

Microbial Examination of Automated Teller Machine (ATM) in Owerri Imo State

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Abstract

This study assessed the microbial examination of Automated Teller Machine (ATM) in Owerri, Imo state. A total of five ATMs were selected from different locations within Owerri Municipal, Imo state was used for this study. Samples were collected from the key-pads and screens of the ATM devices outside banking hails with sterile cotton swabs soaked in physiologic saline. The swab stick was removed in an aseptic manner and corked properly immediately. These were transported to the laboratory inside a sealed plastic bag within 2 hours of collection. The results show the total heterotrophic bacteria count of ranged from 2.2×10^2 cfu/g - 2.9×10^2 cfu/g, the coliform bacterial counts ranged from 2.0×10^2 cfu/g - 3.1×10^2 cfu/g and total fungal count ranged from 1.1×10^2 cfu/g- 2.3×10^2 cfu/g. Bacterial isolated were *Escherichia coli*, *Pseudomonas* specie., *Klebsiella* specie, and *Staphylococcus aureus*. Prevalence of isolation of bacterial isolates shows that *Staphylococcus* specie [15(34.8)] were the prevalent isolates, followed by *E. coli* [10 (23.2)], *Pseudomonas* specie [9(20.9)] and *Klebsiella* specie [9(20.9)]. A total of four fungi were isolated. These comprises of *Aspergillus* specie., *Rhizopus* specie, *Penicillium* specie, and *Candida* specie 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* specie, *Pseudomonas aeruginosa*, *Asterius* specie., *Rhizopus* specie, *Penicillium* specie, and *Candida* specie. If pathogens can be found on ATM keypads, it is easier to comprehend why there are emphases on public health safety.

Keywords: Microbial examination; Automated teller machine Owerri

Introduction

Contamination of environmental objects and surfaces by microorganisms is a common phenomenon. The presence of viable pathogenic bacteria on inanimate objects has been reported by earlier researchers [1]. Several studies of the human environment have demonstrated contamination and colonization of inanimate objects such as door handles, plastics, faucets, phones, money, fabrics, plastics and other fomites by bacteria which is also responsible for the spread of various bacterial infections [2]. Human hands have been shown to play a significant role in the transmission and cross contamination of microorganisms between environmental surfaces [3]. Furthermore, microorganisms found to contaminate of mites have been shown to persist on the surfaces for periods ranging from a few hours to several months, and have been detected and recovered from surfaces after routine conventional cleaning. The ability of inanimate objects to support

viable microorganisms for a prolonged period of time is well documented [4] and such environmental surfaces and objects, especially those in close proximity with persons and frequently touched, poses a lot of threat to human health and is a cause for public concern. Examples of such inanimate objects in the environment that are currently in frequent contact with the hands are the keypads of an automated teller machine (ATM). Automatic teller machines (ATMs), also known as cash machines in the United States are the longest standing and most widely used form of computer driven public technology [5], with an estimated over 2.4 million units in use since their invention and use in the late 1960's, This wide usage 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 [6]. it is readily

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conceivable that pathogens could be transferred among users who share interfaces.

Materials and Methods

Sample area

Imo State is one of the 36 States of Nigeria, located in the southeast region of the country. Formed in 1976 when it split from the former East-Central State, Imo State is bordered by Abia State to the east, Delta State to the West, Anambra State to the north, and Rivers State to the south. The state capital, Owerri, is often described as the entertainment capital of Nigeria. Imo State is a predominantly Igbo speaking state, with Igbo people constituting an estimated 98% of the state's population. During the Nigerian Civil War (1967-1970), the present-day borders of Imo State were part of the Republic of Biafra, a secessionist state formed by Igbo nationalists. Secessionist sentiment remains commonplace in modern Imo State, with the Movement for the Actualization of the Sovereign State of Biafra (MASSOB) being headquartered in the district of Okigwe.

Sample Size

A total of live ATMs was selected from different locations in Imo State.

Ethical Approval

Written permission was obtained from five (5) banking organizations including Zenith, United Bank of Africa, First Bank of Nigeria, Access Bank and Stanbic Bank.

Eligibility of Subject

Inclusion Criteria

Samples were collected from the key-pads and screens of the ATM devices outside banking halls with sterile cotton swabs soaked in physiologic saline. The swab stick was removed in an aseptic manner and corked properly immediately. These were transported to the laboratory inside a sealed plastic bag within 2 hours of collection. All samples were transported to the laboratory as soon as possible without delay and were processed on the same day of collection.

Media preparation

All the media used were prepared according to the manufacturer's guide. Mannitol Salt Agar (MSA), MacConkey and Nutrient, agar plates were used for the isolation of bacteria and Sabouraud dextrose agar was used for isolation of fungi.

Identification of The Bacteria Isolates

Identification of pure colonies using morphological characteristics were based on morphological differences, colonies were isolated

from their axenic culture. Petri-dishes were divided into quadrants and sub-culturing carried out by streaking. Colony morphology observations formed a major identifying criterion for bacteria. The characteristics observed include (circular, irregular, spreading), elevation (flat, slightly raised or markedly raised), pigmentation (red, white, pink, colorless), size (pinpoint, small, medium, large) and texture.

Gram Staining

Culture plates of bacteria, microscopes, wire loops, normal saline, Bunsen burners, timers, microscope slides, oil immersion, distilled water, crystal violet, and Iodine. On a clean slide, a drop of sterile saline was added using a sterilized wire loop. The wire loop was flamed to red hot and allowed to cool, and then, a small number of cells from an isolated colony 24 hours on a culture plate was collected and mixed in a drop of saline and it was spread on the slide to make a thin, uniform smear. The smear was then allowed to air dry before they were heat-fixed by carefully passing them over a Bunsen Burner flame six to eight times. Crystal violet was used to stain the smear for one minute, then it was washed off with distilled water and drained, then Lugol's iodine was added and was left on the smear for 1 minute, which was washed off gently with distilled water and drained. The slides were decolorized with acetone for 3 seconds by adding uniformly and quickly to the slide, it was washed off quickly with distilled water and then saffranin was added and after 1 minute it was washed off with distilled water. The slides were air dried and immersion oil was dropped on the smears and examined microscopically using x100 oil immersion objective of the microscope.

Identification of Bacteria Using Biochemical Methods

Indole Test

Kovac's reagent, peptone water, incubator, wire loop. The test organism was inoculated in sterilized tube containing peptone water broth, the solution was incubated aseptically at 37°C for 24-28 hours, then 0.5ml of Kovac's reagent was added to the broth. The result was noted.

Methyl Red Test

MR-VP medium (glucose broth), methyl red indicator, wire loop, incubator. By using a sterile wire loop, (The test organism was inoculated into the fresh, sterile prepared MR-VP medium and it was incubated at 37°C for 2-5 days. After incubation the broth was obtained from the incubator, 5 drops of 0.04% solution of alcoholic methyl red solution were added and mixed properly. Then the result was read and noted,

Voges- Proskauer Test

MR-VP broth, alpha naphthol, 40% KOH, deionized water. A tube containing the MR/VP broth was inoculated with the pure culture of (he tests organism and was incubated at 35°C at 24 hours. Then 1.5mL of 5% alpha naphthol and 0.5mL of 40% KOH was added and mixed properly. The bottles were left to stand for 5 minutes for aeration. Changes were observed.

Oxidase Test

Oxidase reagent, bacterial cultures, Whatman No 2 filter paper, glass rod. A piece of filter paper was placed in a clean Petri-dish and 2-3 drops of freshly prepared oxidase reagents was added. Using a sterile loop, test bacterial was picked and smeared over a small area of the filler paper. The color change was examined after 10 seconds.

Catalase Test

Bacterial culture, H₂O₂, glass slide, test tubes, glass rod. A drop of 3% H₂O₂ was placed on the opposite ends of a clean grease free glass slide with the help of a dropper. A small portion of (her bacterial culture was transferred on the glass slide containing the H₂O₂ with a sterile glass rod. The sample was examined for immediate bubbling effervescence of O₂.

Identification of Fungi Isolates

The growth pattern, pigmentation and size of colonies were recorded at the incubation period to aid identification of the organisms.

Use Slide Culture Method

The isolate was identified using cultural characteristics and morphology. With the help of sterile petri dishes, sterile filter paper was placed in each of the Petri dishes and 1ml of distilled water was added into the petri dish as sterile U shape glass rod was placed in each of the petri dish. Will) the help of inoculating needle, a cubelike shape of already prepared SDA was cut, four days to 1-week fungal growth was smeared by the four sides of the SDA using a sterile wire loop and a sterile cover slip was placed on the inoculums in the Petri dish. The plate was then covered and kept at room temperature for 4 to 7days as growth was observed daily before examination.

Cell Morphology of Fungi Isolate

A drop of lactophenol (LP) was placed on a clean microscopic slide The cover slip from the glass culture was gently removed and placed in the drop of lactophenol (LP) and also a drop of LP was dropped on the slide from the old culture as the media cultured on the slide was gently removed a sterile cover slip was placed on the slide and observe microscopically. It was first view at X10 to focus the lens well then X40 to get a clearer view.

Results

Table 1 shows the bacterial counts of each sample. The total heterotrophic bacteria count of ranged from 2.2×10^2 cfu/g - 2.9×10^2 cfu/g, the coliform bacterial counts ranged from 2.0×10^2 cfu/g - 3.1×10^2 cfu/g and total fungal count ranged from 1.1×10^2 cfu/g - 2.3×10^2 cfu/g.

Table 1: Total Viable Consult of Bacteria Isolates.

| Total Viable Count (CFU/g) | | | | |
|----------------------------|--------|-------------------|-------------------|-------------------|
| SN | Sample | THC | TCC | TFC |
| 1 | A | 2.2×10^2 | 2.8×10^2 | 1.1×10^2 |
| 2 | B | 2.6×10^2 | 2.5×10^2 | 2.0×10^2 |
| 3 | C | 2.9×10^2 | 3.0×10^2 | 1.8×10^2 |
| 4 | D | 2.8×10^2 | 2.0×10^2 | 1.6×10^2 |
| 5 | E | 2.4×10^2 | 3.1×10^2 | 2.3×10^2 |

Table 2: Morphological and Biochemical identification of bacteria isolates.

| S/N | Morphology | Gramstaining | Catalase | Citrate | Indole | Methylred | Voges proskaur | Oxidase | Motility | Suspected organism |
|-----|--|--------------|----------|---------|--------|-----------|----------------|---------|----------|------------------------------|
| 1 | Yellor, Glassy, Round, Coccie in cluster | + | + | + | - | + | + | + | - | <i>Staphylococcus specie</i> |
| 2 | Greenish, Opague, Flat and Odour colony | - | + | + | - | - | - | + | + | <i>Pseudomonas specie</i> |



| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|--------------------------|
| 3 | Cream, Smoot, Irregular, Short rod in single colony | - | + | + | - | - | + | - | - | <i>Klebsiella specie</i> |
| 4 | Cream, Smooth, Irregular, Short rod | - | + | - | - | + | - | - | - | <i>Escherichia coli</i> |

Table 3: Prevalence of Isolation of Bacterial Pathogens.

| Isolates | Frequency (%) |
|------------------------------|---------------|
| <i>Escherichia coli</i> | 10(23.2) |
| <i>Staphylococcus specie</i> | 15 (34.8) |
| <i>Pseudomonas specie</i> | 9 (20.9) |
| <i>Klebsiella specie</i> | 9(20.9) |
| Total | 43 (100.0) |

Table 4: Colonial and Morphological Features of the Fungi Isolates from A.T.M.

| S/N | Morphological characteristics | Microscopic characteristics | Probable organism |
|-----|---|--|---------------------------|
| 1 | Compact, clusters of dark colony | Hyline conidiophores phylides borne on vesicles, clusters of dark walled conidia with spetate hyphae | <i>Aspergillus specie</i> |
| 2 | Cotton-like, white in color | Simple and form apical, globular sporangia that are supported and elevated by a column shapped columella | <i>Rhizopus specie</i> |
| 3 | The pigmentation was initially white and became green which later turned to pinkish with time. they were filamentous in texture | Spetate hyphae with simple a conidiospores and conidia were observed | <i>Penicillium specie</i> |
| 4 | Creamy colonies that are smooth, convex and with a yeasty odour | Budding spherical to elongated cells forming pesudmycelium | <i>Candida specie</i> |

Table 5: Number and percentage of fungi species isolated from A.T.M.

| Isolates | Number of Isolate | Percentage |
|---------------------------|-------------------|------------|
| <i>Aspergillus specie</i> | 15 | 40.5% |
| <i>Candida specie</i> | 10 | 27.0% |
| <i>Penicillium specie</i> | 7 | 18.9% |
| <i>Rhizopus specie</i> | 5 | 13.5% |
| Total | 37 | 100% |

Keys

THBO - Total Heterotrophic Bacteria count

TCC =Total Coliform count

TSS = Total Salmonella Shigella

Sample A – Zenith bank

Sample B = UIJA

Sample C = First Bank of Nigeria

Sample D = Access Bunk

Sample E, - Stannic Bank

Morphological and Biochemical identification of bacteria isolates

Results from Table 2 shows the morphological appearances and biochemical properties of isolated bacteria. Bacterial isolated were *Escherichia coli*, *Pseudomonas specie*, *Klebsiella specie*, and *Staphylococcus specie*.

Prevalence of isolation of bacterial pathogens

Table 3 shows the prevalence of isolation of bacterial isolates from A.T.M. Of the 43 bacterial isolates obtained, *Staphylococcus* specie [15(34.8)] were the prevalent: isolates, followed by *K. coll* [10 (23.2)], *Pscitcloinonas* specie [9(20.9)] and *Klebsiella* specie [9(20.9)]

Colloidal and Morphological features of the Fungi Isolates from A.T.M

Results as shown in table 4 shows the Colonial and microscopic morphology of isolated fungi associated with A.T.M. A total of four fungi were isolated. These comprises of *Aspergillus* specie, *Rhizopus* specie, *Penicillium* specie, and *Candida* specie.

Number and percentage of fungi species isolated from A.T.M

Table 5 shows the number and percentage of fungi species isolated from A.T.M, which revealed that *Aspergillus* sp. was the most common isolate (1-0.5%), followed by *Candida* specie. (27.0%), *Penicillium* specie. (18.9%), and *Rhizopus* specie (13.5%).

Discussion

ATM machines are one of the most commonly touched surfaces today. The study assessed the microbial contamination of shared surfaces on user hardware interface of five (5) commercial banks randomly scattered within Owerri, Imo state metropolis in south-east Nigeria. A total of 43 bacterial organisms comprising of four (4) different species were isolated. The result obtained in (Table 2) showed that bacteria such as *Staphylococcus aureus*; *Escherichia coli*; *Klebsiella* specie; and *Pseudomonas aeruginosa*, were isolated from the Automated Teller Machine (ATM) located in Owerri, axis Imo State. This result is in agreement with the result obtained by [7,8]. The bacteria isolated from (his study can cause hand-to-mouth infections in man if hands are not sanitized after using the ATM. There is also a possibility of them causing nosocomial infections through medical personnel that used an ATM without thorough sanitation of hands used on ATM in the hospital and its environs as also reported by [9]. These findings are in agreement with the results obtained by [10].

The higher prevalence of *Staphylococcus aureus* (34.8%) obtained might probably be due to high concentration of ATM users. *S. aureus* is a normal flora of the skins. Although *S. aureus* is not always pathogenic, it is a common cause of skin infections such as abscesses, respiratory infections such as sinusitis and food poisoning. In addition, it can result to opportunistic infections. The *Klebsiella pneumonia* isolated could possibly expose the users of the ATMs in the study area to pneumonia if good hygiene practices (especially hand washing after using ATMs) are not adhered to by the users [4]. The statistically significant

relationship between the bacterial isolates and the respective locations could be as a result of several factors such as, poor hygiene practices by ATM users, rainfall, wind and high concentration of users at ATM galleries. Similarly, it has been reported that the keypads of ATMs harbored more bacteria than the computer keypads and this may be due to the fact that, the keypads are exposed to many users, environmental factors such as rain and climatic factors such as wind [11]. The result of this study is also similar with the results of bacterial contamination from obtained the surfaces of the metallic keypads of the ATMs located within Abakaliki metropolis [12]. The abundance of *E. coli* an enteric bacterium is indicative of possible fecal contamination which could also be a pointer also to poor hygienic practices by ATM users [13]. The results of CITJ fungal enumeration is demonstrated in Table 4 Data about fungal contamination in ATMs were described in a few studies. The fungal contamination in this study included *Aspergillus* specie, *Rhizopus* specie, *Penicillium* specie and *Candida* specie. The *Aspergillus* genera were the most dominant followed by *Candida* specie (27.0%), *Penicillium* specie (18.9%), and *Rhizopus* specie (13.5%). *Aspergillus* specie, are widespread in nature because they produce many spores, have wide ecological tolerance, can easily spread through the atmosphere, and they can contaminate almost any kind of surface [14]. *Penicillium* species are common contaminants on various substrates and are known as potential mycotoxin producers. Opportunistic infections leading to mycotic keratitis, otitis media and endocarditis have been reported. Its spores and components in atmosphere entering from respiratory tract affect the human health and may cause allergic reactions. *Rhizopus* species is one of the factors of opportunistic fungal infection, as skin and mucous membranes allergies [15].

Conclusion

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* specie, *Pseudomonas aeruginosa*, *Aspergillus* spp., *Rhizopus* specie, *Penicillium* specie, and *Candida* specie. 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 its barest minimum and if possible, it should be eradicated totally.

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