

CHAPTER ONE

INTRODUCTION AND OBJECTIVES

1.1. Introduction

Fomites when on constant contact with humans or natural habitats of pathogenic organism constitute a major source of spread of infectious diseases (Osterholm *et al.*, 1995). The fomites include door handle of conveniences, showers, toilet, hand lockers especially those found in public offices, hospitals, hotels, restaurants and restrooms (Bright *et al.*, 2010). Beside the day to day interaction of people, which constitute one way of spreading disease, the major source of spread of community acquired infections are fomites (Presscott *et al.*, 1993).

Microorganisms are found everywhere, bacteria and fungi contaminate our body, our houses, work places, and whole environment. Fortunately among many billion of bacteria, only 1500 can be dangerous for our health, causing different disease such as pneumonia or skin infection (Eltablawy and Elhinfnawi, 2009). Microorganisms constitute a major part of every ecosystem. In these environments, they live either freely or as parasites (Sleigh and Timbury, 1998). The hand serve as a medium for the propagation of microorganisms from place to place and from person to person. Although it is nearly impossible for the hand to be free of microorganisms, the presence of pathogenic bacteria may lead to chronic or acute illness (Oranusi *et al.*, 2013).

Human hands usually harbor microorganisms both as part of body normal flora as well as transient microbes contacted from the environment (Lindberg *et al.*, 2004).

In the university environment, students have access to service offices regularly for different purposes. Given that the door handles are not routinely disinfected, the opportunity for the transmission of contaminating microorganisms is great. Although it is accepted that the infection risk in general community is less than that associated with patients in hospital (Scott *et al.*, 1982). The increasing incidence of epidemic outbreaks of certain diseases and its rate of spread from one community to the other has become a major public health concern (Nworie *et al.*, 2012).

1.2. Rationale

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. This is due to the lack of knowledge about where bacteria cause the health problem. Researchers considered that 80% of infections are spread through hands contact with hands or other objects (Al-Ghamdi *et al.*, 2011). The main reasons are difficulties to prevent the transfer of microbes that are already present in human bodies (Lues and Tonder, 2007). Hand washing is fundamental cautionary measure to protect against the spread of diseases and is one of the primary practices to reduce the transfer of bacteria from person to person, or from person to food contact surfaces (Chinakwe *et al.*, 2012). It is established that unwashed hands can transmit

pathogens, especially fecal pathogens, to food product after visit to the toilet. Investigation of food borne illness showed that poor personal hygiene, primarily ineffective hand washing is an important contributor to foodborne illness (Lambrechts *et al.*, 2014). Door handles of offices in Abuja metropolis were investigated for bacterial contamination. The researchers found that 86.7 % were positive (Nworie *et al.*, 2012). This study is expected to highlight the problem of door handles contamination in service offices in some universities in Khartoum State.

1.3 Objectives

1.3.1. General objective

To assess Gram-negative bacteria on door handles of service offices at some universities in Khartoum State.

1.3.2. Specific objectives

1. To isolate and identify G-negative bacteria on the door handles.
2. To estimate the load of bacterial contamination in the door handles.
3. To determine the of bacterial contamination on door handles.

CHAPTER TWO

LITERATURE REVIEW

2.1. Definition of door handles

A door handle is an attached mechanism used to open or close a door. Its location on the door may vary between few inches or centimeters away from the edge of the door to the exact center of the door, depending on the local culture, decorative style or owner preference. The term door handle tends to refer to round operating mechanisms. Door handles and door knobs are the same exact thing ([http://en.wikipedia.org/wiki/Door handle](http://en.wikipedia.org/wiki/Door_handle)).

2.2. History of door handles

The history of door handles is relatively unknown; there is no written history or documentation of who first invented the door handle and how they were fixed to the door. The different areas throughout time show different styles and different materials of door handles, for example in Victorian times the door handles were made from cast bronze with ornamental patterns. Through France and England in the 1800's most door handles were made from China or ceramic. From 1830-1873 there have been many patents for the different styles and forms of door handles, the differences include various materials and shapes. Still today we are constantly changing and inventing new door handle styles and forms, most door handles today are made from

either iron, bronze, brass, aluminum, wrought iron, steel and stainless steel (www.ehardware.co.uk/acatalog/History_of_Door_Handles.html).

2.3. Bacterial contamination

It is generally acknowledged that inanimate objects can carry microorganisms originating from the surrounding environment. These attached microorganisms possess a bio-transfer potential, that is the ability to be transferred to another substratum where growth is possible — for example on food, or on the human body (Joanna Verran, 2012). The spread of infectious diseases through hand contact has been an area of major concern. According to study conducted by Itah and Ben, (2004). Enteric bacteria such as *Escherichia coli*, *Klebsiella* spp, *Citrobacter* spp, were found to contaminate various contact surfaces including door handles and many other common house hold fixtures (Itah and Ben, 2004). Fomites consist of either porous or non porous surfaces or inanimate objects that when contaminated with pathogenic microorganism can transfer them to a new host thereby serving as vehicles in transmission (Greene, 2009). Fomites when in constant contact with humans or natural habitats of pathogenic organism which represent a major source of spread of infection diseases (Osterholm *et al.*, 1995). Such fomites include door handles of conveniences, showers, toilet seats and sinks, lockers, chairs, tables especially those found in public offices, hospitals, hotels, restaurants and restrooms (Bright *et al.*, 2010). Microorganism that cause infections can be found in any environment include soil, air, water and food as well as environmental surfaces or objects (Neely

and Sittig, 2002). Most of the bacteria found by researchers are normal flora in the skin, mouth and nasal passages, that can pass to our hands. Although many of these bacteria won't hurt unless the immune system is become weak because of another illness (oluduro *et al.*, 2011).

2.4. Previous studies

A survey of environmental surfaces in two Atlanta area, day care centers was conducted to determine the prevalence of fecal coliform bacteria, considered a marker for the presence of fecal contamination which might contain pathogenic parasites, bacteria, or viruses. Fecal coliforms were found in 17 (4.3%) of 398 representative samples of building surfaces, furniture, and other objects. These surfaces may be involved in the chain of transmission of enteric diseases among children. Therefore, disinfection of inanimate objects, in addition to good hand washing, may be important in controlling the spread of enteric diseases in day care centers. There were 10 (5.0%) positive plates from one center, and 7 (3.5%) from the other. Positive specimens were obtained from toilets, diapering items; floors, furniture, and a refrigerator handle (Weniger *et al.*, 1983).

Omololu-Aso *et al.*, (2011) investigated two hundred swabs from doctors' stethoscope diaphragm, cell phones of Health Care Workers (HCWS), patients' bed linen, pillows and door knobs at the Obafemi Awolowo University Teaching Hospitals Complex (OAUTHC). Cultures from the swabs were screened for *Staphylococcus aureus*. The

results showed that 18.70% of the doctors' stethoscopes, 20.33% of the doctors' cell phones, 20.33% of the doorknobs were contaminated with *S. aureus*

Oranusi *et al.*, (2013) collected 130 sample consisting of 40 hand swabs; 20 each of food sample and food contact surfaces; 10 each of swabs from banisters table ,table top, door handles, taps handles and toilet flushers were collected from different location of the university campus. They found that about 98% of hand swabs; food contact and the easy contact surfaces were contaminated with organism. Hand swab from the halls of residence and library had higher level of contaminations 2.1×10^5 and 1.9×10^5 cfu respectively. Toilet flushers and Banisters had 8.3×10^6 . Microorganism isolated by their study include *Bacillus* spp; *Staphylococcus* spp; *Streptococcus* spp; *Escherichia coli*; *Salmonella* spp and *Klebsiella* spp.

Baadhaim *et al.*, (2011) indicated that the door handles may aid in the spread of microbes between individuals and that they may be a reservoir of microbial contamination. In their experiments, they assessed the prevalence of Gram negative bacteria that were found on door handles of Olin Hall. It was hypothesized that during times where the building was near its peak usage, a larger percentage of the bacteria sampled from the door handles of Olin Hall would be Gram negative. The results showed that of total microbial colonies observed as 49% were Gram negative bacteria.

Another study held by Nworie *et al.*, (2012) recognized that the increase incidence of outbreaks of certain diseases and its rate of spread from one community to the other

become a major health concern. The sample collected from the Door handles/knobs of public conveniences of selected public offices, motor parks, and markets in Abuja metropolis were investigated for bacterial contamination. Total of 180 swab samples cultured, 156 (86.7%) were positive. The most positive samples from female toilet handles/knobs (41.7%) and bathroom door handles/knobs than males (11.5%). The study also found that toilet door handles/knobs in markets, motor parks and restaurants had higher rate of contamination compared to Government offices, and banks. Contamination was also higher in toilet door handles/knobs (87.2%) than in bathroom door handles/knobs (85%). Most of the bacteria contaminants were Coliforms. The isolated bacterial contaminants were *Staphylococcus aureus* (30.1%), *Klebsiella pneumoniae* (25.7%), *Escherichia coli* (1%), *Enterobacter* species (11.2%), *Citrobacter* species (7.1%), *Pseudomonas aeruginosa* (5.9%), and *Proteus* species (4.5%). This shows that the city's convenient places harbors highly pathogenic bacteria which have the potentials of causing epidemics in the near future

The prevalence of bacterial organisms on toilet door handles in secondary schools in Bokkos Local Government; Jos Plateau State, Nigeria was evaluated by Maori *et al.*, (2011). A total of 120 samples were collected and cultured, 40 from each of the schools (Government Secondary School Bokkos (G. S. S.B), All Nation Academy and Government secondary School Mushere). Out of the 120 samples that were collected 60(50%) yielded growth and 60 showed no growth at all. The following organisms were isolated *Staphylococcus* species (43.3%), *Candida species* (10%),

Escherichia coli (16.7%), *Citrobacter* species (1.7%), *Klebsiella* species (20%), *Proteus* species (6.7%) and *Salmonella* species (1.7%). The result showed that G. S. S. B has the highest contamination (48.3%) followed by All Nations Academy (30%) and then G. S. S. M (21%).

Scott *et al.*, (1982) carried out an investigation about the bacterial flora in over 200 homes. 60 samples collected from bathroom, toilet and kitchen. 9 sample from living room. The result of bacterial contamination as percentage occurring in 200 homes was as following ; *E. coli* 64.5%, *Klebsella Pneumoniae* 29.5%, *Klebsiella* spp, 6%, *Proteus mirabilis* 4%, *Salmonella* spp. 1.5%, *Citrobacter freundii* 42%, *Citrobacter* spp. 29%, *Enterobacter cloacae* 26%, *Enterobacter agglomerans* 7.5%; *Pseudomonus aeruginosa* 4%, *Staphylococcus aureus* 31.5%, *Streptococcus* spp. 16%, the majority of homes were contaminated with *enterobacteria* species and *Pseudomonus* species, many of which are potentially pathogenic. Other potential Pathogens included *Staphylococcus aureus* and *Streptococcus* species .

A study was carried out by Sabra, (2013) on public female restrooms at Taif, Kingdom of Saudi Arabia; Restrooms (RR) from different buildings, in order to characterize the locality of contamination and bacterial loads. 260 sample collected from different rest room (RR) like (RR Door, RR Handle; RR sink; RR Toilet door; RR Toilet handle). Incidence of bacterial growth or positive culture was 187/260 (71.9%). The predominant positive was from RR Toilet Handle in 73/80 (91.3%), then followed by RR Toilet Door in 59/80 (73.8%), RR Sink in 38/60 (63.3%), RR

Handle in 10/20 (50%), finally less positive from RR Door in 7/20 (35%). Different isolated bacteria arranged according to their percentage as *Staphylococcus aureus* 76/187 (40.6%), *Escherichia coli* 42/187 (22.5%), *Bacillus* spp. 40/187 (21.4%), *Klebsiella pneumoniae* 25/187 (13.4%), *Enterococcus faecalis* 18/187 (9.6%), *Citrobacter* spp. 16/187 (8.6%), *Pseudomonas aeruginosa* 13/187 (7%) and *Proteus mirabilis* 10 /187 (5.3%).

As well known that harmful microorganisms can be transferred to hands from contaminated surfaces. These Contaminated hands can transmit disease to own self as well as to others according to a study that done to determine to which extent the hand hygiene practices and toilet door knobs contribute to the bacterial load of hands of toilet users in a medical school. Swabs were taken from a randomly selected sample of 60 medical students for bacterial count from both hands before and after toilet use and from door knobs of six toilets. Only 40 (66.7%) claimed washed their hands with soap. Significantly more females (83%) used soap to wash hands compared to males (50%). Bacterial load in the hands of both males and females showed an increase after toilet use. The increase was significant among male students. The dominant hand had a significantly higher bacterial load than the other. The mean bacterial loads of male toilet door knobs (12 CFU/cm²) were significantly higher than of female toilet door knobs (2.5 CFU/cm²). *Staphylococcus aureus* was isolated from the hands of 21 students (De Alwis et al., 2012).

Fomites are inanimate objects that can serve as vehicles for pathogens transfer. Maryam et al., (2014) conducted a study to determine the pathogenic bacteria isolated

from fomites in a teaching hospital in Nigeria. Exactly 35 samples were used. Twenty three (65.7%) isolates were obtained; the ratio of Gram positive to Gram negative organisms was 12:11. The bacteria isolated were *Staphylococcus aureus* (21.7%), *Staphylococcus epidermidis* (8.7%), *Streptococcus* spp. (8.7%), *Bacillus* spp. (13.0%), *Escherichia coli* (26.1%), *Pseudomonas* spp. (8.7%) and *Klebsiella* spp. (13.0%).

Other a study was conducted to determine the prevalence of some pathogenic bacteria and the general hygienic status on the interior surfaces of some domestic refrigerators (n = 150). *Campylobacter* spp., and *Salmonella* spp. were not recovered from any refrigerators, but *Staphylococcus aureus* was recovered from 9.54%, *Listeria monocytogenes* 3.8%, *Escherichia coli* from 2.1% and *Yersinia enterocolitica* from 1.6% of examined refrigerators. That indicated very poor standards of consumer refrigerator management and hygiene, and posing risks to consumer health (Abdulla et al., 2008).

The occurrence of enteric bacteria in kitchen sponges and dish cloths suggests that they can play a role or lead to the cross-contamination of foods, fomites and hands by food borne pathogens. Koenig, (2014) investigated the occurrence of bacteria in kitchen towels often used to dry dishes, hands and other surfaces in the domestic kitchen. A total of 82 kitchen hand towels samples were collected from households in five major cities in the United States and Canada and the numbers of heterotrophic bacteria, coliform bacteria, and *Escherichia coli* in each towel were determined. Coliform bacteria were detected in 89.0% of the samples and *E. coli* in 25.6% of total coliform bacteria isolated from towels.

CHAPTER THREE

MATERIALS & METHODS

3.1. Study Design

3.1.1. Type of study

This is a descriptive cross-sectional study.

3.1.2. Study Area

The study was conducted in Sudan University of science and technology (SUST) Alneelain University, University of Khartoum. The experimental work was done in the Research Laboratory (SUST)

3.1. 3. Study duration

The study was conducted during the period August-September, 2014.

3.1.4. Sample size

A total of 200 office door handles were included in this study.

3.2. Experimental work

3. 2.1. Collection of samples

The specimens were collected from door handles by means of sterile cotton swabs moistened in sterile nutrient broth. The swab was wiped firmly on the entire surface of the door handle. Each swab was placed in small tube, labeled and immediately transported to the Research Laboratory.

3. 2.2. Culture

The swabs were used to inoculate nutrient agar plates. The plates were incubated aerobically at 35 °C. Bacterial growth was checked after 24-48 hours. At the end of incubation period, bacterial load was estimated semi-quantitatively depending on the number of colony-forming unit (CFU) as follows; 10-20 CFU= ++; 21-30 CFU= +++ and 31-40 CFU= ++++.

3.3. Identification of Gram-negative bacteria

3. 3. 1. Colonial morphology

Different morphological feature of the yielded colonies including color, size, elevation and pigmentation were recorded.

3. 3.2. Gram stain

Gram stain was essential step for the next experimental work to identify Gram-negative isolates. The procedure was carried out according to Cheesbrough (2006) as follows; smear was prepared from overnight culture on a clean and dry slide. The smear was left to air dry. Fixation was done by rapid pass the slide three times through the flame of a Bunsen burner then allowed to cool before staining. Crystal violet stain was added to smear for 30–60 seconds, and then washed by tap water. Lugol's iodine was added for 30-60minutes then washed by tap water and decolorized rapidly (few seconds) with acetone alcohol and washed immediately by tap water. Finally, the smear was covered with saffranin stain for 2 minutes and washed by tap water. The back of slide was wiped clean and placed in a draining rack for smear to air dry. Drop of oil was added to the dried smear and examined under the light microscope (Carl Zeiss, Germany) by oil lens 100X.

3. 3.3. Biochemical tests

3.3.3.1. Sugar fermentation, gas and H₂S production

A tube of KliglerIron Agar was inoculated using a sterile straight wire, first the butt was stabbed then the slope was streaked and incubated at 35–37°C overnight. Lactose fermenting bacteria was appeared as yellow butt and yellow slope, glucose fermenting bacteria was appeared as yellow butt and red slope, non-lactose and non- glucose fermenting bacteria was appeared as red butt and red slope, blackening in the media

indicated hydrogen sulphide production and cracks in the medium was due to gas production (Cheesbrough, 2006).

3.3.3.2. Citrate utilization test

The measurement of this characteristic is important in identification of *Enterobacteriaceae*. Utilization of citrate by tested bacteria was detected in Simmons citrate medium by the production of alkaline byproducts. The medium contained sodium citrate as sole source of carbon and ammonium phosphate as sole source of nitrogen. Bacteria that can use citrate can also extract nitrogen from ammonium salt with production of ammonia. A well-isolated colony was picked from the surface of the medium and inoculated as a single streak on the slant surface of citrate agar tube. The tube was incubated at 35°C for 24 to 48 hours. A positive color was represented by the development of deep blue color within 24 to 48 hour (Cheesbrough, 2006).

3.3.3.3. Indole test

Indole production is an important characteristic in the identification of many species of microorganisms. Indole is one of metabolic degradation products of the amino acid tryptophan. The test is based on the formation of a red complex when indole reacts with aldehyde group of p-dimethylaminobenzaldehyde which is the active chemical in Kovac reagent. Tryptophan broth was inoculated with the test organism and incubated at 37°C for 18 to 24 hour. At the end of incubation period few drops of Kovac reagent were added, and the development of red color at the interface of the reagent and broth

within seconds after adding the reagent was an indicator of presence of indole (Cheesbrough, 2006).

3.3.3.4. Urease test

The test was used to determine the ability of organisms to produce the enzyme urease, which hydrolyzes urea. Hydrolysis of urea produces ammonia and CO_2 , the formation of ammonia alkalizes the medium and the pH shift was detected by the color change of phenol red from light orange to magenta which indicated a positive result. A well-isolated colony was picked from the surface of the medium and inoculated as single streak on the slant surface of Christensen's urea agar (Cheesbrough, 2006).

3.3.3.5. Oxidase test

This test used to determine bacteria that produce oxidase enzyme which oxidized the oxidase reagent (tetramethyl-p-phenylenediaminedihydrochloride) to give a dark-blue color. The test was performed by commercial discs impregnated with the oxidase reagent; a pure colony was smeared on the disc by sterile wooden stick. A positive reaction was indicated by developing deep blue color within 10 seconds (Cheesbrough, 2006).

CHAPTER FOUR

RESULTS

A total of 200 door handle swabs were collected from offices in different universities in Khartoum State. The universities included University of Khartoum, Sudan University of Science and Technology (SUST) and Alneelain University. The frequency and percentages of these swabs were presented in table (1).

Cultivation of these swabs on nutrient agar plates yielded bacterial growth on 87/200 (43.5%) plates. The rest of 113/200 (56.5%) swabs showed no bacterial growth. Those yielded bacterial growth were obtained from Alneelain University 40(46.0%),

SUST 24(27.6%) and University of Khartoum 23(26.4%) (Table2). Of the 87 bacterial growths, 55 were gram- negative bacteria and the rest of 32 gram- positive was excluded.

Study on colonial morphology of bacterial isolates, showed different patterns of feature. The size of the colonies ranged from small to large size. The majority of the colonies were entire circular and few were mucoid.

Bacterial load was recorded semi-quantitatively. The average of bacterial load estimated at different universities were as follows; University of Khartoum high (++++), SUST and Alneelain University moderate (+++) (Table3).

Biochemical tests adopted for identification of Gram-negative isolates were tabulated in table (4).

Gram negative bacteria recovered from door handles were *Pseudomonas* spp. 19 (34.6%), followed by *Klebsiella* spp.13(23.6%), *Escherichia coli* 10(18.2%), *Serratia* spp.7 (12.7%), *Proteus* spp. 3 (5.5%), *Citrobacter* spp .2 (3.6%) and *Yersinia* spp. 1 (1.8%) (Table5).

Table 1. Distribution of investigated swabs specimen according to collection sites

Collection Site	Swabs	

	Frequency	%
University of Khartoum	60	30
SUST	70	35
Al Neelain University	70	35
Total	200	100

Key: SUST= Sudan University of Science & Technology

Table 2. Frequency of Bacterial growth after primary cultivation of door handles swabs according to university

University	Swabs yielded bacterial growth	
	Frequency	%
University of Khartoum	40	46.0
SUST	24	27.6
Al Neelain University	23	26.4
Total	87	100

Key: SUST= Sudan University of Science & Technology

Table 3. Bacterial load estimated according to university

University	Bacterial load
University of Khartoum	++++
SUST	+++
Al Neelain University	+++

Key: 10-20 CFU= ++; 21-30 CFU= +++; 31-40 CFU= ++++

Table 4. Biochemical tests adopted for identification of Gram-negative bacteria

Isolate code	Biochemical tests								Suggested Organism
					KIA				
	Ox	Ind	Cit	Urea	Glu	Lact	H ₂ S	Gas	
1 DH	+	-	+	-	-	-	-	-	<i>Pseudomonas</i> sp.
2 DH	-	-	+	+	+	+	-	+	<i>Klebsiella</i> sp.
3 DH	-	+	-	-	+	+	-	+	<i>E. coli</i>

4 DH	-	-	+	-	+	-	-	-	<i>Serratia</i> sp.
5 DH	-	+	+	+	+	-	+	+	<i>Proteus</i> sp.
6 DH	-	-	+	-	+	+	-	+	<i>Citrobacter</i> sp.
7 DH	-	-	-	+	+	-	-	-	<i>Yersinia</i> sp.

Key: DH = Door handles, Ox = oxidase, Ind = indole, Cit = citrate, Glu = glucose, Lact = lactose, H₂S = hydrogen sulphate, Gas = gas, (+) = positive, (-) = negative.

Table 5. Frequency and percentage of Gram-negative bacteria isolated during this study

Isolated bacteria	Frequency	%
<i>Pseudomonas</i> sp.	19	34.6
<i>Klebsiella</i> sp.	13	23.6
<i>E. coli</i>	10	18.2
<i>Serratia</i> sp.	7	12.7

<i>Proteus</i> sp.	3	5.5
<i>Citrobacter</i> sp.	2	3.6
<i>Yersinia</i> sp.	1	1.8
Total	55	100

CHAPTER FIVE DISCUSSION

5.1. Discussion

The contaminated environmental surfaces such as door handles are commonly touched by hands, which may act as sources for hand transfer. In particular, the contamination of door handles of the service offices is not surprising, and it is usually supposed that the users considered the main source of contamination of the door handles rather than the other parts of the door. In the present study, the level of

contamination was less (33.3%) than that reported (49.0%) by Baadhaim *et al.*, (2011). Moreover, majority of isolated bacteria in this study are potentially pathogens. Similar results were reported by Weniger *et al.*, (1983) and Koenig, (2014) who stated that the occurrence of bacteria on surfaces were coliform bacteria like *E. coli*. Door handles contamination investigated in this study resulted in isolation of many types of Gram-negative bacteria. These are *Pseudomonas* spp. (34.6%), *Klebsiella* spp. (23.6%), *E. coli* (18.2%), *Serratia* spp. (12.7%), *Proteus* spp. (5.5%), *Citrobacter* spp. (3.6%), and *Yersinia* spp. (1.8%). *Pseudomonas* spp. was the prevalent Gram-negative bacterium (34.6 %). This result disagrees with that reported by Nworie *et al.*, (2012) who found that the prevalent bacterium in similar study was *Staphylococcus aureus* (30.1%). The percentage (23.6%) of *Klebsiella* spp. isolated during this study, confirmed the finding of Nworie *et al.*, (2012), who reported the percentage of the same organism as 25.7%. On the other hand, presence of *E. coli* (18.2%) in the door handles is less than that obtained by Sabra, (2013) on public female restrooms door handles at Taif, Kingdom of Saudi Arabia (22.5%). But similar results (5.5%) and (5.3%) respectively were obtained in the two studies regarding *Proteus* spp.

In this study the percentage of *Yersinia* spp. was (1.8%). This result is similar to that obtained by Maryam *et al.*, (2014) who reported 1.6%. The presence of *Yersinia* in the two studies with low percentages is attributed to the fact that this organism causes disease in children and very rare in adults.

To better protect public health in the Sudanese universities, it is important to highlight the need for effective disinfection to minimize the hazard posed or to reduce bacterial

load and contamination by Gram-negative bacteria that may come in contact with door handles of the service offices.

5.2. Conclusion

The study concluded that:

1. The load of bacterial contamination in the door handles of service offices in universities is considerable.
2. Potentially pathogenic Gram-negative bacteria are predominant.
3. The prevalent organism recovered during this study was *Pseudomonas* spp.
4. The use of cleaning methods is totally absent.

5.3. Recommendations

1. Regular cleaning of door handles of the service offices may reduce the load of bacterial contamination.
2. Use of self-disinfecting technology on the door handles to minimize the attachment of microbes or to delay the development of biofilm.
3. Use of the door handles made of a heavy metal such as silver or copper to reduce the microbial load.
4. Further studies are recommended to validate the results of the present study.

REFERENCES

1. **Abdulla, M. A., Siham, E. S., Alian Y. Y. H. A. and Amel, O. B. (2008).** “A study of the microbial content of the domestic refrigerators in Khartoum area (Khartoum North); *Sud .J.Vet.Sci.Anim.Husb.* **47**(1&2 2008)
2. **Al-Ghamdi A. K., Abdelmalek S. M. A., Ashshi A. M., Faidah, Shukri H. and Jiman-Fatani A. A. (2011).** Bacterial contamination of computer keyboard and mice. *J Bio*; **3**(18):2224-3208.

3. **Alice N. Neely, PhD and Dean F. Sitting, (2002).** “Basic microbiology and infection control information to reduce the potential transmission of pathogens to patient via computer hardware” *J. Am Med Info Assoc*; **9**(7):500-508.
4. **Bright K. R., Boone S. A. and Gerba C. P. (2010).** Occurrence of bacteria and viruses on elementary classroom surfaces and the potential role of elementary classroom hygiene in the spread of infectious diseases. *J School Nursing*; **26** (1):33-41.
5. **Baadhaim M., Kusner, D., and Ahmed H. (2011).** “Distribution and Prevalence of Bacteria Found on the Door Handles of Olin Hall, Drake University”. *Conference Poster*.
6. **Cheesbrough M. (2006).** District Laboratory Practice in Tropical Countries, Part 2, Cambridge University Press, United Kingdom, PP. 60-64.
7. **Chinakwe E. C., Nwogwugwu N. U., Nwachukwu I. N., Okorundu S. I., Onyemekara N. N. and Ndubuisi-Nnaji, U. (2012).** Microbial quality and public health implication of hand-wash water samples of public adults in Owerri, South-East Nigeria. *Inter Res J Microbiol.* ; **3**(4):144-146.
8. **De Alwis, W. R., Pakirisamy, P., Wai San, L., and Xiaofen, E. C. (2012).** A Study on Hand Contamination and Hand Washing Practices among Medical Students. *ISRN Public Health*; **3**; 1- 5.
9. **Eltablawy S. Y. and Elhinfnawi H.N. (2009).** Microbial contamination of some computer keyboards and mice in National center for Radiation Research and Technology. *J World Appl Sci*; **6**(4):162-7.

10. **Greene A. (2009).** Fomites, Available from: <http://www.drgreene.com/articles/fomites/>.Reviewed by Le-Bucklin K-V,Hicks R january 12,2009accessed 4 nov 2014.
11. **Itah B., and Ben A. E. (2004).** Incidence of Enteric Bacteria and Staphylococcus Aureus in Day Care Centers in Akwalbom State, Nigeria. *J South E Asi J. Trop Med & Pub Healt*; **35**(1):202-9.
12. **Joanna V. (2012).** School of Healthcare Science, Manchester Metropolitan University, Mancheste, the Microbial Contamination of Mobile Communication Devices. *J Microbiol Biol Educ*; **13**(1): 59–61.
13. **Koenig, D. W. (2014).** Occurrence in Kitchen Hand Towels. *Food Protection Trends*; **34**(5):312-317.
14. **Lambrechts A., Human I. S., Doughari J. H. and Lues J. F. R. (2014).** Bacterial contamination of the hands of food handlers as indicator of hand washing efficacy in some convenient food industries in South Africa. *Pak jMed Sci*; **30** (4): 755-758.
15. **Lindberg E., Adlerberth B., Hesselmar R., Saalman I., Strannegared N. and Aberg I. (2004).** A High rate of transfer of Staphylococcus aureus from parental skin to infant gut flora. *J Clini Microbiol*; **42**(2): 530-534.
16. **Lues J. F. R ,van Tonder .I (2007).** The occurrence of indicator bacteria on hands and aprons of food handlers in the delicatessen section of a retail group, *Food controll*; **8**(18):326-332.
17. **Maryam, A., Hadiza, U. S., and Aminu, U. M. (2014).** Characterization and determination of antibiotic susceptibility pattern of bacteria isolated from some fomites in a teaching hospital in northern Nigeria”. *J Afri Micro Res*; **8**(8): 814-818.

18. **Maori, L., Agbor, V. O., and Ahmed, W. A. (2011).** “The prevalence of bacterial organisms on toilet door handles in Secondary Schools in Bokkos LGA, Jos, Plateau Sate, Nigeria. *IOSR J Pharm Biological sci*; **8** (4); 85-91.
19. **Neely A. and Sittig D. F. (2002).** Basic microbiologic and infection control information to reduce the potential transmission of pathogens to patients via computer hardware. *J. Am Med Info Assoc*; **9**(5):500-508.
20. **Nworie. A., Ayeni. J. A., Eze U. A., and Azi. S. O. (2012).** Bacterial contamination of door handles/knobs in selected public conveniences in abuja metropolis, nigeria; a public health threat. *Continental J Med Res*; **6**(1): 7-11.
21. **Oluduro, A. O, Ubani, E. K. and Ofoezie , I .E, (2011).** “Bacterial assessment of electronic hardware user in interface in Ile-Ife, Nigeria” *J. Bas Appl.pharmaSci*; **32**(3):323-334.
22. **Oranusi S.U., Dahunsi S.O. ,Owoso O. O. and Olatile T. (2013).**“Microbial profile of hands, food, easy contact surfaces and food surfaces: A case study in a University Campus” *J Nov Inter Biotech & Biosci*; **2**(1):30-38.
23. **Osterholm M. T., Hederg C. W. and MacDonald K. I. (1995).** Epidemiology of infectious diseases. In: Mandell, Douglas and Bennett’s principles and practice of infectious diseases. Vol.1, 4th edition, Churchill-Livingstone, NewYork, p.165.
24. **Omololu-Aso, J., Kolawole, D. O., Omololu-Aso, O. O., and Ajisebutu, S O. (2011).** “Antibiotics sensitivity pattern of staphylococcus aureus from fomites in the ObafemiAwolowo University Teaching Hospital Complex (OAUTHC) Nigeria”. *Inter J Medi & Medic Sci*; **3**(2): 32-36.

25. **Prescott L. M., Harvey J. P. and Klein D. A. (1993).** Microbiology, 2nd edition .W.M.C. Brown, England.Pp.706-707,805.
26. **Sabra, S. M. (2013).** Bacterial public Health Hazard in the public Female Restrooms at Taif, KSA, *Middle-East J Scientific Res*; **14**(1):63-68.
27. **Scott, E., Bloomfield S. F. and Barlow C. G. (1982).** An investigation of microbial contamination in the home . *J Hyg*; **89**(2):279-293.
28. **Sleigh D. J. and Timbury M. C. (1998).** Note on Medical Microbiology,5th edition. Churchill-Livingstone, New York. p173.
29. **Van Tonder I and Lues J. F. R. (2007).** the occurrence of indicator bacteria on hands and aprons of food handlers in the delicatessen section of a retail group.*Food control*; **18** (8): 326- 332.
30. **Weniger, B. G., Ruttenber, A. J., Goodman, R. A., Juranek, D. D., Wahlquist, S. P., & Smith, J. D. (1983).**“ Fecal coliforms on environmental surfaces in two day care centers”. *Applied and environmental microbiology*, **45**(2):733-735.
31. [http: //en.wikipedia.org/wiki/Door_ handle](http://en.wikipedia.org/wiki/Door_handle).
32. www.ehardware.co.uk/acatalog/History_of_Door_Handles.html)

APPENDIX

Appendix 1

Ingredient of Media

1. Nutrient Agar:

Formula and preparation	gram/litter
Lab-lemco powder.....	1.0
Yeast extract.....	2.0
Peptone.....	5.0
Sodium chloride.....	5.0
Agar.....	15.0

2. MacConky agar:

Peptone.....	20.0
Lactose.....	10.0
Bile salt.....	5.0
Sodium chloride.....	5.0
Neutral red.....	0.075
Agar.....	12.0

3. Blood agar:

Nutrient agar broth.....	500ml
Sterile defibrinated blood.....	25ml

Appendix 2

Biochemical Test Medium and Reagent:

1.Kligler Iron agar Medium [KIA] : gram/litter

Oxoid dehydrated medium formula

Lab lemco powder	3.0
Yeast extract	3.0
Peptone.....	20.0
Sodium chloride.....	5.0
Lactose.....	10.0
Dextrose [glucose].....	1.0
Ferric citrate.....	0.3
Sodium thiosulphate.....	0.3

Phenol red.....	0.05
Agar.....	12.0
Distilled water	1000ml

2. Citrate Agar Medium :(Simmons Citrate Agar) gram/litter

Magnesium sulphate.....	0.200
Ammonium dihydrogen phosphate.....	1.000
Dipotassium phosphate.....	1.000
Sodium citrate	2.000
Sodium chloride.....	5.000
Bromothymol blue.....	0.080
Agar.....	15.000
Distilled water	1000ml
Final PH [at 25°C].....	6.8±0.2

3. Indole Test Medium:

A-Tryptone broth: gram/litter

Tryptone.....	10.0g
Sodium chloride.....	5.0g
Distilled water.....	1000ml

B-Kovács reagent:

Amyl or isoamyl alcohol, reagent grade	150.0 ml
(butyl alcohol may be substituted)	
p-dimethylaminobenzaldehyde (DMAB).....	10.0 g
HCl (concentrated).....	50.0 ml

4. Urease Test Medium:

Peptone.....	1g
Dextrose.....	1g
Sodium chloride.....	5g
Potassium phosphate monobasic.....	2g
Urea.....	20g
Phenol red.....	0.012g

5. Oxidase Test: [cytochrome oxidase]

Freshly prepared 10 g/l of Tetramethyl-p-phenylenediaminedihydrochloride.

Appendix 3

Gram Stain Reagent:

1. Sodium Chloride, 8.5g/l [0.85%w/v] :

Sodium chloride.....	8.5g
Distilled water.....	1000ml

2. Crystal Violet :

Crystal violet.....	20g
Ammonium oxalate.....	9g
Ethanol or Methanol, absolute.....	75ml
Distilled water.....	1000ml

3. Lugol's Iodine Solution:

Potassium iodide.....	20g
Iodine.....	10g
Distilled water.....	1000ml

4. Acetone-alcohol decolorizer :

Acetone.....	500ml
Ethanol or Methanol, absolute.....	475ml
Distilled water.....	25ml

5. Safranin :

Safranin O.....	2.5g
Ethanol.....	100ml
Distilled water.....	90ml