

# CHAPTER ONE

## 1. INTRODUCTION AND OBJECTIVES

### 1.1. Introduction

Computer is an electronic data processing machine which accepts data from the out-side world in form of an input and manipulates, calculates, computes on the basis of set of instructions supplied and stored in the memory and give the required or desired results, in the form of an output to the user. Because of frequent-dermal contact by numerous users, microbial reservoirs of interest includes the computer keyboard and mouse (Chimezie *et al.*, 2013).

The average number of microorganisms present on multiple-user computer keyboards was significantly greater than on single-user keyboards (Chimezie *et al.*, 2013).

Scientific research has shown that commonly used surfaces such as computers, telephones, headsets, and desks are potential sources of infectious bacteria and viruses leading to the spread of colds, flu, sickness and diarrhea. They are constantly in contact with the environment wherever we go. Bacteria can survive in the microscopic grooves and cracks on surfaces and will go unnoticed. Oils on the skin, dust, grime, moisture and warmth from central heating systems provide an ideal environment for these germs to accumulate (Ashgar and El-said, 2012).

Bacteria are found on keyboards in computer laboratories at Central Connecticut State University. Bacteria are part of normal life; some may end up causing disease but other bacteria, a person's normal flora, are benign or beneficial. People pick up bacteria from environmental sources, including inanimate objects (fomites). There has been much research on bacterial contamination of computer keyboards, these studies usually demonstrate the presence of significant levels of contamination including pathogens (Chery and Davis, 2006).

The increased availability of multiple-user computers in the institutions setting means that these items or equipment are handled by numerous users on a daily basis. Given that computers are not routinely disinfected, the opportunity for the transmission of contaminating microorganisms is potentially great. Understanding of the ubiquity of microorganism in the environment is developing, but the risk or hazard of contamination posed by the computer keyboards and mouse is not yet fully understood. No clear legislation or even widely

recognized guidelines have been formulated on the hazard caused by computer components. This is not in the best interest of campus students especially that computer keyboards and mice could spread significant number of pathogens (Ali *et al.*, 2013).

### 1.2. **Rationale**

Most people do not realize that microbes are found on many common objects. In fact 80%of infections are spread through hand contact with hand or other objects (Reyonlds *et al.*, 2005).

In the Sudan there were few studies that focused on the Gram positive bacteria on computer keyboard and mouse. Furthermore the multiple users of computer keyboard and mouse are potential reservoir for microbial contamination, which routinely used in the universities and have indicated that about 100% of students have access to computer.

This study aimed to highlight the importance of assessment of bacterial contamination in order to investigate the status of

bacterial contamination of computer component mainly keyboard and mouse in the universities.

### **1.3. Objectives**

#### **1.3.1 General objective**

To assess Gram-positive bacteria that exists on computer keyboard.

#### **1.3.2. Specific objectives**

1. To determine bacterial load on computer keyboard.
2. To isolate Gram-positive bacteria on computer keyboard.
3. To identify the isolated bacteria.

## **CHAPTER TWO**

### **2. LITERATURE REVIEW**

Computer is an electronic data processing machine which accepts data from the out-side world in form of an input and manipulates, calculates, computes on the basis of set of instructions supplied and stored in the memory and give the required or desired results in the form of an output to the user (Ravichandran, 2001). Because of frequent-dermal contact by numerous users, microbial reservoirs of interest includes the computer keyboard and mouse (Neely *et al.*, 2005a; Wilson *et al.*, 2006). Anderson and Palambo (2009) documented that the

average number of microorganisms present on multiple-user computer keyboards was significantly greater than on single-user keyboards. Computer hardware has been implicated as a potential reservoir for infectious agents (Neely *et al.*, 2005b). Of increasing concern, however, is the role of keyboards in the non-hospital environment as pathogen reservoirs (Eguia and Chambers, 2003). It follows that the ubiquitous sharing of public computers by a broad user base might facilitate increased transmission and prevalence of pathogenic microorganisms throughout the community (Eltablawy and Elhifnawi, 2009).

Inadequately performed hand hygiene and non-disinfected surfaces are two reasons why the keys and mouse-buttons of laptops could be sources of microbial contamination resulting consequently in indirect transmission of potential pathogens and nosocomial infections (Siegmund *et al.*, 2010; Chimezie *et al.*, 2013).

Computers continue to have an increased presence in almost every aspect of our occupational, recreational, and residential environments. In the university environment, students have indicated that 100% have access to computers, 92.1% regularly use the internet, and 73.3% regularly use e-mail. To accommodate the extensive use of computer technology, universities have developed multiple-user “computer laboratories” on campus for general computer centre. As the popularity of such facilities increases, there is a need to recognize that computer equipment may act as a reservoir for the transmission of potentially hazardous or pathogenic microorganisms. The ability of computers to act as fomites has been previously

documented in hospital and health care environments. In the workplace, contamination of the office environment (including the computer keyboard) with bacteria is also recognized (Chairman *et al.*, 2011).

Keyboards have become reservoirs for pathogens because of the increased use of computers in patient areas (Bures *et al.*, 2000). The risk of transmission of pathogens from computer keyboards to patients would be prevented by compliance with current hand hygiene guidelines. Unfortunately, 34 studies have demonstrated that the mean rate of compliance with the Centers for Disease Control and Prevention guidelines on hand hygiene is approximately 40% among healthcare workers (Boyce and Pittet, 2002), which is a likely explanation for the frequent contamination of computer keyboards. This study was performed to determine the degree of microbial contamination, the efficacy of different disinfectants, and the cosmetic and functional effects of the disinfectants on the computer keyboards (Rutala *et al.*, 2006).

Most people do not realize that microbes are found on many common objects outdoors, in their offices, and even in their homes. Such objects include; playground equipments, ATM keyboards, computer keyboards, escalator handrails, elevator buttons and with the spread of supermarkets and hypermarkets the shopping carts handles. All of the latter objects are places that are most touched by the bare hands of people who are in various hygienic conditions. People believe that microbes are only present in research laboratories or in hospitals and clinics

and thus they have a misleading feeling of security in other places. Lack of knowledge about where germs prowl could be the cause of health problems. In fact 80% of infections are spread through hand contact with hands or other objects (Reynolds *et al.*, 2005).

Reynolds *et al.*, (2005) used an invisible fluorescent tracer for artificial contamination of public surfaces, they found that contamination from outside surfaces was transferred to 86% of exposed individual's hands and 82% tracked the tracer to their home or personal belongings hours later (Reynolds *et al.*, 2005).

The surfaces of computer keyboards and mice are often contaminated with nosocomial pathogens. When those are coming into contact with hands can serve as vehicles for infection transmission (Kramer *et al.*, 2006). The common bacteria that are commonly present on keyboards are coagulase-negative staphylococci (CoNS), diphtheroids and *Bacillus* species (Fukada *et al.*, 2008), however, methicillin resistant *Staphylococcus aureus* (MRSA) is also reported in some studies (Rutala *et al.*, 2006). Several investigations have been done on contamination of computer keyboards (Bures *et al.* 2000; Hartmann *et al.*, 2004), since some harmful bacteria can survive for >24 h on computer keyboards and keyboards in hospitals may therefore

contribute to cross-transmission of bacteria (Devine *et al.*, 2001; Wilson *et al.*, 2005).

Enterococci have been found to survive in dry conditions and on various fabrics utilized in the health care environment. Infection doses of pathogens may be transferred to the mouth after handling an everyday contaminated household object (Rusin *et al.*, 2002).

Ghamdi *et al.*, (2011) investigated the status of bacterial contamination of four daily used objects, computer keyboards, computers mice, elevator buttons and shopping carts handles in the city of Jeddah, Saudi Arabia, and they concluded that 95.5% of the total samples collected were contaminated with mixed bacterial growth. Coagulase-negative staphylococci dominated the isolates.

In order to investigate the status of bacterial contamination of computer components, 50 samples (25 from keyboards and 25 from mice) were collected from the main internet center located in Al-Mustansiriya University, Baghdad, Iraq. A total of 59 isolates comprising 9 bacterial species were recovered from these samples, the frequencies of occurrence of the species were;



*Bacillus* species (25.42%), *Staphylococcus aureus* (18.64%),  
*Staphylococcus epidermidis* (10.17%), (Ali *et al.*, 2013).

study was carried out to isolate and to recognize microorganisms associated with computer keyboards and mouse in computer labs in Himachal Institutes Paonta Sahib (HP), five (5) bacterial species were isolated, The isolates included *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Micrococcus* sp, *Streptococcus* species (Kumar and Srivastava, 2012).

Mehdinejad *et al.*, (2012) investigated the microbial colonization of computer keyboards and mice in our medical school computer center, and the result was all the key boards and 26 (96.29%) out of total tested computer mice were contaminated with at least two kinds of bacteria. Coagulase-negative staphylococci was the most common contaminated bacteria recovered from computer keyboards (36.5%) and mice (38.8%). Other isolated bacteria were *Diphtheroids*, *Bacillus* species, and Enterobacteriaceae as the least isolates bacteria. *S. aureus* was detected on the 5 keyboards and 4 mice. In conclusion, this study has shown the multiple-user computer keyboards and mice as potential reservoir for microbial contamination, some of which are of importance in

transmission the nosocomial infections between medical students and patients in hospital wards.

Potential pathogens isolated from more than 50% of the computers included coagulase-negative staphylococci (100% of keyboards), *diphtheroids* (80%), *Micrococcus* species (72%), and *Bacillus* species (64%). Other pathogens isolated included oxacillin-resistant *Staphylococcus aureus* (ORSA) (4% of keyboards) oxacillin-susceptible *Staphylococcus aureus* (OSSA) (4%), vancomycin-susceptible *Enterococcus* species (12%) (Rutala *et al.*, 2006).

## **CHAPTER THREE**

### **3. MATERIALS & METHODS**

#### **3.1. Study design**

##### **3.1.1. Type of study**

This is a descriptive cross sectional study conducted to assess the Gram-positive on computer keyboard.

##### **3.1.2 Study duration and area**

This study was conducted from April to July 2014 in some Sudanese universities, Khartoum State.

##### **3.1.3. Collection of samples**

The computer keyboards were swabbed with a sterile cotton swab moistened in sterile saline (9% w/v). The cottony part of swab placed in 2 ml sterile normal saline. Laboratory analyses were conducted within 2 hours of samples collection.

#### **3.2. Laboratory diagnosis**

##### **3.2.1. Bacterial load**

Using pour plate method (viable count), the number of living bacteria in liquid culture was counted. A measured amount of the suspension is mixed with molten agar medium in a Petri dish. After incubation, the number of colonies was counted. Counts of pure cultures were made on plates inoculated to yield between 30 and 300 colonies.

A serial 10-fold dilution of the bacterial suspension was prepared.

Pipette 9 ml of diluents (e.g. normal saline) into each of several

sterile test tubes. With a sterile pipette 1 ml suspension was transferred into the first tube of diluents. With a sterile pipette the first dilution was mixed and then transferred 1ml into the next diluents. The remaining dilutions were prepared in the same way, using a fresh pipette for each.

Starting with the greatest dilution, 1 ml of each dilution was pipetted into three Petri dishes and then poured into each dish 15 ml containing clear nutrient agar, mixed and allowed to cool. Then incubated at 37 ° C. the colonies were counted in the three plates and the average number/ plate was multiplied by the dilution factor to obtain the viable count/ml in the original suspension (Collee *et al.*, 1996).

### **3.2.2. Bacterial identification**

#### **3.2.2.1 Gram's stain**

The Gram stain reaction was used to help identify pathogens in specimens and culture by their gram reaction (Gram-positive or Gram-negative) and morphology. Gram-positive bacteria stain dark purple with crystal violet and are not decolorized by alcohol and Gram-negative bacteria stain red because after being stained with crystal violet decolorized by alcohol.

The smears were fixed by dry heat and then covered with crystal violet for 30-60 seconds. The stain was rapidly washed by tap

water and tipped off the slide. The stained smear was then covered with iodine for 30-60 seconds. Iodine washed off and the smear was decolorized with alcohol and immediately washed with clean water. Safranin was added to the smear for 30-60 seconds. The red stain was then washed off with tap water and smear was subsequently air dried and microscopically examined using high resolution objective power (Cheesbrough, 2006).

### **3.2.2.2 Identification of Gram positive bacteria**

#### **3.2.2.2.1 Catalase test**

This test is used to differentiate those bacteria that produce catalase enzyme from non catalase producing bacteria. Catalase catalyst in the breakdown of hydrogen peroxide to oxygen and water. An organism is tested for catalase production by bringing into contact with hydrogen peroxide. Bubbles of oxygen are released if the organisms are catalase producer. The culture should not be more than 24 hours old (Cheesbrough, 2006).

#### **3.2.2.2.2 Coagulase test**

This test was used to differentiate *S. aureus* (which produce coagulase enzyme that convert plasma fibrinogen to fibrin) from other staphylococcal species (Cheesbrough, 2006).

#### **3.2.2.2.3 DNase test**

This test was used to identify *S. aureus* which produce deoxyribonuclease enzyme. DNase hydrolyses deoxyribonucleic acid (DNA). The tested organism was cultured on a medium which contain DNA, after overnight incubation the colonies were tested for DNase production by flooding plate with a weak (1 mole) hydrochloric acid solution. DNase producing colonies were surrounded by clear area due to DNA hydrolysis (Cheesbrough, 2006).

#### **3.2.2.2.4 Fermentation of mannitol**

Mannitol salt medium was used to differentiate *S. aureus* from other staphylococcal species. The suspected colonies were incubated aerobically at 37°C. *S. aureus* that fermented mannitol turned the indicator to yellow (Cheesbrough, 2006).

#### **3.2.2.2.5 Sugar fermentation test**

Prepare broth media and 10% sugar solution, add 99 ml of media with 1 ml of sugar solution to give final concentration 1%, pour 1 ml of each final solution of each sugar in sterile test tube and inoculate the tested organism at 30°C for 5 days and examine daily (Collee *et al.*, 1996).

## CHAPTER FOUR

### 4. RESULTS

This study analyzed 200 samples collected from computer keyboards. 131 samples showed bacterial growth. The growth was distributed as follows; Sudan University of Science and Technology 41, Al-Neelain University 33, University of Science and Technology 35, and Omdurman Alahlia University 22 (Table1). A total of 91 Gram-positive bacterial isolates were recovered from the keyboards. These were Gram-positive cocci 41, and *Bacillus* species 50 (Table 2). The Gram-positive cocci were differentiated as follows; *S. aureus* 11, *S. epidermidis* 12, *S. haemolyticus* 10, *S. schleiferi* 7, and *S. lugdunensis* 1 (Table 3).

**Table 1. Distribution of bacterial growth and load**

<b>Universities</b>	<b>No of sample</b>	<b>Growth</b>	<b>Mean bacterial load CFU/ml</b>
Sudan University of Science and Technology	65	41	30-250x10 <sup>2</sup>
Al-Neelain University	45	33	30-705x10 <sup>2</sup>
University of Science and Technology	55	35	40-787x10 <sup>2</sup>

Omdurman Alahlia University	35	22	300x10 <sup>2</sup>
Total	200	131	30-787x10 <sup>2</sup>

**Table 2. Bacterial isolates**

Isolates	Number (%)
Gram- postive cocci	41 (20.5)
Bacillus species	50 (25)

**Table 3. Types and number of Gram-positive bacterial species isolated from computer keyboards.**

Species	Number (%)
<i>S. aureus</i>	11 (21.9)
<i>S. epidermidis</i>	12 (29.2)
<i>S. haemolyticus</i>	10 (24.3)
<i>S. schleiferi</i>	7 (17.0)
<i>S. lugdunensis</i>	1 (2.4)
Total	41(20.5)



## **CHAPTER FIVE**

### **5. DISCUSSION**

This study investigated the load and types of contaminating microorganisms on the keyboards and of multiple-user computers located in some Sudanese universities.

In the present study the contamination rate of keyboard was high and this shows that microbial contamination also occurs on computer equipment located outside the hospitals and in an environment that is not directly connected to hospital. However most of isolated bacteria in this study were normal flora with coagulase-negative staphylococci (CoNS) as the most common isolated bacteria. In similar studies conducted in a tertiary care center and in a hospital, CoNS was reported as the major isolated organism at the rate of 100% and 96.7% respectively (Rutala *et al.*, 2006; Dogan *et al.*, 2008). Our study also demonstrated that microbial contamination of computer keyboards was prevalent and that commensal skin organisms were the most common contaminating microbes (Schultz *et al.*, 2003).

In this study the bacterial load was ranged from  $30 \times 10^2$  to  $700 \times 10^2$  CFU/m and that indicated the computer keyboards can become contaminated with pathogenic bacteria, this shows that microbial contamination also occurs on computer equipments located outside the hospitals and in an environment, and that is agree with other study which reported by (Chairman *et al.*, 2011).

However most of isolated bacteria in this study was normal flora with CoNS (29%). In similar study conducted in Medical School Computer Center (Chairman *et al.*, 2011), (Rutala *et al.*, 2006). Other studies demonstrated that *Bacillus* spp. was the predominant isolates (Ali *et al.*, 2013, Srikanth *et al.*, 2012). This is agreement with our study.

The rate of isolated *S. aureus* in our study was in concordant to other results as (21%), which was higher than 17.4% reported rate of *S. aureus* isolation in recent study (Lu *et al.*, 2009).

*S. aureus* is a major component of the normal flora of the skin and nostrils, which probably explains its high prevalence as a contaminant, as it can easily be discharged by several human active including sneezing, talking and contact with moist skin.

## **Conclusion**

In conclusion, this study has shown the multiple-user computer keyboards and mice as potential reservoir for microbial contamination, some of which are of importance in transmission the nosocomial infections between medical students and patients in hospital wards.

## **Recommendations**

1. The limitation of the study is that the data presented is based on one-time sampling.

2. Further studies may be carried out to determine the persistence of the microorganisms on computer keyboards over time, and simultaneous sampling of hands of the personnel.
3. Use of plastic covers and simple cleaning of computers with 70% isopropyl alcohol, may decrease the bacterial load.

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