

Journal of Advances in Microbiology Research



E-ISSN: 2709-944X
P-ISSN: 2709-9431
JRM 2022; 3(2): 100-106
© 2022 JAMR

www.microbiojournal.com

Received: 07-08-2022

Accepted: 08-09-2022

Iyevhobu Kenneth O

¹ Department of Public Health, National Open University of Nigeria, Uromi Community Study Centre, Uromi, Edo State, Nigeria

² Lassa fever Enable Study CEPI/ISTH Irrua, Edo State, Nigeria

³ St. Kenny Research Consult, Ekpoma, Edo State, Nigeria

Omolumen Lucky E

Department of Chemical Pathology, Faculty of Medical Laboratory Science, Ambrose Alli University, Ekpoma, Edo State, Nigeria

Ken-Iyevhobu Benedicta A

St. Kenny Research Consult, Ekpoma, Edo State, Nigeria

Oseni David I

¹ St. Kenny Research Consult, Ekpoma, Edo State, Nigeria

² Department of Chemical Pathology, Faculty of Medical Laboratory Science, Ambrose Alli University, Ekpoma, Edo State, Nigeria

Correspondence Author;

Iyevhobu Kenneth O

¹ Department of Public Health, National Open University of Nigeria, Uromi Community Study Centre, Uromi, Edo State, Nigeria

² Lassa fever Enable Study CEPI/ISTH Irrua, Edo State, Nigeria

³ St. Kenny Research Consult, Ekpoma, Edo State, Nigeria

Isolation of bacteria from used handkerchiefs among students

Iyevhobu Kenneth O, Omolumen Lucky E, Ken-Iyevhobu Benedicta A and Oseni David I

Abstract

Bacteria in handkerchiefs can be a reflection of the environment and pathological condition of the individual using the handkerchief. The aim of this study is to isolate bacteria from used handkerchiefs among male and female students of Ambrose Alli University, Ekpoma. Using the streak plate method fifty (50) handkerchiefs from 20 males and 30 females were randomly selected and used for the study. The bacteria isolated were; *Staphylococcus aureus*, *E. coli*, *Proteus* sp, *Klebsiella* sp and *Pseudomonas* sp. *Staphylococcus aureus* had the highest prevalence in the study 57 (48.31%), followed by *Pseudomonas* sp 20 (16.95%), *Klebsiella* sp 19 (16.10%), *Proteus* sp 13 (11.02%) and the least prevalent is *E. coli* 9 (7.63%). In the assessment of organisms isolated from handkerchiefs according to gender, from male handkerchiefs studied, a total of 41 organisms were isolated, in the female handkerchiefs studied, a total of 69 bacteria were isolated. The isolation of these organisms were attributed to poor hand washing, followed by wiping of the dirty hands and face with the handkerchief.

Keywords: Handkerchief, hand hygiene, students, microbes, male, female

1. Introduction

A handkerchief (also called a hankie or, historically, a hand kercher) is a form of a kerchief or bandanna, typically a hemmed square of thin fabric or paper which can be carried in the pocket or handbag, and which is intended for personal hygiene purposes such as wiping one's hands or face, or blowing one's nose. A handkerchief is also sometimes used as a purely decorative accessory in a suit pocket, it is then called a pocket square. It is also an important accessory in many folkdances in many regions like the Balkans and the Middle East; an example of a folkdance using handkerchiefs is Kalamatianos (Hamam, 2014) [10].

Microorganisms are ubiquitous and are found in almost every area around human bodies. Some are specifically found in certain regions of the body as a normal flora where they live as commensals with man. This association is important in protecting the body against other infectious diseases. Each area of the body surface acquires a characteristic flora of organisms well adapted to growth at that particular environment. These residents (normal flora) tend to suppress the intruders either by competition for space and food supply or by production of metabolites that are antagonistic to the survival of the intruder. These residents could be dislodged from their environment when sneezing, coughing, belching, yawning or could be destroyed by regular use of antiseptic soaps or creams on the body surfaces. Handkerchiefs often used for wiping face, closing of the mouth and nose when expressing these reflex activities, therefore constitute an abode for bacteria. Furthermore, bacteria found in handkerchiefs could differ from one individual to another as the bacteria found could be a reflection of the environment and pathological conditions of the individual using the handkerchief. It is generally accepted that the transmission of bacteria and other microorganisms is more likely to occur from wet skin than from dry skin (Gould 1994) [9]. This happens partly because of the ease of water transfer from one surface to another and partly because microorganisms prefer a damp environment and, therefore, may be in a better physiological state to colonize touched surfaces. The amount of residual water left on the hands of users after drying is directly related to the number of bacteria that are transferred by contact: the greater the amount, the more bacteria (Patrick *et al.*, 1997) [23].

Hand hygiene is a fundamental component for controlling the spread of infection (Low bury *et al.*, 1970; Larson, 1981) [17, 14]. Promotion of improved hand hygiene is recognized as an important public health measure.

There is much emphasis on the correct method for hand washing, but less so concerning the options for drying hands. Evidence suggests that efficiency of hand drying is important in the prevention of the transfer of microorganisms from person to person or to the environment (Patrick *et al.*, 1997) [23]. Hands may be a mode of transmission of pathogenic bacteria. Many studies, some of considerable breadth and depth, have centered around the microbial flora of the skin. Relatively few investigations have analyzed the part that hand drying agents and processes play in the cause of human disease.

The normal human skin is colonized by huge numbers of microorganism that live as commensals on its surface (Hay and Adriaans, 2004) [12]. At times microorganism not normally found there may colonize the epidermis and lead rapidly to disease. Apart from these pathogenic organisms, a wide range of organism land fortuitously on the skin, but are unable to multiply. Organisms not normally considered as skin flora may sometimes colonize it (Hay and Adriaans, 2004) [12]. When the skin is inflamed or abnormal, it is often difficult to determine whether an organism isolated is causing or contributing to the observed pathology. If the skin is damaged or the immune status of the subject impaired, microorganisms usually regarded as non-pathogenic in body surface may assume the role of opportunist pathogens. Within a given species, there are also strain differences in virulence (Hay and Adriaans, 2004) [12]. Some strains have a particular tendency to cause disease, perhaps due to greater adherence to epithelial cells or enzyme production (Hay and Adriaans, 2004) [12]. There are some studies investigating skin flora on healthy and ill population to find out any possible relation between disease and microbial flora of skin (Zell *et al.*, 2008) [26]. In this study, we planned to study the species of microorganism colonization on the handkerchiefs among male and female in Ekpoma (Berlau *et al.*, 1999) [4].

Hands may be a mode of transmission of pathogenic microbes. Many studies, some of considerable breadth and depth, have centered on the microbial flora of the skin. Relatively few investigations have analyzed the part that hand drying agents and processes play in the cause of human disease. Disregarding the types of textile towel where users dry their hands on the same area of material as previous users and which have been condemned on hygiene grounds for many years, the three main hand drying methods available in public washrooms have until fairly recently been: paper towels, continuous roller towels (where a fresh area of towel is available for each user) and warm air dryers.

Microbes found on the skin are usually regarded as pathogens, potential pathogens or innocuous symbiotic organisms. Advances in microbiology and immunology are revising our understanding of the molecular mechanisms of microbial virulence and the specific events involved in the host-microbe interaction. Current data contradict some historical classifications of cutaneous microbiota and suggest that these organisms may protect the host, defining them not as simple symbiotic microbes but rather as mutualistic. This review will summarize current information on bacterial skin flora including *Staphylococcus*, *Corynebacterium*, *Propioni-bacterium*, *Streptococcus* and *Pseudomonas*. Specifically, the review will discuss our current understanding of the cutaneous microbiota as well as shifting paradigms in the interpretation of the roles microbes

play in skin health and disease and also its transmission in public places.

Following Semmelweis's observations on the effect of hand washing on the incidence of puerperal fever in a maternity ward in the 19th Century, good hand hygiene has been recognized as an important factor in controlling the spread of infectious disease and, more recently, antibiotic-resistant bacteria in hospitals and in the community. Methods for hand drying vary widely and include paper or cloth towels, warm air dryers or jet air dryers either singly or in combination. Drying with towels may remove remaining microorganisms through friction, while moisture is wicked away into the absorbent material. Warm air dryers evaporate moisture and remove some microorganisms during hand rubbing, although this process may take too long for efficient use, with hands consequently remaining damp (Larson, 1981) [14]. The selection of drying method may depend upon a number of factors including practicality, space availability, cost, or personal preference. Infection prevention considerations may influence the choice of hand-drying method, but the evidence base is weak to make informed decisions (Patrick *et al.*, 1997) [23]. The aim of this study is to enumerate and identify bacteria on used handkerchiefs among male and female in Ekpoma.

2. Materials and Methods

2.1 Study population / sample size

Fifty (50) handkerchiefs from 20 male and 30 female in Ekpoma were randomly used and also 10 control handkerchiefs were also used for this study. Samples were taken by cotton swab from the handkerchiefs of different individuals. Swabs were cultured on different Nutrient agar and incubated at 37°C and sub-cultured into relevant agars. Moreover, different biochemical tests were applied; catalase test, coagulase test and oxidase test. Microorganisms were recognized on the basis of macroscopic, microscopic and differential biochemical tests.

2.2 Study design

This study designed to enumerate and identify bacteria on used handkerchiefs by male and female in Ekpoma. Specimens such as handkerchiefs swab were collected and analyzed in the laboratory using standard methods. Results were presented in tables.

2.3 Specimen collection

Fifty (50) handkerchiefs were used for this study with one swab used for each handkerchief from both male and female. Samples were taken by cotton swab. Swabs were cultured on Nutrient agar, Manitol salt agar, Chocolate agar and Maconkey agar and incubated at 37 °C. The different biochemical tests were applied; catalase test, Indole test, Oxidase test, Citrate test and Coagulase test. Microorganisms were identified on the basis of macroscopic, microscopic and differential tests.

2.4 Sample analysis/methods

The sample analysis was carried out for bacteriological examination in the microbiology laboratory of the Department of Microbiology, Faculty of Life Sciences, Ambrose Alli University, Ekpoma, Edo State.

2.4.1 Bacteriological examination Culture of handkerchief swab

The swab stick was inoculated on each plate of Nutrient and Blood agar by making a primary inoculum on a small area of the agar plate and then streaked out. The growth from the nutrient agar was then sub-cultured into MacConkey agar. The inoculated media was incubated aerobically at 37 °C for 24 hours. Those inoculated on chocolate agar were incubated anaerobically. Identification of bacteria was done by carrying out biochemical tests (Cheesbrough, 2006) [7].

2.5 Data analysis

The data generated was analyzed with simple percentage and presented in tables.

3. Results

This study was carried out for the isolation of bacteria on used handkerchiefs among male and female in Ekpoma. This study is limited to the isolation and identification of bacteria on handkerchiefs used by male and female in Ekpoma and also about ten (10) handkerchiefs were bought which served as control.

In this study, twenty (20) male handkerchiefs were sampled while thirty (30) female handkerchiefs were sampled and ten (10). Out of the 60 samples examined, 57 were positive to bacterial infection which includes; 20 male, 30 female and 7 from control. Five bacterial species were predominantly and frequently isolated from the handkerchiefs sampled in this study. The bacteria isolated are; *Staphylococcus aureus*, *E. coli*, *Proteus* sp, *Klebsiella* sp and *Pseudomonas* sp (Table 1). *Staphylococcus aureus* has the highest prevalence in the study 57 (48.31%), followed by *Pseudomonas* sp 20 (16.95%), *Klebsiella* sp 19 (16.10%), *Proteus* sp 13 (11.02%) and the least prevalent is *E. coli* 9 (7.63%).

In the assessment of organisms isolated from handkerchiefs according to gender (Table 2), from male handkerchiefs studied, a total of 41 organisms were isolated, in the female handkerchiefs studied, a total of 69 bacteria were isolated while in the control samples, a total of 8 organisms were isolated.

The result of the Cultural Characteristics and Biochemical Analysis Bacterial Isolates conducted on the bacteria colonies during fermentation and storage period is shown in table 3. The organisms isolated, some were gram positive and others were gram negative. The organisms on the agar plate showed circular to irregular shape, they were bright yellow, creamy, grey, translucent blue and translucent creamy in colour with surface appearances (consistency)

which were moist, mucoid and dry.

The antibiotic susceptibility patterns of the various isolates for male samples were read using their zones of inhibitions on the sensitivity culture plates, which shows that Ciprofloxacin, Gentamycin, Streptomycin and Refampicin were the most sensitive antibiotics against the gram-positive bacteria isolates (*Staphylococcus aureus*) while other drugs were found to be intermediate and resistant. The gram-negative organisms (*Pseudomonas* sp, *Escherichia coli*, *Proteus* sp and *Klebsiella* sp) were more sensitive to Augmentin and Gentamycin, while Ofloxacin, Peflacin, Ciprofloxacin, Septrin and Ampicillin were intermediate while the other drugs were resistant (Table 4).

The antibiotic susceptibility patterns of the various isolates for Female were read using their zones of inhibitions on the sensitivity culture plates, which shows that Ciprofloxacin, Gentamycin and Streptomycin were the most sensitive antibiotics against the gram-positive bacteria isolates (*Staphylococcus aureus*) while other drugs were found to be intermediate and resistant. The gram-negative organisms (*Pseudomonas* sp, *Klebsiella* sp, *Escherichia coli* and *Proteus* sp) were more sensitive to Augmentin, Ofloxacin and Gentamycin while, Peflacin, Ciprofloxacin, Septrin and Ampicillin were intermediate while the other drugs were resistant (Table 5).

Table 1: Organisms isolated from the study and percentage prevalence

Organisms Isolated	No. isolated	Percentage prevalence (%)
<i>Pseudomonas</i> sp	20	16.95
<i>Klebsiella</i> sp	19	16.10
<i>Staphylococcus aureus</i>	57	48.31
<i>E. coli</i>	9	7.63
<i>Proteus</i> sp	13	11.02
TOTAL	118	100

Table 2: Organisms isolated from the study according to gender

Organisms Isolated	Male N = 20	Female N = 30	Control N = 10
<i>Pseudomonas</i> sp	8	11	1
<i>Klebsiella</i> sp	6	13	0
<i>Staphylococcus aureus</i>	20	30	7
<i>E. coli</i>	3	6	0
<i>Proteus</i> sp	4	9	0
Total	41	69	8

Table 3: Cultural Characteristics and Biochemical Analysis Bacterial of the Isolates

Organisms Isolated	Cultural characteristics					Biochemical analysis							Organism
	Gram	Shape	Shape of colony	Consistency	Colour	Motility	Catalase	Oxidase	Indole	Coagulase	Citrate	Urease	
B1	-	Rod	Circular	Mucoid	Translucent cream	-	+	-	-	-	+	+	<i>Klebsiella</i> sp
B2	-	Rod	Irregular	Moist	Light green	+	+	+	-	-	+	-	<i>Pseudomonas</i> sp
B3	+	Cocci in cluster	Circular	Moist	Golden yellow	-	+	-	-	+	+	+	<i>Staphylococcus aureus</i>
B4	-	Rod	Irregular	Moist	Bright yellow	+	+	-	+	-	-	-	<i>Escherichia coli</i>
B5	-	Rod	Irregular	Mucoid	Colourless	+	+	-	+	-	+	+	<i>Proteus</i> sp

KEY

+ = Positive

- = Negative

B = Bacteria

Table 4: Antibiotic Susceptibility Patterns of Bacterial Isolates from Male

Antibiotics	Mircoorganisms			
	<i>Staph aureus</i>			
Gram Positive Disc	<i>Staph aureus</i>			
Ciprofloxacin	S			
Norfloxacin	R			
Gentamycin	S			
Amoxil	R			
Streptomycin	S			
Refampicin	S			
Erythromycin	R			
Chloramphanicol	S			
Ampiclox	S			
Levofloxacin	R			
Gram Negative Disc	<i>E. coli</i>	<i>Klebsiella</i> sp	<i>Pseudomonas</i> sp	<i>Proteus</i> sp
Ofloxacin	R	S	S	S
Peflacin	S	R	S	S
Ciprofloxacin	R	S	S	S
Augmentin	S	S	S	R
Gentamycin	S	S	S	S
Streptomycin	R	S	R	S
Ceporex	R	R	R	S
Naliolixic acid	R	R	R	R
Septin	S	S	S	R
Ampicillin	R	S	S	S

KEY: S = Sensitive (>3mm) R = Resistance (<3mm)

Table 5: Antibiotic Susceptibility Patterns of Bacterial Isolates from Female

Antibiotics	Mircoorganisms			
	<i>Staph aureus</i>			
Gram Positive Disc	<i>Staph aureus</i>			
Ciprofloxacin	S			
Norfloxacin	R			
Gentamycin	S			
Amoxil	S			
Streptomycin	S			
Refampicin	R			
Erythromycin	R			
Chloramphanicol	S			
Ampiclox	S			
Levofloxacin	R			
Gram Negative Disc	<i>E. coli</i>	<i>Proteus</i> sp	<i>Pseudomonas</i> sp	<i>Klebsiella</i> sp
Ofloxacin	R	S	S	S
Peflacin	R	R	S	S
Ciprofloxacin	R	S	R	S
Augmentin	S	S	S	R
Gentamycin	S	S	S	S
Streptomycin	S	S	R	R
Ceporex	R	R	R	S
Naliolixic acid	R	R	R	R
Septin	S	R	S	R
Ampicillin	R	S	S	S

Key: S = Sensitive (>3mm) R = Resistance (<3mm)

4. Discussion

Unhygienic handling and use of the handkerchief contributed to the observed gross contamination with bacteria. Result of the current study clearly revealed limitation of experience on hygienic handling of handkerchief and poor sanitization strategies tracked to diminish pathogenic bacteria. In the current report, diverse microorganisms including probably pathogenic microbes were encountered in the handkerchiefs studied. Among these microbes are *Staphylococcus aureus* with percentage prevalence of 48.31%, *E. coli* with percentage prevalence of 7.63%, *Proteus* sp with percentage prevalence of 11.02%, *Klebsiella* sp with percentage prevalence of 16.10% and *Pseudomonas* sp with percentage prevalence of 16.95%

(Table 1). As reported earlier (Olsen *et al.*, 2000) [22], microorganisms, together with pathogenic species, frequently exist in handkerchiefs and hand towels. The results showed that brand new handkerchiefs picked from the shelves tested positive for microbes. An average of 8 organisms' which were 2 bacteria (*Staphylococcus aureus* and *Pseudomonas* sp) were present on unused handkerchiefs when the bacteria culture solution was plated. The main source of microbes on new handkerchiefs could have been human skin of merchandisers or customers during the shopping activities and possibly wind. Contamination during experimentation was minimal because sterile surgical gloves were used to handle the towels while conducting experiments. Human skin harbours approximately 1×10^5

colony forming units per cm² of *Staphylococcus aureus* (Spicer, 1959) [24]. The presence of microbes on the handkerchiefs indicated that handkerchiefs either did not contain antimicrobial finishes or were treated with antimicrobial finishes that could not eliminate all microbes. However antimicrobial finishes can actually be damaging to health as their overuse can create a drug-resistant microbes that will not respond to any prescription available because the microbes such as bacteria mutate to create “super germs” (Ministry of Health, 1997) [18].

Staphylococcus aureus is relatively widespread in the environment but is found mainly on the skin and mucous membranes of animals. The organism is a member of the normal microbial flora of the human skin and is found in the nasopharynx of 20–30% of adults at any one time. Staphylococci are occasionally detected in the gastrointestinal tract and can be detected in sewage. *Staphylococcus aureus* can be released by human contact into water environments such as swimming pools, spa pools and other recreational waters which can find their way into this handkerchief when used after activities in these environments. It has also been detected in drinking-water supplies (Antai, 1987) [2]. Hand contact is by far the most common route of transmission. Inadequate hygiene can lead to contamination of food. Foods such as ham, poultry and potato and egg salads kept at room or higher temperature offer an ideal environment for the multiplication of *S. aureus* and the release of toxins. The consumption of foods containing *S. aureus* toxins can lead to enterotoxin food poisoning within a few hours (LeChevallier and Seidler, 1980) [5]. The presence of *Staphylococcus aureus* on handkerchiefs means that handkerchiefs can be sources of food poisoning since *Staphylococcus aureus* is pathogenic. This bacterial species causes boils and localized swollen areas of tissue. It can also lead to blood stream invasion, fever and general malaise (Chakwana and Nkiwane, 2014) [6]. The fact that this bacteria was found on handkerchiefs, means that handkerchiefs are suitable vehicles for staphylococcal food poisoning since their degree of bacterial contamination.

The *Pseudomonas* cells cultured in non-selective nutrient agar appeared pink on the optical microscope when observed after the Gram stain reaction. The presence of *Pseudomonas* sp on handkerchiefs shows that handkerchiefs can be vehicles for the transmission of disease (Kimberly, 2003) [13]. *Pseudomonas* sp is an opportunistic pathogen which causes toe mob infection characterized by thick white scaling areas between toes (Kimberly, 2003) [13]. It also causes Green nail syndrome which is the greenish coloration of nail plates (Kimberly, 2003) [13]. *Pseudomonas* sp is opportunistic human pathogen. They are “opportunistic” because it seldom infects healthy individuals. They are pathogenic if it enters the body via wounds, abscesses and burns (Blomfield *et al.*, 2011) [5]. The handkerchiefs users with wounds such as minor cuts are therefore susceptible to toe mob infection and green syndrome since *Pseudomonas* sp cells were found in the handkerchiefs. The towel users are vulnerable when they come in contact with the bacteria during hand and face drying. *Pseudomonas* sp can cause a range of infections but rarely causes serious illness in healthy individuals without some predisposing factor. It predominantly colonizes damaged sites such as burn and surgical wounds, the respiratory tract of people with underlying disease and physically damaged eyes (Bartram, 2003). From these sites, it may invade the body, causing

destructive lesions or septicaemia and meningitis. Cystic fibrosis and immunocompromised patients are prone to colonization with *P. aeruginosa*, which may lead to serious progressive pulmonary infections (de Victorica and Galván, 2001) [8]. Water-related folliculitis and ear infections are associated with warm, moist environments such as swimming pools and spas. Many strains are resistant to a range of antimicrobial agents, which can increase the significance of the organism in hospital settings. *Pseudomonas aeruginosa* is a common environmental organism and can be found in faeces, soil, water and sewage. It can multiply in water environments and also on the surface of suitable organic materials in contact with water. *Pseudomonas aeruginosa* is a recognized cause of hospital-acquired infections with potentially serious complications. It has been isolated from a range of moist environments such as sinks, water baths, hot water systems, showers and spa pools (Hardalo and Edberg, 1997) [11].

The presence of *Escherichia coli* on handkerchiefs means that handkerchiefs can be sources of food poisoning since *Escherichia coli* is pathogenic. This bacterial species causes gastroenteritis which is an inflammation of the stomach and intestines and causing vomiting and diarrhoea (Chakwana and Nkiwane, 2014) [6]. Members of *Escherichia coli* are almost universal inhabitants of the intestinal tract of humans and they may play a nutritional role in the intestinal tract by synthesizing vitamins, particularly vitamin K (Moyo and Baudi, 2004) [19]. Though *Escherichia coli* species are rarely pathogenic they have shown some implications in diarrhoea in infants and urinary tracts in older people (Moyo and Baudi, 2004) [19]. This shows that handkerchiefs are suitable vehicles for *E. coli* in skin and wound since their degree of bacterial contamination was found to be high. *Escherichia coli* is present in large numbers in the normal intestinal flora of humans and animals, where it generally causes no harm (O'Connor, 2002) [21]. However, in other parts of the body, *E. coli* can cause serious disease, such as urinary tract infections, bacteraemia and meningitis. A limited number of enteropathogenic strains can cause acute diarrhoea (Nataro and Kaper, 1998; O'Connor, 2002) [20-21]. As few as 100 Enterohaemorrhagic *Escherichia coli* (EHEC) organisms can cause infection. Enterotoxigenic *Escherichia coli* (ETEC) produces heat-labile or heat-stable *E. coli* enterotoxin, or both toxins simultaneously, and is an important cause of diarrhoea in developing countries, especially in young children. Symptoms of ETEC infection include mild watery diarrhoea, abdominal cramps, nausea and headache. Infection with enteropathogenic *Escherichia coli* (EPEC) has been associated with severe, chronic, non-bloody diarrhoea, vomiting and fever in infants. EPEC infections are rare in developed countries, but occur commonly in developing countries, with infants presenting with malnutrition, weight loss and growth retardation. The pathogens have been detected in a variety of water environments. Infection is associated with person-to-person transmission, contact with animals, food and consumption of contaminated water. Person-to-person transmissions are particularly prevalent in communities where there is close contact between individuals, such as nursing homes and day care centres (Nataro and Kaper, 1998) [20].

Michael *et al.*, (2005) described *Proteus* sp as an organism that occur in the intestine of human and in a wide variety of animals, in polluted water, soil and they are opportunistic pathogens. This might have been incriminated in the handkerchief after eating, use of the handkerchief in cleaning classroom chairs before sitting, some students spread their handkerchief to sit on or used to clean hands

after playing with pets.

Klebsiella sp have been identified as colonizing hospital patients, where spread is associated with the frequent handling of patients (e.g., in intensive care units). Patients at highest risk are those with impaired immune systems, such as the elderly or very young, patients with burns or excessive wounds, those undergoing immunosuppressive therapy or those with HIV/AIDS infection (Ainsworth, 2004) ^[1]. Colonization may lead to invasive infections. On rare occasions, *Klebsiella* sp, notably *K. pneumoniae* and *K. oxytoca*, may cause serious infections, such as destructive pneumonia. *Klebsiella* sp are natural inhabitants of many water environments, and they may multiply to high numbers in waters rich in nutrients, such as pulp mill wastes, textile finishing plants and sugar-cane processing operations. In drinking-water distribution systems, they are known to colonize washers in taps. *Klebsiella* can cause nosocomial infections, and contaminated water and aerosols may be a potential source of the organisms in hospital environments and other health care facilities (Bartram, 2003) ^[3].

The results show that the level of microbial contamination was higher in females than males. The ability of the handkerchiefs to shelter bacteria increased with time of their usage even though they were washed daily. This could have been due to increased moisture conditions. Dust could have accumulated between threads during sun drying in an open environment. Drying in warm and humid open environments accelerate microbial growth as indicated by Rodger *et al.* (1979) ^[27]. Although literature has indicated that microbes can be destroyed by the sun's ultraviolet (UV) rays, the requirement will be that the environment, in which the towel is dried will be exposed (Liam and Hudson, 2004). In this research the isolation of bacteria was attributed to poor hand washing, followed by wiping of the dirty hands and face with the handkerchief as Ekpoma is known to be a dusty town during the dry and also having bad road network.

5. Conclusion

In conclusion, handkerchiefs could be hazardous to health if they are not laundered on a daily basis as they could harbour different types of microbes. There is need to have a handkerchief for each specific purpose/occasion as evidenced by the results that showed. Disinfecting handkerchiefs every other day with bleaching agents such as sodium hypochlorite, and rinsing them thoroughly over and over again would reduce the risk of poisoning due to chemicals and at the same time prevent the handkerchiefs from becoming shelter to pathogenic microorganisms. Bleaching handkerchiefs however would lead to their quick disintegration and the need to purchase new ones frequently. Individuals preferred using this method of laundering even though it led to the frequent purchasing of towels, as compared to home laundering which would not eliminate microbes completely but this applies to the financially enabled individuals. Using handkerchiefs manufactured from synthetic micro fibres could be another solution to reduce transfer of organisms because, micro fibres have been engineered to possess properties such as good liquid absorption and wicking, easy wash-ability and quick drying, properties desired in kitchen towels (Chakwana and Nkiwane, 2014) ^[6]. Being synthetic micro fibres, are not palatable to microbes and therefore do not provide a breeding environment for pathogenic microbes.

6. Acknowledgement

The authors would like to thank all the Laboratory and

technical staffs of St Kenny Research Consult, Edo State for their excellent assistance and for providing medical writing support/editorial support in accordance with Good Publication Practice (GPP3) guidelines.

7. Conflict of Interest

The authors declare no conflicts of interest. The authors alone are responsible for the content and the writing of the paper.

8. Funding

This research did not receive any grant from funding agencies in the public, commercial, or not-for-profit sectors.

10. References

- Ainsworth R. Safe, piped water: Managing microbial water quality in piped distribution systems. IWA Publishing, London, for the World Health Organization, Geneva; c2004. p. 45-52.
- Antai SP. Incidence of Staphylococcus aureus, coliforms and antibiotic-resistant strains of Escherichia coli in rural water supplies in Port Harcourt. Journal of Applied Bacteriology. 1987;62:371-375.
- Bartram J. Heterotrophic plate counts and drinking-water safety: the significance of HPCs for water quality and human health. WHO Emerging Issues in Water and Infectious Disease Series. London, IWA Publishing; c2003. p. 4-8.
- Berlau J, Aucken H, Malnicl IT. Distribution of Acinetobacter species on skin of healthy humans. Eur J Clin Microbiol. 1999;18(3):179-83.
- Blomfield F, Martin E, Carlo S, Kumar JN, Elizabeth AS. The Infection Risks Associated with Clothing and Household Linens in Home and Everyday Life Settings and the role of Laundry. International Scientific forum on Home Hygiene (IFH) Ed; c2011. p. 67-71.
- Chakwana C, Nkiwane LC. Development of a Low Cost Re-usable Microfibre Sanitary Pad. Textiles and Light Industrial Science and Technology; c2014. p. 13-17.
- Cheesbrough M. Medical laboratory manual for tropical countries. London, Publishing; c2006. p. 200-234.
- De Victorica J, Galván M. *Pseudomonas aeruginosa* as an indicator of health risk in water for human consumption. Water Science and Technology. 2001;43:49-52.
- Gould D. The significance of hand drying in the prevention of infection. Nursing Times. 1994;90:33-36.
- Hamam J. History of the Towel; c2014. <http://www.towelusage.org/history/2014/09/usda>. Retrieved 28 August 2018.
- Hardalo C, Edberg SC. *Pseudomonas aeruginosa*: Assessment of risk from drinking-water. Critical Reviews in Microbiology. 1997;23:47-75.
- Hay RJ, Adriaans BM. Bacterial infections. In: Burns T, Breathnach S, Cox Neil, Griffiths CT, Rook's textbook of Dermatology. Black Well Science Ltd, USA; c2004. p. 271-276.
- Kimberly C, Species BEIOB. Alberta Canada University of Alberta Edmonton. McGraw-Hill Companies, Boston, USA; c2003. p. 56-61.
- Larson EL. Persistent carriage of gram-negative bacteria on hands. Am J Infect control. 1981;9:112e-119e.

15. LeChevallier MW, Seidler RJ. Staphylococcus aureus in rural drinking-water. Applied and Environmental Microbiology. 1980;39:739-742.
16. Liam SH, Hudson SM. Application of Fibre-Reactive Chitosan Derivative to Cotton Fabric as an Antimicrobial Textile Finish, Carbohydrated Polymers; c2004. p. 54-60.
17. Lowbury EJ, Thom BT, Lilly HA, Babb JR, Whittall K. Sources of infection with *Pseudomonas aeruginosa* in patients with tracheostomy. J Med Microbiol. 1970;3:39e-56e.
18. Ministry of Health, Governing Microbial Standards for Foodstuffs and Related Matters, In: HEALTH (Ed.); c1997. p. 45-51.
19. Moyo DZ, Baudi I. A Bacteriological Assessment of the Cleaning and Disinfection Efficacy at the Midlands State University Canteen, Zimbabwe. Pakistani Journal of Biological Science. 2004;7:1996-2001.
20. Nataro JP, Kaper JB. Diarrheagenic Escherichia coli. Clinical Microbiology Reviews. 1998;11:142-201.
21. O'Connor DR. Report of the Walkerton Inquiry: The events of May 2000 and related issues. Part 1: A summary. Toronto, Ontario, Ontario Ministry of the Attorney General, Queen's Printer for Ontario; c2002. p. 78-95.
22. Olsen S, MacKinnon L, Goulding J, Bean N, Slutsker L. Surveillance for food borne disease outbreaks, 1993–1997, Surveillance Summaries. 2000;49:1-62.
23. Patrick DR, Findon G, Miller TE. Residual moisture determines the level of touch-contact-associated bacterial transfer following hand washing. Epidemiology and Infection. 1997;119:319-325.
24. Spicer CC. The Survival of Shigella sonnei on Cotton Threads. Journal of Hygiene. 1959;57:210-215.
25. World Gazzetter. Population of Cities, news, divisions; c2007. <http://worldgazzetter.com/ng.php>. Retrieved on 23/10/2016.
26. Zell CH, Resch M, Rosenstein R. Characterization of toxin production of coagulase-negative Staphylococci isolated from food and starter cultures. Int J Food Microbiol. 2008;127:246-51.
27. Rodger SA, Holmes IH. Comparison of the genomes of simian, bovine, and human rotaviruses by gel electrophoresis and detection of genomic variation among bovine isolates. Journal of Virology. 1979 Jun;30(3):839-46.

How to Cite This Article

Iyevhobu KO, Omolumen LE, Ken-Iyevhobu A, Oseni DI. Isolation of bacteria from used handkerchiefs among students. Journal of Advances in Microbiology Research. 2022;3(2):100-106.

Creative Commons (CC) License

This is an open access journal, and articles are distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International (CC BY-NC-SA 4.0) License, which allows others to remix, tweak, and build upon the work non-commercially, as long as appropriate credit is given and the new creations are licensed under the identical terms.