found to be contaminated with potentially pathogenic bacteria, highly resistant to some commonly used antibiotics. These interfaces are therefore potential vehicles for the transmission of clinically important pathogens.

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