BACTERIA ASSOCIATED WITH HAND-WASHING BOWLS AND WATER IN PUBLIC RESTAURANTS

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ABSTRACT
Aims: To determine the bacterial associated with sharing of hand-washing bowls and water in public eateries. Study Area: This study was performed in six restaurants located around Rivers State University, in Port Harcourt Local Government Area, Rivers State, Nigeria. Methodology: The samples were collected by swabbing the bowl (Nsw) directly, the stored water (Nso), after rinsing of hand (Nri), and after hand-washing (Nwa) and were cultured on nutrient agar incubated at 37°C for 18 to 24 hours. Colony counts were done and the percentages of bacteria isolated were determined. Bacteria isolated were identified using morphological, cultural, chemical and biochemical tests. Results: The percentage occurrences of bacteria isolated from the four sources examined were: Staphylococcus aureus 9 (20.5%), Escherichia coli 11 (25.0%), Bacillus sp 10 (22.7%) and coagulase negative Staphylococci sp 4 (9.1%). The most predominant bacteria isolated were E. coli followed by Klebsiella sp and Bacillus sp, then S. aureus and coagulase negative Staphylococci sp. These bacteria were isolated from all the four sources examined. Conclusion: Normal flora and potentially pathogenic microorganism are associated with the use of same bowl in public eateries. This could serve as a medium of transfer of these bacteria from one person to another.

KEYWORDS: Public restaurant, Normal flora, Hand washing, Water.

1. INTRODUCTION
Lack of proper hygiene could lead to the spread of microorganisms especially bacteria which are potential pathogens. An easy route to transmit pathogens is via direct contact. The hand is a common means of direct contact with inanimate surfaces. In the process of touching surfaces of objects, the hands could pick up infectious agents from the surfaces (Pittet et al., 2000). It has been noted that the hands on the average, touches the body at least eighteen times in an hour. This suggests the high tendency microbes can easily adhere and be transmitted through hands (WHO, 2009).

Eating in a public place especially in most West African countries involves the direct use of fingers and or hands. This is partly due to traditional beliefs or the satisfaction derived from such practice as well as the nature of the food served. In restaurants, it is a common practice to use bowls containing water for washing hands prior to eating. WHO had noted that hand washing does not completely eliminate microbes especially if not done for at least twenty seconds (WHO, 2009). Numerous works had shown that compliance to hand hygiene can reduce the transmission and spread of different pathogens Curtis et al., (2000; Kamf et al., 2009).

Most African countries still uphold eating in the traditional ways. In Nigeria most ethnic groups prefer eating with the hands rather to use cutleries. Using cutleries in some settings may be seen as a difficult task, but even though hands are washed before eating, this is done mostly without soap. Soaps are sometimes used in hand washing after eating food that contains oil (fats) and it is difficult to wash ordinarily with water. In such situations, if the hands and utensils are washed without soap, they may serve as a medium of transferring bacteria. In some restaurants, most traditional foods are eaten with hands, for example gari or fou-fou. There is the possibility of hands, plates and dishing materials becoming vehicles of transmitting bacteria pathogens.

The basins used for hand washing are re-used by other customers for the same purpose. In some cases there is no prior washing of the basin before other customers are served water for washing with it. This practice could lead to the transfer of bacteria especially via the faecal oral route. Diarrhea causing bacteria are commonly transmitted through the faecal-oral route (Curtis, 2009). Hand hygiene has been shown as a significant preventive measure against diarrhea infections (Curis & Cairncross, 2003). It is also a cost effective technique in preventing the transmission of bacteria (Borghi et al., 2002).
Although the practice of hand-washing and the factors that influence hand washing behavior among individuals in communities are complex (Hoguo, 1995).

Washing hands without soap or with soap might be linked with knowledge of the advantages of the practice, availability of water and soap. Soap and water do not only remove pathogenic bacteria mechanically but lethal chemically to some de-contaminants the colonizing flora (Han, 1989; Rotter, 1999). The aim of this research are to determine bacteria associated with washed hands water, the washing hand bowl, the stored water, rinse water after initial washing respectively.

2. MATERIAL AND METHODS

2.1 Study Area
This study was carried out in six restaurants located around Rivers State University, in Port Harcourt Local Government Area, Rivers State, Nigeria. This study was conducted between June-July 2017. Confidentiality of the restaurants identities were maintained by naming them by alphabets.

2.2 Sample Collection
Sterile universal bottles were used to collect water samples used for hand washing by mixing the water in bowl properly and decant about 20 ml and rinsing from the main source of water, which was labeled as ‘source’. Another sample was obtained from the bowl containing water for hand-washing before hand washing and labeled as ‘wash’ and a third sample was taken from the bowl containing water used for rinsing and was labeled ‘rinse’. The wash hand bowl was swabbed with a sterile swab stick and labeled ‘swab’. The collected samples (both the water and the swabs) were transported on ice packs in cooler to the Medical Microbiology Laboratory at the Department of Medical Laboratory Science, Rivers State University for examination.

2.3 Media Preparation and Isolation of Bacteria
Nutrient and MacConkey agar powder were weighed, dissolved and sterilized according to the manufacturer’s instructions. The sterilized molten agar was cooled to about 47°C and 15-20 ml was dispensed into sterile disposable petri dishes. Blood agar was made by adding 5% human blood to nutrient agar. They were allowed to set and stored in refrigerator at 4°C for subsequent uses.

2.4 Inoculation of Media
Commercially purchased swab sticks were used to swab each wash hand basin. Each swab stick was directly used to inoculate blood, nutrient and MacConkey agar plates, and streaked out with a sterile wireloop. The plates were incubated at 37°C and examined for growth after 18-24 hours. A sterile swab stick was inoculated onto blood agar plate to serve as quality control to monitor sterility of the swab sticks and one of the prepared plates was incubated un-inoculated to check the sterility of agar plates. For water samples, 0.01 ml of each sample was cultured by spread plate technique. These were also incubated and examined as above.

2.5 Identification of Isolated Bacteria
Bacteria identification were performed using morphological, Gram’s staining reaction, chemical and biochemical tests such as catalase, coagulase, Indole, and Urease tests (Cheesbrough, 2002).

3. RESULTS
3.1 Types of bacteria isolated
The overall percentage occurrences of isolated bacteria were: S. aureus 9 (20.5%), Escherichia coli 11 (25.0%), Bacillus sp 10 (22.7%), Klebsiella sp 10 (22.7%), and coagulase negative Staphylococci sp 4 (9.1%) respectively. From the washed water, S. aureus 2 (20.0%), E. coli 3 (30.0%), Klebsiella sp 1 (10.0%), Bacillus sp 3 (37.3%) and coagulase negative Staphylococci sp 1 (10.0%) respectively. Rinse water, S. aureus 3 (27.3%), E. coli 3 (27.3%), Klebsiella sp 3 (27.3%), Bacillus sp, 1 (9.1%) and coagulase negative Staphylococci sp 1 (9.1%) respectively. From source water, S. aureus 1 (9.1%), E. coli 3 (27.3%), Klebsiella sp 1 (27.3%), Bacillus sp 3 (27.3%) and coagulase negative Staphylococci sp 1 (9.1%) respectively. Rinse water, S. aureus 3 (27.3%), E. coli 3 (27.3%), Klebsiella sp 3 (27.3%), Bacillus sp, 1(%) and coagulase negative Staphylococci sp 1 (9.1%) respectively. Swab from wash hand bowl: S. aureus 3 (25.0%), E. coli 2 (16.7%), Klebsiella sp 3 (25.0%), Bacillus sp 3 (25.0%) and coagulase negative Staphylococci sp 1 (8.3%) respectively.
Figure 3.1. Types of organisms isolated from hand-washing bowls. a) Hand washing water b) Rinse water c) Stored water d) Hand washing bowl swab. S: *Staphylococcus* sp, E: *E. coli*, B: *Bacillus* sp, K: *Klebsiella* sp and C: coagulase negative *Staphylococci* sp. Key: bowl swab (Nsw), stored water (Nso), hand rinsing water (Nri), hand-washing water (Nwa).

3.2 Percentage occurrences of microorganisms isolated

A total of 44 bacteria were isolated from different water sources. The percentages are represented in fig 3.2 below.
4. DISCUSSION
The bacteria isolated from the four sources examined were *S. aureus*, *E. coli*, *Bacillus sp* and *Proteus* coagulase negative *Staphylococci* sp. *Staphylococcus* is a common inhabitant of humans (normal flora) of the hands, nostrils, mouth, skin etc. It was noted that bacteria could be transferred from office lock handles to toilet lock handles by human hands as the vehicle of transmission (Amala and Ade, 2015). Transmission of *Staphylococci* from humans is by contact with the nose and other parts of body that harbour the bacterium as normal microbiota. The possibility of inanimate objects such as computer keyboard, cellular phones, transferring bacteria to human hands for onward transmission was noted by (Amala and Ejikema 2015, Amala and Nwokah, 2016). *E. coli* and *Klebsiella* are both coliform bacteria, the presence of both organisms suggest faecal contamination from human or animals and contaminated water. *E. coli* and *Klebsiella* are associated with gastroenteritis and isolating both bacteria should be of public health interest or concern. The two bacteria are mostly transmitted through the faeco-oral route (Curtis, 1999). The infective dose of *E. coli* O517 was estimated at 100 to 500 colony forming units (cfu) but the immunological status of the host may determine the initiation of infection (Hockin et al., 1989).

*Bacillus sp* are spore bearers and their bacterial spores are geophilic thereby giving them easy access to contaminate vegetables, local spices, legums and other ingredients used in cooking. Both bacteria are implicated in food borne illness. *Bacillus cereus* causes both food infection and food poisoning. The bacterial isolates could also contaminate human hands via contact with inanimate objects and subsequently transmitted to wash hand basins and water. The hands are commonly contaminated by contact with inanimate surfaces. It was observed that in the process of touching surfaces of objects the hands could pick up infectious agents from the surfaces and transmit (Pittet et al., 2000). The isolation of the four genera of bacteria from the four sources examined may suggest the trading of these potential pathogens among these sources. Biofilm may be formed in the plastic drums used to store water for hand washing and rinsing and washing of plates, thereby distributing the bacteria continuously to other sources. Hand wash does not completely eliminate bacteria, if not done for at least 20 seconds (WHO, 2010). Some persons may not be patient to wash properly for the prescribed period thereby caring over some bacteria into the meal and eventually end up in the gastrointestinal tract (GIT) and infection may ensue depending on the immunological status of the individual.

The current study isolated five (5) bacteria which were also isolated in previous studies and were tagged as potential pathogens (Amala and Monsi, 2017).

5. CONCLUSION
Bacteria isolated from hand washing bowl and water might have been carried over from previous users.

Competing Interests
Authors have declared that no competing interests exist.

Authors’ Contributions
ASE designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. MTP managed the analyses and literature searches of the study. All authors read and approved the final manuscript.

REFERENCES