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Isolation of Microbes Associated With Automated Teller Machine (ATM) Keypads Studied at Rumuokoro Port Harcourt, Rivers State

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Authors' contributions

This work was carried out in collaboration among all authors. Author BCA designed the study, wrote the protocol and the first draft of the manuscript. Author IMI performed the statistical analysis and managed the analyses of the study. Author HOS managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

The keypads of Automated Teller Machines (ATMs) are subjected to several microbial contaminations due to their large dermal contact by numerous users and different personal hygiene practice. The study investigated the bacterial diversity and level of contamination obtainable on the ATM keypads during transactions and antibiotics susceptibility pattern of the isolates. The population of culturable bacterial isolates was determined by plating. Isolates were characterized culturally, morphologically and biochemically. Antibiotic susceptibility pattern of the isolates was determined using the disc diffusion method. The total culturable heterotrophic bacterial counts ranssged from 5.23 to 9.25 log cfu/g. The bacterial identified and frequency of occurrence is *Staphylococcus aureus* (17.5%), *Escherichia coli* (22.5%), *Bacillus* spp (17.5%), *Salmonella* spp (10.0%), *Pseudomonas aeruginosa* (10.0%), *Proteus* spp (7.5%) and *Klebsiella* spp (15.0%) respectively. *Staphylococcus aureus* were more susceptible to Chloramphenicol (37mm) and were more resistant to Rifampicin (00mm) and Levofloxacin (00mm) respectively. The Gram negative isolates in the study were susceptible to ciprofloxacin and gentamycin and more

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resistant to ceporex, nalidixic acid, septrin and ampicillin respectively. The highest gram negative isolates that showed more susceptible to all the used gram negative antibiotics were *Escherichia coli* and *Salmonella* spp while the lowest were *Klebsiella* spp and *Proteus* spp respectively. The study has revealed that bacterial contamination on ATM keypads is of health significance and could result to public health challenges if not properly managed. Therefore, adequate hand-washing hygienic practices and cleaning agents are advocated towards reducing the related ill-health among ATM users.

Keywords: Automated teller machines; contamination; antibiotics; susceptibility; keypads.

1. INTRODUCTION

Automatic teller machine (ATMs), which may be described as a cash point machine, cash machine or a cavity in a wall, is a computer supported telecommunication equipment that help the client of a financial institution to perform banking transactions from almost anywhere in the world were an ATM is present or made available [1]. People who perform bank transactions (bank workers and bank customers) have a great influence on the introduction of microorganisms unto the ATM keypad due to their individual unhygienic lifestyle such as sneezing, coughing and using the washrooms are equally route of microbial penetration as a result of inadequate hand sanitation before and after using the machine [2]. Also, these devices have become a medium for transmission of infectious diseases, once the keyboard is contaminated, the user as such may end up picking these microbes after utilizing the ATM machine [2].

Moreover, an ATM machine in the bank has no restriction as regarding to who has the ability to use the facility and also no guidelines to cover the sanitary usage. But like all surfaces, when there is lack of adequate cleaning regimen in the areas for most of these ATM equipment's, it exposes them to microbial colonization and attack which therefore, lead to infection to the users (Stephen and Kwaku, 2011). They are basically located in city centers, trade environments, campuses and within the health facilities where it can be assessed by consumers [3] Contamination of environmental inanimate objects and surfaces such as ATM machine by microbes is a normal process because they are regarded as ubiquitous in the environment [4].

Similarly, various studies of the man's environment have indicated contamination and colonization of inanimate objects like door handles, phones, money, fabrics, plastics and other fomites by microorganisms that are equally in charge for the spread of different microbial infections [5,4]. For instance, the most inanimate objects in the environment that are usually in regular contact with the hands are phones, money, keypads of an automated teller machine (ATM) [6].

Hence, microorganisms seen to contaminate fomites have been indicated to persist on the surfaces for periods ranging from a few hours to several months, and have been detected and recovered from surfaces after regular conventional sterilization (French *et al.,* 2004).

Therefore, Human hands have been reported to play a crucial role in the transmission and cross-contamination of organisms between environmental surfaces [4,7].

The capacity of non-living objects to facilitate viable microorganisms for a prolonged period of time is properly documented [6] and like environmental surfaces and objects, particularly those in close proximity with persons and constantly touched, subject human health into threat and is a cause for public concern [5,4].

Hundreds of people whose socio-economic levels and sanitary measures are quite different from each other utilized the ATM machines on a regularly bases, thereby, increasing the microbial load and disease contraction [3].

Recognizing that, 80% of infections are distributed through hand contact with contaminated hands, fomites or environment and the growth incidence of antimicrobial resistance by various pathogenic organisms which is the aim of public health management is now on disease prevention as opposed to treatment [8,4].

There is no scientific report to the best of our knowledge on the isolation and identification of bacteria from ATMs in Rumuokoro Community and its environs Rivers State, Nigeria.

To this end, this research considering that most users of the ATM are greatly ignorant of the potential hazards they face each time they utilizes an ATMs machine, this research investigates the presence of bacteria, that could be isolated from ATM keypads and public health impact.

2. MATERIALS AND METHODS

2.1 Study Area

This study was conducted within Rumuokoro Area of Port Harcourt in Rivers State. The study was undertaken from March 7th to June 20th 2021.

2.2 Sample Collection

Sixteen (16) Automated Teller Machines (ATMs) of eight (8) various banks (UBA Bank ATM 1,UBA Bank ATM 2, First Bank, Ecobank, Access Bank, Zenith Bank, Fidelity Bank, GT Bank) situated along Rumuokoro area was used for the study. The single sterile swab sticks moistened with sterile physiological saline before swabbing the buttons surfaces of the metallic keypads of ATMs. The swab sticks were labelled appropriately, kept on Ice pack medium and was transported to the Madonna University Microbiology Research Laboratory Complex, Elele, Rivers State for microbiological analysis within two hour of collection (Nworie *et al.,* 2012).

2.3 Sterilization of Materials

All the glassware's and other materials used were sterilized by autoclaving at 121^{o}C for 15 min at 15 psi or by dipping into ethanol (95% alcohol) and flaming where necessary.

2.4 Media Preparation

2.4.1 Nutrient agar

Nutrient agar (28 g) was suspended in one liter of distilled water in a conical flask mixed well and stopped with cotton wool and foil. The medium was sterilized at 121^{o}C for 15min at 15psi in an autoclave and permitted to cool at 50^{o}C before it was dispensed into plates.

2.5 Preparation of Physiological Saline

This serves as a diluents and it was prepared by dissolving 8.5 g of analytical sodium chloride in 1 litre of deionized water. This was followed by dispensing 9ml of the solution into test tubes and sterilized by autoclaving at 121°C for 15 minutes at 15 psi.

2.6 Enumeration of Bacteria

Serial dilution was carried out by transferring 1 ml of the sample into 9mls of normal saline. The dilution continued until diluents that can provide well separated colonies were established.

The spread plate technique as described by American Public Health Association [9] was used. The prepared media was poured out into sterile 9cm glass Petri dishes and left to solidify.

Duplicate plates were inoculated with 0.1ml aliquot of each dilution and spread using a flame sterilized hockey stick. Inoculated plates were incubated at 37^{o}C for 24hrs. Selected plates between 30-300 colonies were observed and recorded.

2.7 Culture Purification

Single colonies of bacteria were randomly selected from the cultured plates based on their cultural morphology and these bacteria cultures were isolated by streaking to get pure cultures. The plates were incubated at 37^{o}C for 24hrs in preparation for identification using biochemical test (Cruickshank *et al.,* 1975). (18)

2.8 Characterization and Identification of Bacterial Isolates

2.8.1 Gram stain reaction

Gram staining was done as described by Anele et al. [10] "A loopful of water was placed in a grease free sterile slide and then a portion of the organism was spread to make a smear. The smear was air dried and heat fixed. The smear was covered with crystal violet and allowed to stand for one minute, the stain was washed off and excess water was drained. The smear was covered with Gram's iodine and allowed to stand for one minute. The excess iodine was drained off and rinsed gently. 75% alcohol was also used as a decolourizer and spread on the smear until the drops coming off the slide were a pale violet colour, for 20seconds. The slide was washed gently with water. The smear was counterstained with safranin for 45 seconds. It was washed with water and the smear was allowed to blot dry. A

drop of the immersion oil was placed on the smear and the slide was viewed under the microscope at the oil immersion objective. Gram positive cells appeared purple under the microscope and Gram negative cells appeared pink or red under the microscope.

2.8.2 Biochemical test

The biochemical test that were carried out are as follows: Indole test, Sugar fermentation test, Oxidase test, Citrate test, Catalase test, Methyl Red Voges Proskauer test (MRVP), Motility test and Triple Sugar Iron test as described by Cheebrough, [11]; Nworie et al. [12]; Anele et al. [10].

2.9 Antibiotic Susceptibility Testing

The antibacterial susceptibility testing of the isolates were performed using disc diffusion method described by Bauer et al. [13] Muller-Hinton agar (Difco Laboratories GmbH, Augsburg, Germany) was prepared and poured into sterile plates. The agar medium was allowed to solidity at room temperature on a flat bench. Then some few colonies of the fresh culture of the isolates were streaked on the surfaces of the well-dried agar plates. Then multiple antibiotics disc containing (Erythromycin, Septrin, Pefloxacin, Gentamycin, Ampiciox, Amoxacillin, Rocephin, Cirpoflaxicin, Streptomycin and Zinnacef) were gently and firmly placed on the agar plates using a sterile forceps. The plates were then incubated at 35-37^{o}C for 18-24 hours. Inhibition zones observed were then measured to the nearest millimetre and recorded. Isolates were classified as resistant, intermediate or sensitive based on the definition of the Clinical and Laboratory Standard Institute and in accordance with WHO requirements Cheesbrough, [14]; Cheesbrough, [15]. An isolates was considered multi- drug resistant if it were resistant to at least three of the antibiotics tested.

2.10 Statistical Analysis

The results obtained from this study were edited, coded and subjected to different statistical investigation. Mean occurrence was determined for various samples. Analysis of variance (ANOVA) was used to determine the significance at 95% internal ANOVA.

3. RESULTS

3.1 Total Culturable Heterotrophic Bacterial Counts

The total culturable heterotrophic bacterial counts are shown in Fig. 1: Total culturable heterotrophic bacterial counts from the different bank's ATM within Rumuokoro, Port Harcourt ranged from (5.23 to 9.25) log cfu/g.

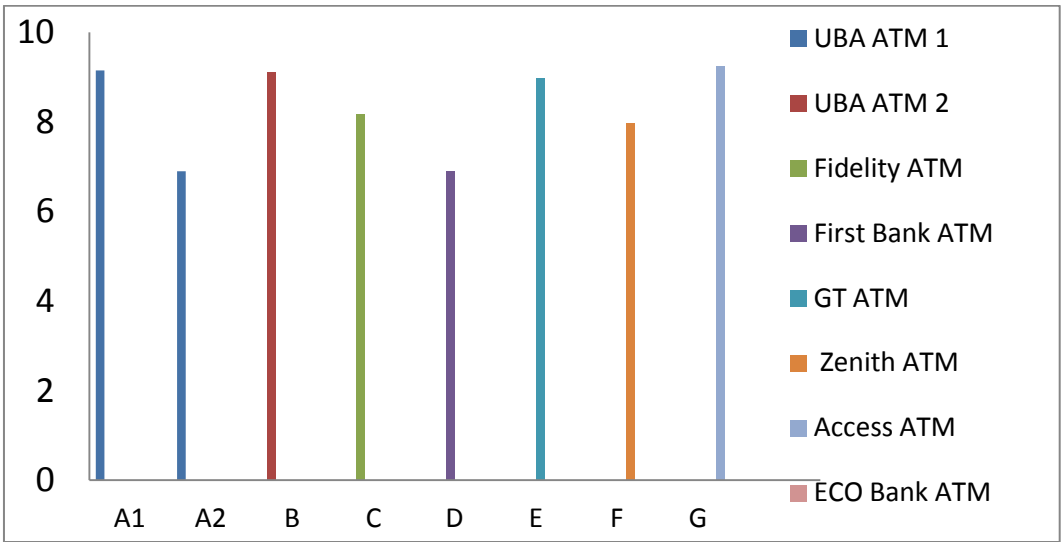

Fig. 1. Bacterial counts obtained from banks ATMs machines keypads from different points at Rumokoro Port Harcourt, Rivers State

Keys: A1= UBA, A2= UBA, B=Fidelity, C= First Bank, D= GT Bank, E= Zenith Bank, F= Eco Bank, G= Access Bank.

Table 1. Frequency of occurrence (percentage) of Different Bacterial isolates from different bank ATM keypads in Rumuokoro, Port Harcourt, Rivers state, Nigeria

Bacteria Isolates	**A1**	**A2**	**B**	**C**	**D**	**E**	**F**	**G**	**Freguency**	**Percentage (%)**
Staphylococcus aureus	3	2	0	0	0	0	2	0	7	17.5
Escherichia coli	2	3	1	1	1	0	0	1	9	22.0
Klebsiella spp	1	0	1	2	1	1	0	0	6	15.0
Salmonella spp	1	0	1	0	1	0	1	0	4	10.0
Proteus spp	0	1	1	1	0	0	0	0	3	7.5
Bacillus spp	2	1	1	1	1	1	0	0	7	17.5
Pseudomonas aeruginosa	1	0	0	0	0	1	1	1	4	10.0
Total									40	100

Keys: AI= UBA ATM, A2= UBA ATM, B=Fidelity ATM, C= First Bank ATM, D=GT Bank ATM, E= Zenith Bank ATM, F=ECO Bank ATM, G=Access Bank ATM.

Table 2. Antibiotic Sensitivity pattern of Gram Positive Bacterial Isolate obtained from different Banks ATMs machines keypads at Rumuokoro, Port Harcourt, Rivers State

Bacterial Isolate	**Antibiotics/diameter of zone of inhibition (mm)**									
	CPX	**NB**	**CN**	**AML**	**S**	**E**	**RD**	**CH**	**APX**	**LEV**
Staphylococcus aureus	12	10	32	21	36	20	0	37	22	0

Resistance range 0-13mm, Sensitive range 15mm and above

Keys: CPX= Ciprofloxacin, NB= Norfloxacin, CN=Gentamycin, AML=Amoxil, S= Streptomycin, E= Erythromycin, RD= Rifampicin, CH= Chloramphenicol, APX= Ampiclox, LEV=Levofloxacin.

Table 3. Antibiotic Sensitivity Pattern of Gram Negative Bacterial Isolate obtained from different Banks ATMs machines keypads at Rumuokoro, Port Harcourt, Rivers State

Bacterial isolates	**Antibiotic/Diameter of Zone of Inhibition (mm)**									
	OFX	**PEF**	**CPX**	**AU**	**CN**	**S**	**CEP**	**NA**	**SXT**	**PN**
E. coli	17	27	32	30	37	27	0	0	30	0
Klebsiella spp	11	13	34	6	30	7	1	0	6	0
Proteus spp	3	4	35	4	32	3	0	1	1	0
Salmonella spp	12	10	37	8	33	9	2	0	5	1
Bacillus spp	15	20	32	14	31	17	21	19	15	13
P. aeruginosa	16	27	30	21	34	19	24	16	14	12

Resistance range 0-13mm, Sensitive range 15mm and above

Keys: OFX= Tarivid, PEF= Reflacine, CPX=Ciprofloxacin, AU= Augementin, CN=Gentamycin S= Streptomycin, CEP= Ceporex, NA= Nalidixic Acid , SXT= Septrin PN= Amplicin

3.2 Frequency of Occurrence (Percentage) of Bacterial Isolates in bank ATM Keypads

The percentage frequency of occurrence of bacterial is shown in Table 1. The bacterial organisms isolated from the study and frequency of occurrence include *Staphylococcus aureus* (17.5%), *Escherichia coli* (22.5%), *Bacillus* spp. (17.5%), *Salmonella* spp (10.0%), *Pseudomonas aeruginosa*. (10.0%), *Proteus* spp. (7.5%) *Klebsiella* spp (15.0%). *Escherichia coli* were the highest occurring organisms while *Pseudomonas aeruginosa* and *Salmonella* spp. were jointly the least predominant.

3.3 Antibiotic Susceptibility Pattern of Bacterial Isolate from bank ATM from Different Location in Rumuokoro, Port Harcourt, Rivers State

Results of the antibiotic susceptibility pattern of bacterial isolates are shown in Table 2. The antibiotics used in the study include Erythromycin, Norfloxacin, Gentamycin, Ampiclox, Levofloxacin, Amoxacillin, Chloramphenicol, Cirpoflaxicin, Streptomycin and Rifampicin.

The antibiotics Chloramphenicol with (37mm) showed more activity against *Staphylococcus*

aureus than other antibiotics used for study in (Table 2) followed by Streptomycin (36mm), and Gentamycin (32mm) the lowest was Norfloxacin with (10mm). *Staphylococcus aureus* showed more resistant to levofloxacin and Rifampicin respectively.

Table 3 revealed that *P. aeruginosa* and *Salmonella* spp showed more susceptible to all the Gram negative antibiotics used in the study. More also, Gentamycin (37mm) showed more.

4. DISCUSSION

ATM machines are one of the most commonly touched surfaces today. The study assessed the bacteria contamination of shared surfaces on user hardware interface of eight (8) commercial banks randomly scattered within Rumuokoro, Port Harcourt metropolis in south-south Nigeria. A total of 40 bacterial organisms comprising of seven (7) difference species were isolated. The result obtained in (Table 1) showed that bacteria such as *Staphylococcus aureus*; *Escherichia coli*; *Klebsiella* spp; *Salmonella* spp; *Pseudomonas aeruginosa*; *Bacillus* spp and *Proteus* spp were isolated from the Automated Teller Machine (ATM) located in Rumuokoro, axis Rivers State. This result is in agreement with the result obtained by Nworie et al. [12]. Some of these isolated bacteria belong to the family of *Enterobacteriacae* which have the capacity to cause hand-to- mouth infections in man if hands are not properly sanitized after utilising the ATM machines. There is also a tendency of them causing nosocomial infections through medical personnel that used this ATM without adequate sanitization of their hands on ATM in the hospitals and its environments that exposes the users of such ATM to this dangerous hospital acquired infection [16].

The isolation of *Staphylococcus aureus* also with the second highest percentage occurrence of 7(17.5%) agrees with the finding of Nwankwo and Offiah [2]. *Staphylococcus aureus* is a major component of the normal floral of the skin and nostrils, which probably explain its high prevalence as a contaminant, as it can be easily discharged by several human activities including sneezing, talking, and contact with moist skin [17].

It has also been associated with numerous infectious disease conditions and nosocomial infections. It follows that since users constantly touch interface and often sneeze, there is every high possibility of introducing S. *aureus* on the interface in use. More also, airborne organisms can be transported from users to passer-by through hand shake [12]. Despite the fact that the lowest number (percentage) of 3(7.5%) was recorded in *Proteus* spp there is still a high possibility of infections caused by *Proteus* spp that could adversely affect the users if there is inadequate hand washing practices among them.

More so, the ATM situated near a busy road e.g. UBA also have some percentage of bacteria load, because of movement of vehicle causing disturbance of dust particles, smoke, infectious dust, droplets that increase the rate of microorganism in the air [4]. The antibiotic susceptibility result of this study in (Table 2) showed that *Staphylococcus aureus* was (0mm) percentage resistant to Rifampicin and Levofloxacin, followed by Norfloxacin (10mm) and Ciprofloxacin (12mm) while susceptible to Chloramphenicol (37mm), followed by Streptomycin (36mm), Gentamycin (32mm), Ampiclox (22mm), Erythromycin (20mm) and Amoxil (21) respectively. This is a clear indication that the antibiotics are more proactive against the isolated organism and to combat infectious diseases emulating from the isolated bacteria which is in agreement with a similar work performed by (Nwnkwo and offiah 2016). In the same vein, (Table 3) showed the Gram negative antibiotic susceptibility testing against the bacteria isolates which Ciprofloxacin showed more susceptible to *Salmonella* spp with (37mm), followed by *Proteus* spp (35mm), *Klebsiella* spp (34mm), *Bacillus* spp (32mm), *E. coli* (32) and lowest of susceptibility was P. *aeruginosa* (30mm) respectively. Also, Gentamycin equally demonstrated highest susceptibility to E. coli (37mm), followed by P.*aeruginosa* (34mm), *Salmonella* spp (33mm), *Proteus* spp (32mm) *Bacillus* spp (31mm) and *Klebsiella* spp (30mm) respectively this result is in harmony with the finding of Nworie et al. [12]. Ceporex, Nalidixic acid Septrin and Ampicillin respectively showed resistant to the tested Gram negative bacteria isolates this result affirmed the finding of Nworie et al. [12]. This simply means that the resistant antibiotics seem to be losing the battle against bacterial isolates as high resistance to all the Gram positive and negative bacterial isolates recorded in this study in (Table 2 and 3) respectively.

5. CONCLUSIONS

This study has revealed the presence of bacterial contamination on ATM keypads, with possible

health challenges. The organisms isolated were *Staphylococcus aureus*, *Escherichia coli, Klebsiella* spp, *Proteus* spp, *Salmonella* spp, *Bacillus* spp and *Pseudomonas aeruginosa*. The result of the antibiotics test showed that Chloramphenicol, Streptomycin and Gentamycin are drugs of choice for *Staphylococcus aureus*. Also, ciprofloxacin and gentamycin showed to be the drugs of choice for *Salmonella* spp, *Escherichia coli, Bacillus* spp, *Klebsiella* spp, P. *aeruginosa* and *Proteus* spp. If pathogens can be found on ATM keypads, it is easier to comprehend why there are emphases on public health safety. The need to combine technological innovation with safe and healthy use is therefore strongly advocated in the light of current findings. Therefore, this showcase a general responsibility to see that measures are adequately place to ensure that transfer of infections through ATMs is reduced to it barest minimum and if possible it should be eradicated totally.

6. RECOMMENDATIONS

1. Good hand washing and other hygienic practices should be properly observed by the users of ATMs.
2. There should be provision of bowls containing sanitizer by the bank management at every ATM locations as to promote disinfecting of users hands before and after ATM transaction.
3. Banks management should endeavour to employ ATM cleaners so that they can disinfect the metallic buttons at intervals using compatible disinfectants.

CONSENT

Approval letter of consent was obtained from the management of all the banks to use the facilities.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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