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COMPARATIVE STUDY ON THE LEVEL OF BACTERIOLOGICAL CONTAMINATION OF AUTOMATIC TELLER MACHINES, PUBLIC TOILETS AND PUBLIC TRANSPORT COMMERCIAL MOTORCYCLE CRASH HELMETS IN KIGALI CITY, RWANDA

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ABSTRACT

**Background:** The environments can be contaminated by infectious agents that constitute a major health hazards as sources of community and hospital-acquired infections due to various activities.

**Objective:** A comparative study on the level of bacteriological contamination of automatic teller machines (ATMs), public toilets and commercial motorcycle crash helmets were conducted in Kigali city during the period of January to March, 2013.

**Design:** Samples were collected from selected ATMs, public toilets and commercial motorcycle crash helmets surfaces. Micro-organisms identified from these samples were associated to infecting organisms recovered from unwashed hands surfaces and recorded results in the nearby hospital.

**Setting:** Samples from each device and subject were transported to the laboratory where they were analysed for the presence of coliforms and other airborne, human skin and intestinal disease causing microorganisms. Microbiological methods including spread plate techniques and some biochemical tests were used to partially identify the microorganisms.

**Subjects:** Subjects involved in this study were consented students from University of Rwanda and Kigali motorcyclists for collections of samples from hands and crash helmets respectively.

**Results:** The following pathogenic bacteria have been found on the devices, *Staphylococcus aureus*, *Staphylococcus epidermis*, *Streptococcus species*, *Escherichia coli*, *Salmonella*, *Klebsiella*, *Enterobacter aerogenes*, *Pseudomonas*. The commercial motorcycle crash helmets had the highest level of bacteriological contamination compared to ATMs and public toilets. There was no growth observed on samples collected after treatment from ATMs, public toilets, and commercial motorcycle crash helmets. Attempt to correlate this finding with infecting organisms recovered from unwashed hands surfaces and recorded results in the nearby hospital show that the presences of some of these infectious pathogens.

**Conclusion:** This study has revealed the ability of these public devices to serve as vehicle of transmission of microorganisms with serious health implications. To improve and ensure the safety of these public devices the use of disinfectants is of high importance on reducing bacteriological load on those public devices. Proper cleaning regimen to sanitise these facilities regularly and public education on their hygienic usage are recommended to reduce the associated risks.

## INTRODUCTION

Micro-organisms are found everywhere and constitute a major part of every ecosystem. In these environments, they live either freely or as parasites (1). In some cases, they live as transient contaminants in fomites or hands where they constitute a major health hazards as sources of community and hospital-acquired infections (2).

ATM machine can be contaminated by user breathe out, sneezing, digging nose during work by hands or other actions that leaves the key boards contaminated. The nasal droplets, mouths water or mucus from mouth and nose, nail cavities, bank notes when handled with contaminated hands and air-borne microorganisms brought by winds and settle on ATMs (3). ATM represents a good transient environment for development of pathogenic microbes especially *Salmonella*, *Escherichia coli* (4). The faecal coliform group includes genera such as *Enterobacter* and *Klebsiella* which are commonly associated with public devices (5). ATMs should be cleaned properly, as they are operated by all sections of people frequently. If these are not cleaned properly, they become source of potentially pathogenic microorganisms in people (6).

Research has shown that public toilets are vital components for breeding places for disease-causing bacteria (7). Sanitary conditions in public places have always been a major problem, especially rest rooms. Health departments are continually checking the cleanliness and safety of these bacterial breeding grounds to prevent the spread of sickness and disease. Barker and Bloomfield in 2000 (8) found that in four out of six toilets tested, *Salmonella* bacteria existed. Public toilets represent a suitable environment for development of pathogenic microbes' especially faecal coliform group such as *Enterobacter*, *Klebsiella* and enteropathogens such as *Salmonella enteritidis*, and *Escherichia coli* (8). It was found that ATMs have similar levels of *Pseudomonads* and *Bacillus*, bacteria known to cause sickness and diarrhea, as in public toilets (9).

Transportation is vital to every human community. One means of transportation in most part of the world is the motorcycle. Motorcycles are a popular means of public transportation especially for short distances in Kigali city. Its frequent use and patronage is very high because it is faster, convenient and can easily maneuver through the regular traffic jam and get to any destination provided there is road (10). However, motorcycle crash helmet could constitute a health hazard to the users. Constant handling and use of MCHs by different individuals could create a prime condition for proliferation of many microorganisms such as bacteria and mildews (11), and a possible transmission of pathogenic microorganisms as well

as communicable diseases among users.

The human skin is constantly in contact with environmental microorganisms and become readily colonised by certain microbial species. The adult human skin supports about 10 cfu/ml bacteria (12). The normal microbiota of the skin include among 12 others, coagulase negative *Staphylococcus*, *Diphtheroides*, *Staphylococcus aureus*, *Streptococcus (various species)*, *Bacillus spp.*, *Mallassezia furfur*, *Candida spp.* and occasionally, *Mycobacterium spp.* are found on the skin (11). However, this normal microbiota can produce disease condition if introduced into foreign locations or compromised hosts (13).

ATMs, public toilet and commercial motorcycle crash helmets are fomites which are used by many people. It is necessary to investigate the possibility of the transmission of infectious agents through the use of these often used materials. It is in this context a comparative study on the levels of bacterial contamination of highly used ATM machines, public toilets and commercial motorcycle crash helmets in selected areas located in Kigali city has been undertaken. The effects of cleaning and disinfecting ATM machines, public toilets and commercial motorcycle crash helmets on the microbial load has also been studied. Findings were correlated with infecting organisms of body surfaces and recorded results of the nearby hospitals which perhaps may provide interesting information relevance to the community.

## MATERIALS AND METHODS

### 1. Collection of samples from toilets, ATMs and CMHs

*Experimental design:* Kigali is a province-level city governed by a city council. The city is split into three administrative districts: Gasabo, Kicukiro, and Nyarugenge (14). From the three districts the Nyarugenge was selected by using a lottery system of randomisation. From the selected district again sectors were selected by using the same system. Samples were collected from ATMs selected at interval of 5 ATMs within the selected sectors. Samples were collected from existing public toilets within the selected sectors. CMHs samples were collected from motorbike cyclists at parking stations within the selected sectors. From those who consented to provide samples the lottery system was used to select for sample collection. In this study, a total number of 120 samples were collected. Distribution of the samples were as follows; 40 (20 before and another 20 after application of disinfectants) from ATMs, 40 (20 before and 20 after) from public toilets and 40 (20 before and 20 after) from CMHs.

*Sample procedure:* Samples were collected by adopting the cotton swab rinse technique (15, 16 and 17).

Packages of cotton swab and glass tubes were sterilized by autoclaving and allowed to dry before use. To maintain aseptic technique, personnel wore sterile surgical gloves. Cotton swabs were moistened with 5 ml of sterile water before use. From the same surface but different locations one sample was collected before applying disinfectant and another after applying disinfectant. The sample surface was wiped with the cotton swab in order to pick up microorganisms. In order to effectively remove pathogens cotton swab on surface was covered a second and third time with the wiping motion rotated 90°. Each cotton swab was placed into a dry sterile glass tube container. Disinfectants that have been used were seventy percent alcohol based disinfectants there were commonly found in every super market around Kigali (produced and supplied by KIPHARMA S.A.R.L., Kigali, Rwanda). Samples were transported to CHUK laboratory and processed within 1 hr. Two well oriented personnel were involved in the process of sample collection.

*Culture media and biochemical test:* From each sample, serial dilution was made to reduce the microorganism's concentration in the sample. Serial dilution was carried out using sterile saline water as diluents, 10 ml was aseptically measured and added in the cotton swab applicator and shaken thoroughly and a one ten dilution of that solution was prepared by adding 1 ml of that solution to 9 ml of sterile saline water with a sterile pipette and thoroughly mixed.

For counts of total viable bacteria, Gram positive bacteria (*Staphylococcus*), Gram negative (*coliforms*); from each dilution 0.1 ml was plated using the pour plate method. Plating was done in duplicates on the following culture media: Nutrient agar (NA) for total viable count, Blood agar (BA) for *Streptococcus*, Mannitol salt agar (MSA) for *Staphylococcus aureus*, MacConkey agar (MCA) for coliforms, *Salmonella-Shigella* agar (SS) for *Salmonella* and *Shigella* count. NA, MSA, BA, SS and MCA inoculated plates were incubated for 24 hours at 37°C.

After incubation, colonies that developed on the plates were counted using a colony counter. The number of colonies per unit volume of the used quantity is the colony forming unit (CFU) of the sample was thereafter calculated for each sample. Bacteria isolates were identified based on morphological characteristics using Gram staining and viewing under the light microscope and a series of biochemical tests: Citrate test, Urease test, Coagulase test, Catalase test, Malonate test, Indole production, Triple Sugar Iron (TSI) test and Methyl red test. Standard biochemical test procedure was followed (18).

## 2. Enumeration, isolation and identification of bacteria from humans hands

*Sample collection and culture media:* Out of 31 fourth year Applied Biology students at the University of Rwanda 25(81%) of them consented to provide samples. Samples were collected from unwashed hands paying special attention to the backs of hands, wrists, inbetween fingers and underneath fingernails, during February 2014. The process of collection the samples from the body surfaces was the same as mentioned above by applying the cotton swab rinse method. A laboratory technician was involved for sample collection and carrying out the laboratory analysis.

After proper labeling and serial dilution as outlined above a total of 25 samples were then streaked on nutrient agar (NA), MacConkey agar (MA), Mannitol salt agar (MSA), Eosin methyl blue (EMB) and Selenite Cysteine Agar (SCA).

Those plates were incubated aerobically at 37°C for 24-48 hours. Pure cultures of bacterial isolated were characterized based on morphology and biochemical tests. Finally Gram-positive and Gram-negative bacteria were identified by microbiological procedures. Bacterial colonies were differentiated based on the colony morphology and color, by Gram staining procedures. Suitable biochemical tests were done for further identification of the bacterial isolates.

*Characteristics of colonies from different media:* Nutrient Agar (NA) was a medium used for total viable bacteria count of both Gram-positive and Gram-negative bacteria. MacConkey agar (MA) medium was differential and selective for Gram-negative bacteria. The Mannitol salt agar (MSA) was selective and differential for Gram-positive bacteria, especially for *Staphylococci*. Eosin methyl blue (EMB) medium was differential for lactose and non-lactose fermenting bacteria facilitate the isolation of Gram-negative enteric bacteria. Selenite Cysteine Agar (SCA) medium was selective for Gram-negative bacteria.

*Gram staining:* Gram staining was done by picking separate colonies from NA culture media and then Spread it on slides. A thin smear was made using a wire loop, fixed it on air and over the flame.

During staining, firstly, a drop of crystal violet was poured on smear and waited for one minute then it was washed with tap water. Secondly, iodine was poured and washed after waiting for one minute. Thirdly, alcohol was applied and washed immediately. Finally, a counter red stain (safranin) was poured, waited for one minute, then it was washed with sterile water, dried on air and observed under microscope using oil immersion.



**Inoculation of colonies for biochemical tests:** Aerobic bacteria from nutrient agar white and yellow colonies suspected to be either Gram positive or Gram negative were picked separately and inoculated on the following biochemical reagents: Simmons citrate agar, malonate broth, urease broth, Triple sugar iron agar and Methyl red-Voges Prausker reagent, incubated at 37°C for 24 hours. Colonies from NA were also inoculated into catalase test and coagulase test by using hydrogen peroxide and rabbit plasma respectively. Staphylococcal species from Mannitol salt agar: A colony was picked from MSA plates and used for catalase and coagulase tests using hydrogen peroxide and rabbit plasma respectively. Gram negative bacteria from Mcconkey agar: Pink and yellow colonies were picked separately and inoculated into test tubes containing the following biochemical reagents: Simmons citrate agar, Malonate broth, urease broth, Triple sugar iron agar and Methyl red-Voges Prausker reagent (MR-VP), incubated at 37°C for 24 hours. Detailed procedure for each biochemical test was followed the method described by McFadden, 2000 (18).

### 3. Retrospective bacteriological examination data

Retrospective bacteriological examination results were obtained from Kigali Kanombe Hospital Laboratory in order to correlate the present finding with infecting organisms recorded in the nearby hospital.

## RESULTS

### 1. Results for toilets, ATMs and CMHs samples

**Bacterial count results:** The bacteriological counts of both untreated and disinfectant treated ATMs, public toilets and commercial motorcycle crash helmet samples are summarised in Table 1. The results revealed that total viable count on MCHs was higher than total viable count of ATM and public toilets. The results also showed that there were no growth observed on treated with disinfectants samples. The study has shown that CMHs are 2.2 times more contaminated than public toilets and 1.06 times more contaminated than ATMs. ATMs were shown to be 2.1 times more contaminated than public toilets.

**Table 1**

*Summary of the bacteriological counts of both untreated and disinfectant treated ATMs, public toilets and commercial motorcycle crash helmet samples (in cfu/ml)*

| Type of bacteria   | ATMs                 |                   | Public Toilets       |                   | MCHs                 |                   |
|--------------------|----------------------|-------------------|----------------------|-------------------|----------------------|-------------------|
|                    | Without disinfectant | With disinfectant | Without disinfectant | With disinfectant | Without disinfectant | With disinfectant |
| Total viable count | $0.4325 \times 10^5$ | *                 | $0.2053 \times 10^5$ | *                 | $0.4608 \times 10^5$ | *                 |
| Gram positive      | $0.2392 \times 10^5$ | *                 | $0.1625 \times 10^5$ | *                 | $0.1752 \times 10^5$ | *                 |
| Gram negative      | $0.1897 \times 10^5$ | *                 | $0.1985 \times 10^5$ | *                 | $0.2818 \times 10^5$ | *                 |

\*: means that there were no microbial counts on Petri dishes, because the numbers of colonies chosen to be counted were within ranges between 30 and 300.

cfu= colony forming unit

**Gram staining results:** During this study, Gram staining techniques were performed for all colonies used in biochemical test. Different results were obtained:- Gram-negative bacilli rods: a characteristic of *Enterobacteriaceae*, such as *E. coli*, *Salmonella*, *Klebsiella*, *Shigella* and *Pseudomonas*;

Gram positive cocci in clusters: a characteristic of *Staphylococcus spp.*, such as *S. aureus* and *Staphylococcus epidermis* and *Streptococcus* species.

**Biochemical tests results:** Table 2 shows the biochemical tests results from ATMs samples, public toilets

samples and samples from commercial motorcycle helmets. The different bacteria species identified with biochemical tests from the different sources are summarised. The presence of these bacteria explains the bacteriological contamination of ATMs, public toilets and commercial motorcycle helmets which can be the source of contamination and spreading of a bacteriological infection. In this study eight species of bacteria were isolated which include *Staphylococcus aureus*, *Staphylococcus epidermis*, *Streptococcus species*, *E. coli*, *Salmonella*, *Klebsiella*, *Enterobacter aerogenes* and *Pseudomonas*.

**Table 2**  
 Biochemical tests results from ATMs, public toilets and commercial motorcycles helmet samples

| Bio. Test ORG                   |      | Cata-lase | Coa-glase | C i t -rate | Gas  | H <sub>2</sub> S | MR   | VP   | Urease | Malonate | TSI         | Indole |
|---------------------------------|------|-----------|-----------|-------------|------|------------------|------|------|--------|----------|-------------|--------|
| <i>Staphylococcus aureus</i>    | ATMs | Pos.      | Pos.      | Neg.        | Neg. | Neg.             | Pos. | Neg. | Neg.   | Neg.     | A l k / Alk | Pos.   |
|                                 | PTs  | Pos.      | Pos.      | Neg.        | Neg. | Neg.             | Pos. | Neg. | Neg.   | Neg.     | A l k / Alk | Pos.   |
|                                 | CMHs | Pos.      | Pos.      | Neg.        | Neg. | Neg.             | Pos. | Neg. | Neg.   | Neg.     | A l k / Alk | Pos.   |
| <i>Staphylococcus epidermis</i> | ATMs | Pos.      | Neg.      | Neg.        | Neg. | Neg.             | -    | -    | Neg.   | Neg.     | A / A       | Neg.   |
|                                 | PTs  | Pos.      | Neg.      | Neg.        | Neg. | Neg.             | -    | -    | Neg.   | Neg.     | A / A       | Neg.   |
|                                 | CMHs | Pos.      | Neg.      | Neg.        | Neg. | Neg.             | -    | -    | Neg.   | Neg.     | A / A       | Neg.   |
| <i>Streptococcus spp.</i>       | ATMs | Neg.      | -         | Pos.        | Neg. | Neg.             | -    | -    | Pos.   | -        | A / A       | Pos.   |
|                                 | CMHs | Neg.      | -         | Pos.        | Neg. | Neg.             | -    | -    | Pos.   | -        | A / A       | Pos.   |
| <i>E. coli</i>                  | ATMs | Pos.      | Neg.      | Neg.        | Pos. | Neg.             | Pos. | Neg. | Neg.   | Neg.     | A / A       | Pos.   |
|                                 | PTs  | Pos.      | Neg.      | Neg.        | Pos. | Neg.             | Pos. | Neg. | Neg.   | Neg.     | A / A       | Pos.   |
|                                 | CMHs | Pos.      | Neg.      | Neg.        | Pos. | Neg.             | Pos. | Neg. | Neg.   | Neg.     | A / A       | Pos.   |
| <i>Salmonella</i>               | ATMs | Pos.      | Neg.      | Pos.        | Pos. | Pos.             | Pos. | Neg. | Neg.   | Neg.     | Alk / A     | Pos.   |
|                                 | PTs  | Pos.      | Neg.      | Pos.        | Pos. | Pos.             | Pos. | Neg. | Neg.   | Neg.     | Alk / A     | Pos.   |
|                                 | CMHs | Pos.      | Neg.      | Pos.        | Pos. | Pos.             | Pos. | Neg. | Neg.   | Neg.     | Alk / A     | Pos.   |
| <i>Klebsiella</i>               | ATMs | Neg.      | Neg.      | Pos.        | Pos. | Neg.             | Neg. | Pos. | Pos.   | Pos.     | Alk / A     | Neg.   |
|                                 | PTs  | Neg.      | Neg.      | Pos.        | Pos. | Neg.             | Neg. | Pos. | Pos.   | Pos.     | Alk / A     | Neg.   |
|                                 | CMHs | Neg.      | Neg.      | Pos.        | Pos. | Neg.             | Neg. | Pos. | Pos.   | Pos.     | Alk / A     | Neg.   |
| <i>Enterobacter aerogenes</i>   | ATMs | Pos.      | Neg.      | Pos.        | Pos. | Neg.             | Neg. | Pos. | Neg.   | Pos.     | A / A       | Neg.   |
|                                 | CMHs | Pos.      | Neg.      | Pos.        | Pos. | Neg.             | Neg. | Pos. | Neg.   | Pos.     | A / A       | Neg.   |
| <i>Pseudomonas</i>              | ATMs | Pos.      | Neg.      | Pos.        | Neg. | Neg.             | Neg. | Neg. | Neg.   | Neg.     | A l k / Alk | Neg.   |
|                                 | PTs  | Pos.      | Neg.      | Pos.        | Neg. | Neg.             | Neg. | Neg. | Neg.   | Neg.     | A l k / Alk | Neg.   |
|                                 | CMHs | Pos.      | Neg.      | Pos.        | Neg. | Neg.             | Neg. | Neg. | Neg.   | Neg.     | A l k / Alk | Neg.   |

Pos. =positive, Neg. = negative, A/A= lactose and or sucrose/glucose positive, Alk/A= lactose and or sucrose negative/ glucose positive, Alk/Alk = lactose and or sucrose/ glucose negative, ORG= organism, Bio. Test= biochemical test, PTs = Public Toilets, CMH= Commercial Motorcycle Helmets.

**Table 3**  
Occurrence of isolated bacterial species among the total examined study subjects

| Bacterial species               | Number positive/<br>Total examined | Percentage<br>positive | Test method positive  |
|---------------------------------|------------------------------------|------------------------|---|
| <i>Staphylococcus aureus</i>    | 10/25                              | 40%                    | Catalase, Coagulase and Gram stain positive                               |
| <i>Staphylococcus epidermis</i> | 6/25                               | 24%                    | Catalase and Gram stain positive  |
| <i>Streptococcus spp.</i>       | 7/25                               | 28%                    | Gram stain positive   |
| <i>Escherichia coli</i>         | 22/25                              | 88%                    | Citrate, Urease, Catalase, Gram stain negative and colonies features      |
| <i>Salmonella</i>               | 11/25                              | 44%                    | TSI, Citrate, Urease, Catalase, Gram stain negative and colonies features |
| <i>Klebsiella</i>               | 3/25                               | 12%                    | MR-VP, Malonate, Gram stain negative and colonies features                |
| <i>Enterobacter aerogenes</i>   | 1/25                               | 4%                     | MR-VP, Malonate, Catalase, Gram stain negative and colonies features      |

MR-VP= Methyl red-Voges Prausker reagent, TSI= Triple sugar iron agar

**Table 4**  
Bacteriological examination recorded results from Kanombe Hospital registry book between 2011 and 2013

| Ser No | Organism                     | Specimen type   | 2011            |        | 2012            |        | 2013            |        | Total           |        |
|--------|------------------------------|-----------------|-----------------|--------|-----------------|--------|-----------------|--------|-----------------|--------|
|        |                              |                 | % post Tot exam | % post | % post Tot exam | % post | % post Tot exam | % post | % post Tot exam | % post |
| 1      | <i>Salmonella spp.</i>       | Blood/stool     | 23/255          | 9%     | 22/205          | 10.7%  | 3/73            | 4.1%   | 48/533          | 9.0%   |
| 2      | <i>Pseudomonas spp.</i>      | Blood/stool/pus | 9/139           | 6.5%   | 2/77            | 2.6%   | 4/108           | 3.7%   | 15/324          | 4.6%   |
| 3      | <i>Staphylococcus aureus</i> | Blood           | 8/164           | 4.9%   | 13/166          | 7.8%   | 2/109           | 1.8%   | 23/439          | 5.2%   |
| 4      | <i>Escherichia coli</i>      | Stool           | 2/46            | 4.3%   | 0/58            | 0      | 5/118           | 4.2%   | 7/222           | 3.2%   |

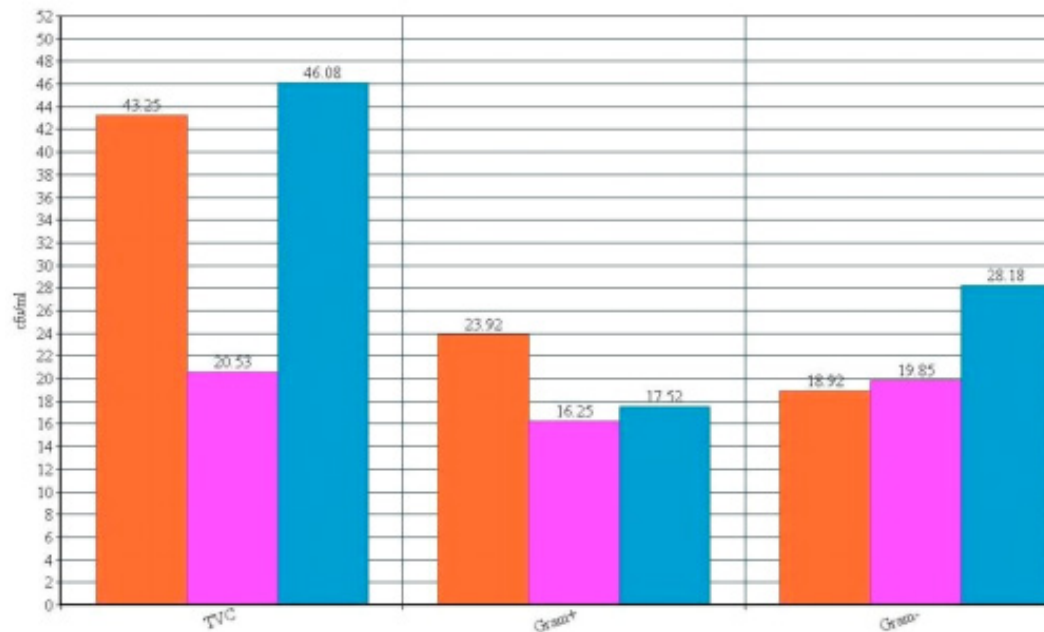
No pos= Number positive, Tot exam= Total examined, % post= Percent positive

*Level of bacteriological contamination tests results:* Figure 1 describes the level of bacteriological contamination of ATMs, Public toilets, and commercial motorcycle crash helmet samples. The results show that high

number of Total Viable Count (TVC) was found on MCHs samples, the high number of gram positive was found on ATMs samples and the high number Gram negative was found on MCHs samples.

**Figure 1**

Level of bacteriological contamination test results where on Y-axis CFU/ml and on X-axis microorganisms are shown



TVC= Total Viable Count, Gram+= Gram positive, Gram- = Gram negative

*Results for samples collected from unwashed hands surfaces:* Analysis from culture media, biochemical test and Gram staining show that seven species of bacteria were isolated from unwashed hands of the study subjects. The result is presented in Table 3. The bacteria were *Staphylococcus aureus*, *Staphylococcus epidermis*, *Streptococcus* species, *Escherichia coli*, *Salmonella*, *Klebsiella* and *Enterobacter aerogenes*. In increasing order the most predominant were *Staphylococcus aureus* (40%), *Salmonella* (44%) and *Escherichia coli* (88%). In decreasing order the less occurred in the subjects were *Streptococcus* species (28%), *Staphylococcus epidermis* (24%), *Klebsiella* (12%) and *Enterobacter aerogenes* (4%).

*Results for the retrospective bacteriological examinations:* Table 4 shows bacteriological examination recorded results from Hospital registry book between 2011 and 2013. Four different bacterial organisms, namely, *Salmonella spp.*, *Pseudomonas spp.*, *Staphylococcus aureus* and *Escherichia coli* were identified from different types of specimens. Of the total specimens examined *Salmonella spp.* was recovered from 9.0% (48/533) samples, *Pseudomonas spp.* from 4.6% (15/324), *Staphylococcus aureus* from 5.2% (23/439) and *Escherichia coli* from 3.2% (7/222).

## DISCUSSION

ATMs, public toilets and commercial motorcycle crash helmets play a vital role in our daily life whilst the contribution of hands contaminated with pathogenic

and non-pathogenic microorganisms to the spread of infectious disease has been recognised for many years (19, 20).

The results obtained from the bacteriological analysis of ATMs, public toilets and commercial motorcycle crash helmets indicate that high numbers of bacteria were found in surfaces which is accordance to work done by Anderson and Palombo, 2009 (21); who found between 99-100% contaminations of dry surfaces in a research Centre. Depending on environmental conditions, pathogens may remain infectious on surfaces for weeks after the contamination event. The longer the survival of a bacterium on a surface like the keyboard, then the potential of that bacterium being picked up by someone becomes considerably increased (22). Contamination of these surfaces is aided by personal hygiene and lifestyle of users of these equipments. Thus MCHs were highly contaminated which could be attributed to the frequency of their uses. Previous works have shown that frequently used fomites are most likely contaminated and therefore carry higher heterotrophic bacterial loads (23). The fact that the contaminants were at high level in this study is of great concern.

Our results indicate that *Staphylococcus aureus* was the most frequently isolated bacteria on ATMs. This confirms the study done by Stephen and Kwaku in 2011 (24), which found that their presence on the fingers, even after washing if washing was not thoroughly done especially as it is a known resident micro-flora of the skin (19). This study is also

substantiating that this bacteria species was detected in 40% of the unwashed hands of the study subjects. An attempt made to correlate this finding with infecting organisms recovered from the unwashed hands of study individuals' show that five species of bacteria were isolated as infectious organisms (Table 3). Hands as primary mode of transmission of respiratory, intestinal and skin infections as well as diarrheal diseases is well recorded (19, 20). Similar association was done with hospital bacteriological retrospective data. Four of the organisms isolated from this finding were also recorded in the hospital found in Kigali city between 2011 and 2013. Three of them were known to be infectious to human beings (27, 28, and 31).

On MCHs *Staphylococcus aureus* was the highest isolated bacteria, this also confirm the study done by Adamu and his colleagues in 2012 (25). On public toilets the highest isolated bacteria was *E. coli*. The latter organism was also detected from 88% of the present study persons. This indicates that the existence of fecal contaminations.

The results show that ATMs has the high number of isolated bacteria than public toilets. This finding also shows that the ATMs that were inside the house were showed less contaminated by bacteria than those which were not inside the house. Besides, there were more bacteria in women toilets than in men toilets; which is similar to the findings of Kennedy *et al.*, 2005 (26). This may be due to certain habits of women which tend to enhance contamination.

After treating with disinfectants, there were no bacteria growth on all samples obtained from ATMs, public toilets and commercial motorcycle crash helmets.

In this study eight species of bacteria were isolated. Five of them were recorded as pathogenic (26, 27, 28, and 29). This has a lot of health implications (11). Many of the isolates were the same in all samples except two species (*Streptococcus spp.* and *Enterobacter aerogenes*) that were not on public toilets.

*Staphylococcus aureus* is known to cause boils, abscesses, wound infections, toxic shock syndrome, pneumonia and other disease (27, 28). On the other hand, *Staphylococcus epidermis* is a common skin resident responsible for endocarditic and infections of patients with lowered resistance (29). Their presence indicates that the use of ATMs, public toilets and commercial motorcycle crash helmets can lead to transmission of serious skin infections.

*Streptococcus spp.* is one type of bacteria that is found on toilet seats in this investigation. *Streptococcus A* strains are found in the throat and skin and can cause strep throat and impetigo, a common skin infection that primarily affects children (26).

The occurrence of *Enterobacter aerogenes* and *E. coli* indicate possible faecal contamination of the

ATMs, public toilets and commercial motorcycle crash helmets. The implication of this is that the handling of ATMs, public toilets and commercial motorcycle crash helmets could be a potential source of food poisoning when infected hands are used in eating and food preparation without proper hygiene of hand washing (28).

*Pseudomonas* was recovered from all three surfaces studied. This bacterium infects people with low immune resistance such as cystic fibrosis patients. It also invades burns and causes urinary tract infection (27).

Four species of microorganisms, namely, *Escherichia*, *Klebsiella*, *Pseudomonas* and *Salmonella*, which have been isolated from ATMs and CMHs their pathogenic properties is well documented. Almost all serovars and species of *Salmonella* are known to be pathogenic (30, 31). *Salmonella spp.* has been found to survive on dry surfaces for long periods, making its presence significant (32).

ATM machines and commercial motorcycle crash helmets were shown to be heavily contaminated with bacteria; to the same level as nearby public toilets. In addition the bacteria we detected on ATMs were similar to those from the toilet, which are well known as causes of common human illnesses. The low counts on public toilets could be attributed to the fact that public toilets were thought of as the dirtiest places and many efforts in cleaning were done to keep it clean thus toilets tend to get cleaned frequently, which is different from ATMs and commercial motorcycle crash helmets often left out of the cleaning routine.

In this study, women's toilets were shown to have a higher bacteriological count than men's toilets. It is speculated that women's toilets contain more bacteria because of higher use frequency and toilet seats are often wet creating a favorable environment aiding bacteria to persist for longer periods of time. Humid conditions were found to increase survival times for most types of bacteria (33) that may be the reason why there are more bacteria in women's toilets.

Some samples from toilet seats were collected immediately after being cleaned by attendants and the results have shown it is not enough to keep the toilets seat clean, disinfecting is also needed as it has been found that disinfecting a toilet seat can eliminate pathogenic bacteria.

The effect of treating ATMs, public toilets and commercial motorcycle crash helmets with disinfectants has been demonstrated in this study. It was found that there were no growths at all on all samples treated with disinfectants (Table 1). This was due to the capacity of diffusion through the cell membrane as there were mixed with purified water (34). Disinfectants that have been used were alcohol based disinfectants, there were easily found in every super markets around Kigali at a low cost. In addition, the disinfectants left a good smell where there have



been used which is advantageous compared to other disinfectants that leave unpleasant smell on surfaces. In this study for sample collection the cotton swab rinse method was employed. This method is the standard technique to determine microbial contamination (17). The method is appropriate for high level of bacterial contaminations (16, 17). The limitation of this study was it does not utilised sample collection methods which have higher recovery performance described by others (15, 16).

### CONCLUSION

This investigation has revealed the ability of ATMs, CMHs and public toilets to serve for the transmission of microorganism with serious health implications. Cleaning with water has been shown to be ineffective, thus disinfectants are highly recommended on reducing bacteriological load on those public devices. Cleaning of these public devices will improve the image of their owners leveraging customers' loyalty and satisfaction. From these findings, it is clear that hygienic practices on public devices are far below expectations and it can be concluded that users are less informed of the risks involved in their usage of public devices and that these surfaces could act as carriers or vehicles/sources of potential pathogens. This situation calls for different role players in the public health sector to awake to their responsibilities in sensitising the public through different means the potential risk involved in publicly used contaminated surfaces in devices. If the public are informed of the microorganisms associated with public devices, it would help in reducing the risk of cross-transmission of bacterial infections through contaminated dried surfaces and would improve hygienic practices through regular and adequate disinfection of the surfaces. Appropriate agencies should also set standard for the public devices operators and monitor the practices from time to time as this will go a long way in helping to reduce microbiological contamination. Using passengers' hygienic head cover (Akanozasuku) to protect from unhygienic related diseases from helmets while on Motorcycle is also suggested.

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