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Bacteriological Assessment of Students' Pen

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Authors' contributions

This work was carried out in collaboration between all authors. Authors AI and AK designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author NGE managed the analyses of the study. Author AYM managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Pen, as a writing instrument that is passed from hand to hand, is likely to be contaminated with diseases causing microorganisms especially if handled with unclean hands or kept in dirty surroundings. This study was carried out to investigate the prevalence of bacterial contamination among the pen of students. 50 samples of the pen were collected randomly from students in Unity Secondary School Zungeru, Niger State, Nigeria, under aseptic conditions and were brought into the laboratory for analysis. Cooked meat media, MacConkey, and nutrient agar were used in quantification and isolation respectively. The bacterial were further sub cultured, identified and characterized by growing them on MacConkey and Chocolate agar, observing their morphological characteristic then Gram reaction and biochemical test were used for confirmation. Out of the 50 samples collected, 40(90%) were contaminated with different bacterial species which includes *Streptococci* sp. *Staphylococcus aureus, E. coli, Clostridium* Sp., *Neisseria* Sp., and *Klebsiella* Sp., and $3.2x10^5$, $5.0x10^5$, $2.6x10^5$, $1.8x10^5$, $1.2x10^5$ and $1.6x10^5$ CFU/g respectively, with *Staphylococcus aureus* and *Neisseria* Sp, having the highest and the lowest colony forming unit respectively. However the population density of the bacteria isolates based on the frequency of

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occurrence includes *E. coli* (34%), which was dominant followed by *Staphylococcus aureus* (24%), *Streptococci* Sp. (12%), *Clostridium* Sp. (4%), *Klebsiella* Sp. (4%) and *Neisseria* Sp. (2%). This shows that pen can be a potential format for the transmission of pathogens. Therefore it is important to encourage higher compliance to hand washing and personal hygienic practices.

Keywords: Pen; contamination; students; fomite; personal hygiene.

1. INTRODUCTION

Bacteria constitute a large domain of prokaryotic microorganisms. They are typically few micrometers in length. Bacterial are ubiquitous in nature, adaptable to extremely conditions and can survive wherever they find themselves [1]. The vast majority of the bacteria in the body are rendered harmless by the protective effects of the immune system, though many are beneficial particularly in their normal location in the host (e.g gut flora). However several species of bacteria are pathogenic and cause infectious diseases such as cholera, diarrhea, syphilis, anthrax, leprosy, bubonic plague etc when introduced into foreign locations through contaminated objects, food or water [2].

The contamination of objects could be from several sources like atmosphere, during storage, handling or production [3]. The microorganisms that are mostly isolated from surfaces of objects included members of the genus *Staphylococcus* species*, enterobacteria,* and *bacilli* sp*.* Most objects contaminants are environmental microorganisms and those arising from human skin flora such as *Staphylococcus aureus* [4]. The presence of bacteria on objects is a reflection of the local environmental situation and personal hygiene.

The pen is an instrument made of plastic or metal with a pointed tip used to apply ink to a surface, usually paper, for writing or drawing [5]. The use of pen has been part of mankind since about 3000 BC used for writing on parchment and pen as an object that is passed from hand to hand is likely to be contaminated with diseases causing microorganisms especially if handled with unclean hands or kept in dirty surroundings [6]. Pen, therefore, present a particular risk to public health since communicable diseases spread through contact with fomites [7]. Ogo et al. [8] reported that the source of contaminations and infections could be as a result of poor or negative handling practices like chewing contaminated objects in the mouth or scratching of body parts with its edge or head. Placing pen on dirty surfaces like other objects exposes it to contamination, making it a convenient habitat for pathogens [9]. Ordinarily, the exposure of pen to the atmosphere could bring about contamination, depending on the environment in question. For instance, handling pen with unwashed hand after visiting the toilet has a high tendency of being contaminated with pathogenic bacteria. Other attitudes such as cleaning the ear with pen and under the nails could lead to the possible transfer of infectious dose of pathogenic bacteria from such medium to the pen which could later be transferred to the mouth where it could cause infection or disease on a healthy individual [10].

This study was conducted to determine bacterial contamination of student's pen in Unity Secondary School Zungeru, Wushishi Local Government, Niger State, Nigeria and to identify the most important bacterial species associated with these pens in order to take the necessary remedial measures.

2. MATERIALS AND METHODS

2.1 Sample Collection

Fifty (50) samples of the pen were collected randomly from students of Unity Secondary School Zungeru, Niger state, Government Area of Niger State, Nigeria, on coordinates 9° 48'N 6° 9'E and elevation of 149 m (489 ft) in May 2017. under the aseptic condition and transported to the laboratory for analysis.

2.2 Media Preparation

The media used were Nutrient agar, cooked meat media, MacConkey and Chocolate agar. The media were all prepared according to the manufacturer's instructions and sterilized by autoclaving at 15ibs pressure (121°C) for 15 minutes, they were allowed to cool to about 45°C and 20 ml each was poured into sterile plates according to the method described by Cheesbrough [11].

2.3 Sample Preparation and Inoculation

Each of the pen samples was soaked in sterile cooked meat media in a test tube with vigorous shaking to dislodge the cells into the media. The

suspensions were then serially diluted and transferred into the prepared Nutrient and MacConkey agar media plates using streak plate method and were incubated at 37°c for 18-24 hours aerobically and anaerobically. The colonies were counted using colony counter [11].

2.4 Characterization and Identification of Isolated Bacteria

After isolating pure cultures, the bacterial isolates were then identified and characterized by observing their growth on MacConkey and Chocolate agar for the morphological characteristic. Gram staining reaction and biochemical tests such as catalase test coagulate test and indole test were used to confirm the isolated bacteria as described by Cheesbrough [11].

3. RESULTS

Fig. 1 shows the prevalence of bacteria in student pen, Table 1 shows Identification and biochemical characterization of bacteria isolated from pen while Table 2 The average colony count of bacterial isolated from the pens of student.

4. DISCUSSION

The study revealed the prevalence of bacteria on pen used by students which were not unexpected due to the ubiquitous nature of bacteria and their ability to survive on surfaces according to Pope and co-worker (2006) who demonstrated in their study that bacteria were capable of growing on the surface of objects like currency notes. However, Out of the 50 samples collected 40 (90%) were contaminated with different bacteria genus and species which includes *Streptococci* Sp. *Staphylococcus aureus, E. coli, Clostridium* Sp., *Neisseria* Sp., *and Klebsiella* Sp., and the average colony count were $3.2x10^5$, $5.0x10^5$, $2.6x10^5$, $1.8x10^5$, $1.2x10^5$ and $1.6x10^5$ CFU/g respectively. The viable bacteria count ranged from $1.2x10^5$ to $5.0x10^5$cfu/g With *Staphylococcus aureus* and *Neisseria* Sp. having the highest and the lowest colony forming unit respectively. These bacteria may probably have found their entry to the pens through the skin and hand to hand mechanism since they are normal microbiota of the skin as advanced by Pope et al. [7]. This is a cause for concern since the greater the concentration of bacteria on the object, the longer their ability to survive and these pathogenic bacteria may cause disease in anyone who gets contaminated while using the object according to Reynolds et al. [12].

The bacteria isolates based on the frequency of occurrence include *E. coli* (34%), which was dominant followed by *Staphylococcus aureus* (24%), *Streptococci* Sp. (12%), *Clostridium* Sp. (4%), *Klebsiella* Sp. (4%) and *Neisseria* Sp. (2%). These bacterial isolated shows that pen can be a potential format which is similar to other formats such as currency note in circulation, mobile phone and doorknob [13]. The handling of pen-like other formats could make it easy for the transfer of pathogenic bacteria and thus cross-contamination.

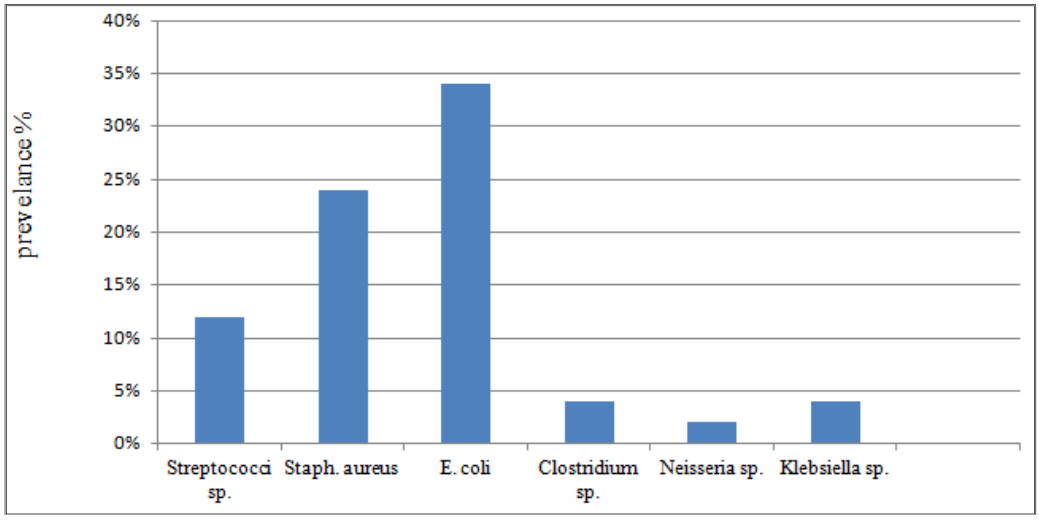

Fig. 1. Shows the prevalence of bacteria in student pen

Table 1. Identification and biochemical characterization of bacteria isolated from pen

Cultural characteristics	Gram reaction	Shape	Arrangement	Catalase	Coagulase	Indole	Mortality	Suggested organism
Discoid colonies	+	Cocci	Chains	+	-	-	-	*Streptococci* species
Round, smooth, raised, glistening, and deep golden colonies	+	Cocci	Clusters	+	+	-	-	*Staphylococcus aureus*
Circular, convex, smooth colonies with distinct edges	-	Bacilli	Singly	-	-	+	+	*E. coli*
Large raised colonies	+	Bacilli	Singly	-	-	-	+	Clostridium *species*
Convex, glistening, elevated mucoid colonies	-	Cocci	Pairs	-	-	-	-	*Neisseria* species
Large and very mucoid colonies	-	Bacilli	Singly	-	-	-	-	*Klebssiella* species

Key: (+) =Positive test, (-) = Negative test

Table 2. The average colony count of bacterial isolated from the pens of student

Organism isolated	Average colony count (CFU/g)
Streptococci species	3.2×10^5
Staphylococcus aureus	5.0×10^5
Escherichia coli	2.6×10^5
Clostridium species	1.8×10^5
Neisseria species	1.2×10^5
Klebsiella species	1.6×10^5

The high prevalence of *E. coli* suggests fecal contamination of these pens which can result in disease outbreak and community-acquired infection. The presence of *Staphylococcus aureus, Streptococci* Sp., and *Clostridium* Sp. confirm the ubiquitous nature of these bacterial and the ability of *Clostridium* spores to resist environmental changes and survive when exposed to an extreme environment with dry heat. However, *Streptococci* sp, *Staphylococcus aureus* and *Neisseria* sp. which are usually found in the nose are often propelled from the respiratory tract into the air when coughing or sneezing [7] and eventually settle on the palm and are transferred to the pen surface.

5. CONCLUSION

The result of this study has shown that pen can be colonized by bacteria and could serve as a potential source of infection or fomite to transmit pathogenic bacteria from one individual to another. Therefore it is important to encourage higher compliance to hand washing, personal hygienic practice and routine surface disinfection of personal items, especially among the students to prevent the spread bacterial infections.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Choi CQ. Microbes thrive in the deepest spot on earth. Live science. Journal of Health System Pharmacology. 2013;15:4. (Accessed 12th Sep., 2017)
2. WHO. Overcoming antimicrobial resistance. World health organization report on Infectious Diseases; 2000. (Accessed 12th Sep., 2017) Available:http://www.who.int/infectious-disease report/2000/index.html
3. Liu LT, Wang DQ, Zheng YX. A survey on the status of bacterial contamination of currency notes in Guangzhou city. International Journal of Recent Scientific Research. 2009;5(9):1533-1540.
4. Xu J, Moore JE, Millar BC. Ribosomal DNA (rDNA). Identification of the culturable bacterial flora on monetary coinage from 17 currencies. Journal of Environmental Health. 2005;67:51-55.
5. Abington H. The Hutchinson Unabridged Encyclopedia with Atlas and Weather guide. Helicon, Credo Reference Web. 2012;76.
6. Umeh E, Juluku JU, Ichor T. Microbial contamination of Naira (Nigerian currency) notes in circulation. Research Journal of Environmental Science. 2007;1:336-339.
7. Pope TM, Ender PT, Woelk WK, Koroscil MA, Koroscil TM. Bacterial Contamination of Paper money. Southern Medical Journal. 2002;95:1408-1410.
8. Ogo NI, Ajayi JA, Ajayi OO, Madukeke A. Eggs and Cysts of parasites contamination on Nigerian currency notes. African Journal of Natural Sciences. 2004;7:40-42.
9. Podhajny MR. How dirty is your money? Paper, Film and Foil Converter (PFFC). Penton Media Inc. 330 N. Wabash, Suite 2300, Chicago, IL., USA. 2004;60611-3998.
10. Ulger F, Esen S, Dilek A, Yanik K, Gunaydin M, Leblebicioglu H. Are we aware of how contaminated our mobile phones are with nosocomial pathogens? Annuals Clinical Microbial Antimicrobial. 2009;8:7.
11. Cheesbrough M. District laboratory practice in tropical countries. Part 2, ELBS, London. 2000;157-234.
12. Reynolds KA, Watt PM, Boone SA & Gerba CP. The occurrence of bacteria and biochemical markers on object surfaces. International Journal of Environmental Health Research. 2005;15: 225-234.
13. Khin NO. Contamination of currency notes with enteric bacterial pathogens, Journal of Diarrhea. 2005;l11(12):30-4.

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