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ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF BACTERIAL CONTAMINANTS FROM AUTOMATED TELLER MACHINES IN KADUNA

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ABSTRACT

Hand contact is a major means of spreading infectious diseases. Automated teller machines are widely used in our society today by all and sundry through direct hand contact. There is great possibility of spreading drug-resistant bacteria strains unconsciously through the use of the machines. This study was aimed at isolating bacterial contaminants from some automated teller machines in Kaduna and determining their multidrug resistance pattern. Fifty automated teller machines (ATMs) were randomly selected from Ahmadu Bello Way, Kaduna. Using swab sticks, one sample was taken from the keypads of each ATM and transferred into nutrient broth and incubated overnight. Identification of bacteria isolates was done using microscopic and biochemical tests. The antibiotic sensitivity tests were carried out using the modified Kirby Bauer disc diffusion method. The total viable bacteria count from the ATMs was 1.6 x 10^3- 6.6 x 10^6cfu/ml. The following bacteria were isolated *Staphylococcus aureus* 19 (38%), *Escherichia coli* 15 (30%), *Klebsiella species* 7 (14%), *Bacillus subtilis* 6(12%) and *Proteus vulgaris* 3(6%). The antibiotics susceptibility test of the isolated organisms showed that ciprofloxacin was the most active antibiotic while the beta lactams were least active, different pattern of multidrug resistance was observed. The Multiple antibiotics resistance index (MARI) showed that 98% of isolates from ATMs had MARI greater than 0.2 indicating that the isolates originated from high risk source of contamination where antibiotics are often used. The findings of this study showed that the ATMs sampled were highly contaminated with bacteria which were resistant to commonly used antibiotics.

KEYWORDS: **Automated teller machines; Antibiotics; Resistance; Bacteria.**

INTRODUCTION

Automated Teller Machine (ATM) is a computerized telecommunication device which makes banking easier today. It is a means by which a customer of a bank can perform financial transactions without the need of a cashier, or bank teller [1]. ATMs were developed as cash dispensers and have evolved to provide many other bank-related functions such as paying routine bills, fees and taxes, updating passbooks, cash advances, cheque processing module, money transfer between linked accounts and deposit currency recognition, acceptance and recycling.

Use of ATM has a variety of benefits which include: time saving, accuracy, availability of daily 24 hours service, cash withdrawal, checking of bank balance at convenience without entering the bank, banking services like deposits, withdrawal, transfer of funds etc. can be accessed by customers from any part of the world and lots more. The ATM is likely to be contaminated with many different kinds of microorganisms both pathogenic and non-pathogenic due to their vast usage by many people in a day especially in an overcrowded environment [2].

The spread of infectious disease through hand contact has been an area of major concern in the public health care system because of the frequent contact of the hand with fomites which are potential carriers of pathogenic organism. This may lead to an alarming rate of outbreaks of infections transmitted

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by fomite. It had been shown that hard and non-porous surfaces such as ATMs keypad and door handles have the highest rate of transfer of bacterial to human hands [3]. Also lack of knowledge of the role of such surfaces in the transmission of micro-organisms and infectious disease can further lead to the spread of these microorganisms ignorantly.

ATM machine is likely to be contaminated with various microorganisms due to their vast dermal contact by multiple users [4]. Some of the pathogenic microorganism which had been isolated from ATMs include *Staphylococcus aureus, Escherichia coli, Klebsiella species, Proteus vulgaris* and *Enterobacter* species [4]. The isolated pathogenic bacteria from the family *Enterobacteriaceae* can cause hand-to-mouth infections in man if hands are not sanitized after using ATM. There is also a possibility of these pathogenic organisms causing nosocomial infections through medical personnel coming directly from the hospital and using immediately an ATM without thorough sanitation of hands before use [3].

It has also been shown that microbes once attached to hands and some surfaces may survive for a while and may be difficult to remove [5]. In fact 80% of infections are spread through hand contact with hands or other objects [6]. ATM once contaminated becomes vehicles for transmission of infection, such that the user may succeed in picking these pathogens after making use of the Automated Teller Machine, since there is no restriction as to who has access to the facility, and no guidelines to ensure hygienic usage [6]. Disease causing microbes have increasingly become resistant to most of the antibiotic commonly used today; these resistant strains can be spread through the use of ATMs. Over use or inappropriate use of antibiotics in our community may be responsible for antimicrobial resistance being experienced today [7].

The aim of this research work was to isolate bacterial contaminants from Automated Teller Machine keypads in Kaduna, Nigeria and to determine their multidrug resistance pattern.

MATERIALS AND METHOD

Sample site

In August, 2018, samples were collected from different ATMs in Ahmadu Bello area of Kaduna metropolis after taking permission from the bank managements. The banks include; Guaranty Trust Bank (GTB), Union Bank, Access, Eco Bank, Keystone, Polaris, Unity Bank, Union Bank of African (U.B.A), First Bank, and First City Monument Bank (FCMB).

Collection of sample and pre-treatment

Fifty automated teller machines (ATMs) were randomly selected from Ahmadu Bello Way, Kaduna. Using swab sticks, samples were taken from the keypads of these ATMs and transferred into nutrient broth (Titan Biotech Ltd, India) and incubated overnight.

Determination of total bacterial count

Standard plate count method was used to determine the total colony count of the bacteria in the samples [8]. A ten-fold serial dilution of the samples were performed and plated out on nutrient agar plates using the pour plate technique. The plates were incubated at 37^{o} C for 24 hours, the colonies were counted and total bacteria count was calculated. The bacteria isolates were refrigerated and kept on slants for further tests.

Isolation and identification of the bacteria

Colonies that grew on nutrient agar plates were gram stained in accordance with standard Gram staining procedure described by Cheesebrough [9] and microscopic examination, catalase test, coagulase test, urease test, indole test, triple sugar iron test were carried out [9].

Antibiotic susceptibility testing

Antibiotic susceptibility test of the isolated organisms against commonly prescribed antibiotics was carried out using Kirby-Bauer modified disc diffusion method according to CLSI guideline [10]. The standard antibiotics disc used against Gram negative bacteria isolates were: Co-trimoxazole (30µg), Chloramphenicol (30µg), Sparfloxacin (10µg), Ciproflaxacin (10µg), Amoxicillin (30µg), Amoxicillin-Clavulanate (30µg), Gentamicin (10µg), Pefloxacin (30µg), Ofloxacin (10µg), Streptomycin (30µg). The following antibiotics discs were used against Gram positive isolates: Co-trimoxazole (30µg), Ciprofloxacin (10µg), Amoxicillin (30µg), Gentamicin (10µg), Pefloxacin (10µg), Streptomycin (30µg), Ampicillin-cloxacillin (30µg), Cefuroxime (20µg), Ceftriaxone (25µg), Erythromycin (10µg).

Determination of multidrug resistance

According to European Centre for Disease Control (ECDC) and Centre for Disease Control & Prevention (CDC), Atlanta, Multidrug resistant (MDR) was defined as acquired non-susceptibility to

at least one agent in three or more antimicrobial categories [11]. Therefore all the isolates that were resistant to three or more classes of antibiotics were classified as being multidrug resistant. Multiple antibiotic resistance (MAR) index was defined as the number of antibiotics to which test isolate displayed resistance divided by total number of antibiotics to which the test organism has been evaluated for sensitivity [12]. This was calculated for each of the test isolate.

RESULTS

The number of samples collected from each bank is presented in Table 1. The average total viable bacteria count from the ATMs was 1.6 x 10^3- 6.6 x 10^6 cfu/ml. The average total viable count from each ATM is as shown in Table 2. The following bacteria were isolated from the ATMs: *Staphylococcus aureus* 19 (38.0%), *Escherichia coli* 15 (30.0%), *Klebsiella species* 7 (14.0%), *Bacillus subtilis* 6(12.0%) and *Proteus vulgaris* 3(6.0%). The bacteria isolate from each bank is presented in Table 3. The antibiotics profile showed that the isolates were generally resistant to the antibiotics used (Tables 4 and 5). In Gram-positive, the highest level of resistance was observed with Erythromycin followed by amoxicillin (Figure 1) while the Gram-negative isolates were highly resistant to ofloxacin and chloramphenicol. (Figure 2). Multidrug resistance was observed in 98% of the isolates (Tables 6 and 7). Likewise, 98% of the isolates had MAR index greater than 0.2.

DISCUSSION

The isolation of bacteria from ATM in this study confirms that ATM might serve as a mediator playing an important role in the transmission of pathogenic bacteria in the environment. They provide favorable conditions such as substrate acquired from human body and due to handling as well as dust from the environment. Most of the bacteria encountered in this study are members of the human flora. This suggests that humans are the major source of bacteria contaminant on the ATM.

The high level of contamination is possibly as a result of frequency of usage, poor hygiene status of users and environmental condition around the ATMs. These bacteria could be regarded as potential pathogens in some high-risk susceptible hosts such as older, and/or immune-compromised individuals [13]. They could be detrimental to the health of ATMs users.

Staphylococcus aureus is a major component of the normal flora of the skin and nostrils, which probably explains its high prevalence as bacteria contaminant, and it can easily be discharged by several human activities, like sneezing, talking and contact with moist skin [14]. On the other hand, *Escherichia coli,* an enteric pathogen, spread diseases through touch or improper sanitary activities of individuals. Transient pathogens are excreted in feces, various body fluids or tissues by persons infected or colonized by these pathogens. *E. coli* can cause diseases such as gastroenteritis, urinary tract infection, septicemia, dysentery, vomiting, stomach cramps and flatulence [15]. *Proteus species* are part of the Enterobacteriaceae family of Gram-negative bacilli. They are most commonly found in the human intestinal tract as part of normal human intestinal flora, along with *Escherichia coli*, of which *E. coli* is the predominant resident. *Proteus* is also found in multiple environmental habitats, including long-term care facilities and hospitals. *Proteus species* are known to cause infections such as urinary tract infections, Cystitis, Pyelonephritis and Prostatitis [16]. *Klebsiella pneumonia* is a gram-negative non-motile, rod-shaped bacterium found in the normal flora of the mouth, skin and intestines. It can cause destructive changes to human and animal lungs if aspirated especially to the alveoli resulting in bloody sputum [17].

In comparison with previous studies, the prevalence of bacteria contaminant observed in this study is a little higher than that isolated from ATM in a study in Umuaiah, Nigeria [18] and Abakaliki [19]. Furthermore, Okoro et al [20] in a study on assessment of some selected ATMs in Kaduna metropolis for bacteria contamination reported a lower prevalence of bacteria contaminants, other bacteria including *Ps. aeruginosa, Shigella dysenteria* and *Salmonella typhimurium* were also isolated [20]. Many types of pathogenic bacteria had also been isolated from computers' mice and keypads which were used in hospitals and education institutions [20]. In this study the high prevalence of *Staphylococcus aureus* and *Escherichia coli* observed compared to other organism isolated maybe due to vast dermal contact with the ATM keypads. Enteric bacteria like *Escherichia coli*, *Proteus vulgaris* and *Klebsiella* species found on the ATM keypads maybe an indication of recent fecal contaminant which can cause disease and food poisoning in humans.

Table 1: Number of samples collected from each bank

Sample sites	Number of samples collected from each ATM per bank
Guaranty Bank	7
Union Bank	4
First City Momentum Bank	5
Access Bank	6
First Bank	8
Eco Bank	5
Keystone	5
Union Bank of Africa	6
Polaris Bank	2
Unity Bank	2
Total	50 samples

Table 2: Mean total viable bacteria count from ATMs

	GTB (CFU/ML)	UNION BANK (CFU/ML)	FCMB (CFU/ML)	ACCESS (CFU/ML)	FIRST BANK (CFU/ML)	ECO (CFU/ML)	KEYSTONE (CFU/ML)	UBA (CFU/ML)	POLARIS (CFU/ML)	UNITY (CFU/ML)
1	5.18×10^4	5.1×10^3	5.1×10^4	3.9×10^4	4.9×10^5	4.9×10^3	3.5×10^3	1.75×10^3	5.9×10^6	3.5×10^3
2	2×10^4	5.5×10^5	3.2×10^3	5.9×10^3	5.9×10^4	5.3×10^4	5.9×10^3	3.0×10^3	3.5×10^4	2.7×10^4
3	2.4×10^6	1.7×10^3	2.5×10^4	4.2×10^3	3.6×10^4	5.5×10^6	3.8×10^3	2.0×10^3		
4	3×10^6	6.4×10^6	2.4×10^6	4.1×10^6	2.4×10^6	5×10^3	3.9×10^6	1.2×10^6		
5	4.2×10^3		3.9×10^4	1.6×10^3	5.5×10^3	7.3×10^3	3.0×10^6	2.7×10^3		
6	4.8×10^6			5.1×10^3	2.9×10^6			6.6×10^6		
7	5.8×10^3				1.9×10^3					
8					7.8×10^3					

Table 3: Bacteria isolated from ATM keypads from each bank

Organism	GT Bank	Union Bank	Access	FCMB	First Bank	Eco Bank	Keystone Bank	UBA	Polaris	Unity Bank
S. aureus	2	2	2	2	3	2	2	2	1	1
E.coli	2	1	2	2	2	1	1	2	1	1
Klebsiella pneumonia	1	0	2	0	2	1	0	1	0	0
Proteus vulgaris	0	1	0	0	0	0	1	1	0	0
Bacillus subtilis	2	0	0	1	1	1	1	0	0	0
Total (100%)	7(14%)	4(8%)	6(12%)	5(10%)	8(16%)	5(10%)	5(10%)	6(12%)	2(4%)	2(4%)

Table 4. Antibiotics susceptibility pattern of Gram-positive isolates

Antibiotics	*S. aureus* n = 19 (%)			*B. subtilis* (n = 6) (%)		
	S	I	R	S	I	R
Pefloxacin 10µg	12(63.2)	1(5.3)	6(31.6)	3(50.0)	1(16.7)	2(33.3)
Gentamicin 10 µg	9(47.4)	2(10.5)	8(42.1)	2(33.3)	0	4(66.7)
Ampicilli-cloxacillin 30 µg	6(31.6)	0	13(68.4)	3(50.0)	2(33.3)	1(16.7)
Cefuroxime 25µg	4(21.1)	6(31.6)	9(47.4)	1(16.7)	2(33.3)	3(50.0)
Amoxicllin 30 µg	1(5.3)	0	18(94.7)	3(50.0)	1(16.7)	2(33.3)
Ceftriaxone 25 µg	3(15.8)	9(47.4)	7(36.8)	2(33.3)	0	4(66.7)
Ciprofloxacin 10 µg	13(68.4)	6(31.6)	0	4(66.7)	2(33.3)	0
Streptomycin 30 µg	7(36.8)	2(10.5)	10(52.6)	1(16.7)	3(50.0)	2(33.3)
Cotrimoxazole 30 µg	7(36.8)	3(15.8)	9(47.4)	4(66.7)	1(16.7)	1(16.7)
Erythromycin 10 µg	1(5.3)	6(31.6)	12(63.2)	0	3(50.0)	3(50.0)

Table 5. Antibiotics susceptibility pattern of Gram negative isolates

Antibiotics	*E. coli* n = 15 (%)			*K. pneumonia* n = 7 (%)		
	S	I	R	S	I	R
Cotrimoxazole 30µg	7(46.7)	7(46.7)	1(6.7)	2(28.6)	3(42.9)	2(28.6)
Chloramphenicol 30 µg	7(46.7)	2(13.3)	6(40.0)	0	1(14.3)	6(85.7)
Sparfloxacin 10 µg	8(53.3)	1(6.7)	6(40.0)	1(14.3)	1(14.3)	5(71.4)
Amoxicillin-Clavulanate 30 µg	12(80.0)	0	3(20.0)	1(14.3)	2(28.6)	4(57.1)
Amoxicillin 30 µg	3(20.0)	1(6.7)	11(73.3)	3(42.9)	0	4(57.1)
Gentamicin 10 µg	5(33.3)	0	10(66.7)	4(57.1)	0	3(42.9)
Ciprofloxacin 10 µg	10(66.7)	1(6.7)	4(26.7)	6(85.7)	1(14.3)	0
Streptomycin 30 µg	4(26.7)	1(6.7)	10(66.7)	1(14.3)	1(14.3)	5(71.4)
Ofloxacin 10 µg	8(53.3)	2(13.3)	5(33.3)	1(14.3)	1(14.3)	6(85.7)
Pefloxacin 10 µg	2(13.3)	0	13(86.7)	3(42.9)	0	4(57.1)

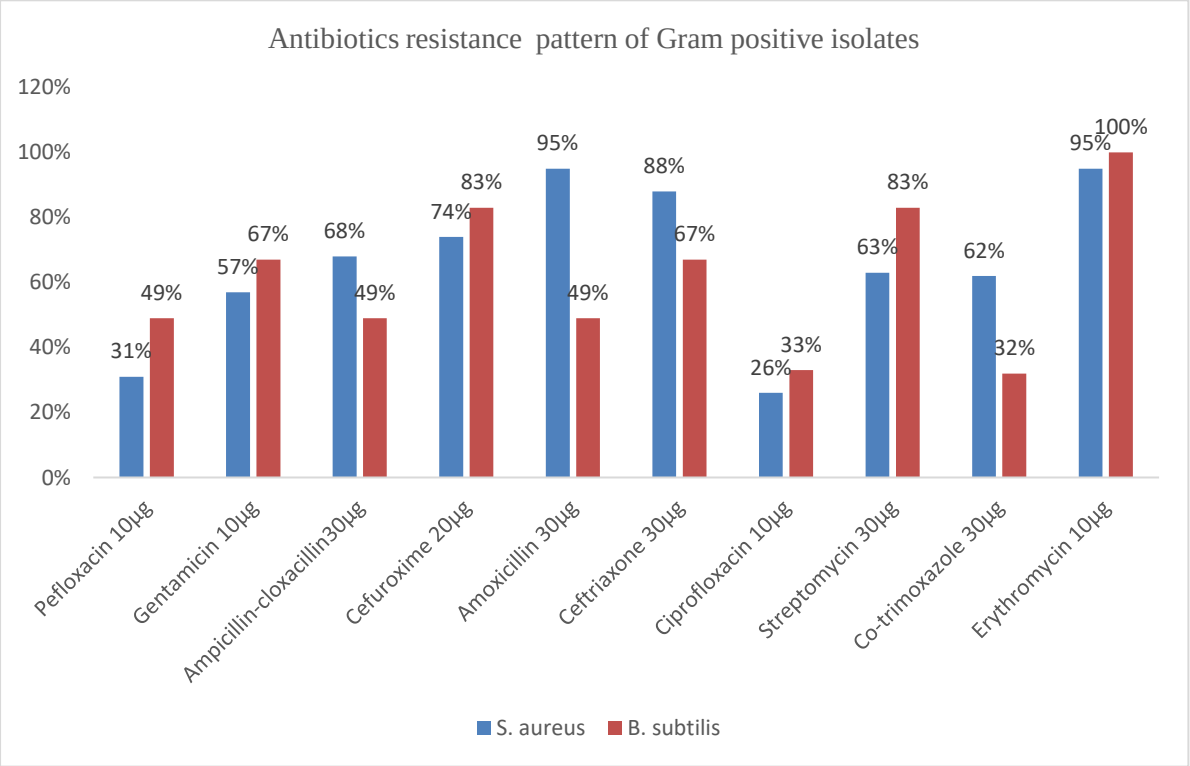

Figure 1: Resistance profile of Gram-positive bacteria isolates.

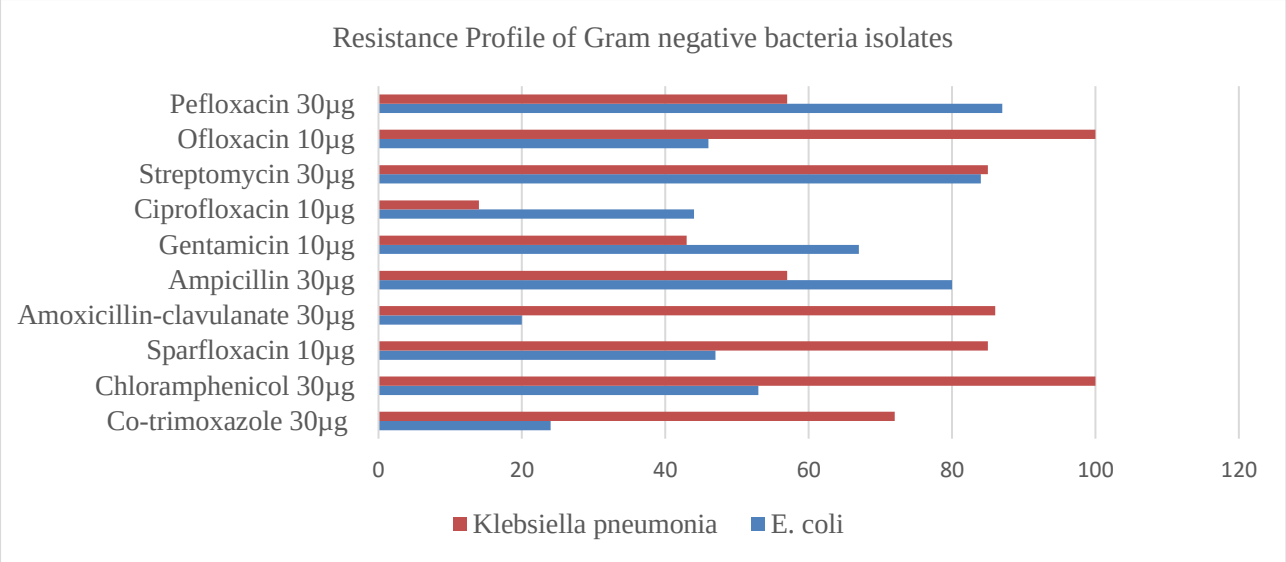

Figure 2: Resistance Profile of Gram-negative bacteria isolates.

Table 6: Multiple antibiotics resistance pattern of Gram-positive isolates.

Isolates	Code	Antibiotics resistant to	MARI	MDR
S. aureus	GT1	APX, Z, AM, R, CPX, SXT, E	0.7	MDR
	GT2	APX, Z, AM, R, E	0.5	MDR
	U1	APX, Z, AM, R, S, SXT, E	0.7	MDR
	U2	CN, APX, AM, R, SXT, E	0.6	MDR
	ACC1	CN, Z, AM, S, SXT, E	0.6	MDR
	ACC2	CN, APX, AM, R, CPX, S, SXT, E	0.8	MDR
	FC	CN, Z, AM, R, CPX, S, SXT, E	0.8	MDR
	FC2	CN, APX, Z, AM, R, SXT, E	0.7	MDR
	FB1	CN, APX, Z, AM, S, SXT, E	0.7	MDR
	FB2	CN, APX, Z, AM, R, SXT, E	0.7	MDR
	FB3	CN, APX, Z, AM, R, S, E	0.7	MDR
	ECO1	Z, AM, R, CPX, S, E	0.6	MDR
	ECO2	PEF, APX, Z, AM, R, SXT, E	0.7	MDR
	K1	PEF, CN, APX, Z, AM, R, S, SXT, E	0.9	MDR
	K2	PEF, AM, R, S, E	0.5	MDR
	UB1	R, S, SXT, E	0.4	MDR
	UB2	PEF, AM, R, CPX, E	0.5	MDR
	P	PEF, CN, APX, Z, AM, R, S, E	0.8	MDR
	U	PEF, CN, APX, Z, AM, R, S, SXT, E	0.9	MDR
Bacillus subtilis	GT1	Z, AM, R, CPX, S, E	0.6	MDR
	GT2	PEF, CN, APX, AM, R, S, SXT, E	0.8	MDR
	FC	Z, CPX, S, SXT, E	0.5	MDR
	FB	CN, Z, AM, S, E	0.5	MDR
	ECO	PEF, CN, APX, Z, R, S, E	0.7	MDR
	K	PEF, CN, APX, Z, R, S, E	0.7	MDR

KEY

SXT=Co-trimoxazole (30µg)
PEF=Pefloxacin (10µg)
AM=Amoxicillin(30µg)
APX=Ampicillin-cloxacillin (30µg)
CN=Gentamicin (10µg)
CPX=Ciprofloxacin (10µg)
S=Streptomycin (30µg)
Z=Cefuroxime (20µg);
R=Ceftriaxone (25µg); E=Erythromycin (10µg)

GT = Guaranty Trust Bank
A = Access Bank
FB = First Bank
FCMB = First City Momentum Bank; KB = Keystone Bank
ECO = Eco Bank; UBA = Union Bank of African
UB = Union Bank; P = Polaris Bank; U = Unity Bank
MARI= Multiple antibiotics resistant index
MDR: resistance to at least 3 classes of antibiotics (multiple drug resistance).

Table 7: Multiple antibiotics resistance pattern of Gram-negative isolates

Isolates	Code	Antibiotics resistant to	MARI	MDR
Klebsiella	GT	SXT, CHX, SP, AU, AM, CPX, S, OFX, PEFN	0.9	MDR
pneumonia	ACC1	SXT, CHX, SP, AU, S, OFX	0.6	MDR
	ACC2	CHX, SP, AU, AM, CN, S, OFX	0.7	MDR
	FB1	CHX, SP, AU, S, OFX, PEFN	0.6	MDR
	FB2	SXT, CHX, SP, AM, PEFN	0.5	MDR
	ECO	SXT, CHX, SP, AU, CN, S, OFX, PEFN	0.8	MDR
	UB	SXT, CHX, SP, AU, AM, CN, OFX	0.8	MDR
E. coli	GT1	SXT, AM, CN, S, PEFN	0.5	MDR
	GT2	AM, CN, PEFN	0.3	MDR
	U	SXT, AM, S, PEFN	0.4	MDR
	ACC1	SXT, CN, CPX, OFX, PEFN	0.5	MDR
	ACC2	SXT, AM, CN, OFX, PEFN	0.5	MDR
	FC1	SXT, CHX, AU, AM, S, PEFN	0.6	MDR
	FC2	SXT, CHX, SP, AM, CPX, S, OFX	0.6	MDR
	FB1	S, PEFN	0.2	No MDR
	FB2	CHX, SP, AM, CPX, S, OFX	0.6	MDR
	ECO	SP, CN, S, OFX, PEFN	0.5	MDR
	KS	CHX, AM, CN, S, PEFN	0.5	MDR
	UB1	CHX, SP, AM, CN, S, PEFN	0.6	MDR
	UB2	CHX, SP, AM, CN, CPX, OFX, PFN	0.7	MDR
	P	SXT, CHX, SP, AU, AM, CN, S, PEFN	0.8	MDR
	U1	SXT, CHX, SP, AU, AM, CN, CPX, S, OFX, PEFN	1.0	MDR
Proteus vulgaris	U2	SXT, AU, CN, CPX, S, PEFN	0.6	MDR
	K	SXT, CHX, SP, AU, CN, CPX, S, PEFN	0.8	MDR
	UBA	SXT, CHX, AU, CN, S, OFX,	0.6	MDR

KEY

SXT=Co-trimoxazole (30µg)
AM=Amoxicillin (30µg)
CN=Gentamicin (10µg)
S=Streptomycin (30µg)
PEFN=Pefloxacin (30µg)
CH=Chloramphenicol (30µg)
OFX=Ofloxacin (10µg)
S=Sparfloxacin (10µg)
AU=Amoxacillin-Clavulanate (30µg)
CPX=Ciprofloxacin (10µg)
MARI= Multiple antibiotics resistant index
MDR: resistance to at least 3 classes of antibiotics (multiple drug resistance).

The results of the antibiotics susceptibility test in this study showed that the isolates were generally resistant to the antibiotics used. The level of resistance observed was similar to that observed in the study by Nwanko and Offiah [18] on bacteria contamination of user interface of ATM of various banks in Umuahia metropolis, Abia state, Nigeria. However, Hassan *et al*., [21] in a report on antibiogram of bacteria isolated from automated teller machines in Hamadan, West Iran reported a higher level of susceptibility to gentamicin.

Considering all the antibiotics used in this study, ciprofloxacin was the most active. The difference in the activities observed might have been due to the difference in the class and mechanism of action of the various antibiotics. All the bacteria isolates were resistant to two or more antibiotics evaluated. This indicates a high level of multidrug resistance to the commonly used antibiotics. The result of the multiple antibiotics resistance index showed that 98% of bacteria isolates from ATM had MAR index greater 0.2. This suggests that the isolates originated from a high-risk source of contamination where antibiotics are often used [22]. The high MAR index of the isolates indicated a high level of antibiotics abuse in our society. Isolation of multidrug-resistant bacteria on ATM had been reported [23, 24]. This presence of MDR bacteria isolates on ATM and the potential hazards should be of great concern considering the huge number of people accessing ATM daily.

CONCLUSIONS

This study revealed that Automated Teller Machines (ATMs) sampled were highly contaminated with pathogenic bacteria which were resistant to some commonly used antibiotics. These bacteria can be transported from the contaminated surface to the humans by direct contact. As recommendation, hand washing posts should be set up very close to every ATM point, to encourage simple hand washing exercise before and after use of ATMs. Also, notices should be placed close to the machines directing users on the best hygiene practices when using the ATMs.

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