Prevalence and Antibiogram of Bacteria Isolated from Saloon Equipment in Rivers State, Nigeria

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Abstract

The rate of new saloons in Rivers State is alarming and the conditions with which the operators carryout their activities, calls for public health concern. Hence, the prevalence and antibiogram of bacterial isolates from barbing saloon equipment in barber shops located in Bori, Omoku, and Port Harcourt in Rivers State was investigated using standard microbiological techniques. Samples for analysis were collected by swabbing the surfaces of clippers, brushes, and combs using moist swab sticks. The total heterotrophic bacteria and Staphylococci were evaluated using nutrient and mannitol salt agar respectively. The Kirby Bauer disc diffusion method was used to determine the susceptibility pattern of the isolates. The mean heterotrophic bacterial counts ranged from 17.62±2.75x10^3 to 40.95±14.49x10^{3}CFU/cm^2, the total Staphylococcal count ranged from 11.67±1.29x10^2 to 39.69±20.61 x10^{2}CFU/cm^2. A total number of 77 isolates belonging to six genera were isolated and they include; Acinetobacter baumannii (2.59%), Arthrobacter mysorens (16.88%), Bacillus cereus (20.78%), Micrococcus luteus (12.99%), Serratia marcescens (9.1%), Staphylococcus aureus (37.66%). The prevalence of these organisms was more on the brush with a mean of 2.68 compared to clipper and comb with means of 1.63 and 0.5, respectively. Statistical analysis showed no significant difference among the Port Harcourt isolates but showed a significant difference for Bori and Omoku isolates at p≤0.05. The susceptibility pattern showed Staphylococcus aureus was resistant to Ofloxacin, Levofloxacin, Cephaflash and Cotrimoxazole while Bacillus cereus and Arthrobacter mysorens were susceptible to all eight antibiotics. The pattern of occurrence on different equipment suggests that less attention is usually given to brush in these establishments. Regulatory policies to check the activities of these saloons to prevent an outbreak of disease is advocated.

Key words: *Prevalence, Barbing saloon equipment, Bacterial isolates, Antibiogram*

INTRODUCTION

The establishment of barbing saloons in Rivers State seems to steadily be on the increase and because the establishment is not regulated, there is a steady increase in the establishment of new ones. Background knowledge on infectious disease control which should be a prerequisite for saloon operators is sadly lacking in the majority of these saloon operators. Known for its aesthetic activity, research however shows the possibility of it making its patrons feel sick by the acquisition of contagious diseases after visiting the saloon (Kondo *et al*., 2006). All individuals (male and female) have approximately 300,000 hairs on their scalp with a growth rate of approximately half an inch per month (Elewski, 2000). Therefore, there would be at least a monthly visit to the barbing saloon for a haircut. The advent of electric clippers for barbing operations replaced the traditional use of razor blades and other sharp objects following technological advancement (Mackenzie *et al*., 2005). The inappropriate disinfection or sterilization methods used in many barbing saloons and the re-use of barbing equipment has heightened the concern regarding communicable diseases associated with the scalp.

The use of kerosene, diesel, ethanol, fuel, and other cleaning agents for the sterilization of clippers, combs, and brushes is common practice among barbers in Nigeria (Kligman *et al*., 2011). Bacterial infections such as impetigo-like lesions have been reported to be infections associated with barbing operations. Opportunistic pathogens are usually present in non-living cornified layers of the skin and its appendages (Kligman *et al*., 2011).

Host resistance and the inoculum size are major determinants in the establishment of a microbial infection during barbing. Although, the severity of the infection depend mostly on the immunologic status of the host (Mackenzie *et al*., 2005).

Many bacterial species live as normal flora of the skin and mucous membranes of humans with *Staphylococcus aureus* being one of the most important and pathogenic species (Ryan and Ray, 2004). Some other bacterial species including *S. epidermidis*, are considered commensals, or normal residents of the skin surface. Even though they are harmless in most individuals, the bacteria are capable of causing various infections of the skin and other organs. Abrasions on the skin are a predisposing factor for these infections (Ryan and Ray, 2004).

Infection of the hair follicles caused by *S. aureus* is one of the most common causes of folliculitis (Smoloinski, 2003). Other skin infections caused by this bacterium include impetigo (crusting of the skin), cellulitis (inflammation of deeper layers of the skin and connective tissues under the skin). Scalded skin syndrome is a rare but possible serious complication that can develop from infection by *S. aureus* (Smolinski *et al*., 2003). Therefore, this study aims at assessing the prevalence and antibiogram of bacteria isolated from saloon equipment in Rivers State Nigeria with a view of providing information that would help policy makers in controlling out outbreaks communicable diseases.

MATERIALS AND METHODS

Study Area

Three major towns were selected for this study in Rivers State: Port Harcourt (PHALGA), Omoku (ONELGA), and Bori (KHANA LGA). The three local government areas represent the three senatorial districts of Rivers state. The choice of each locations depend on the population of the area.

According to Demogrophia, 2016, Port Harcourt is the capital and largest city in Rivers State and has an estimated population of 1,865,000 inhabitants. Omoku is a town located in the northern part of Rivers state in Rivers West senatorial district and the capital of the Ogba/Egbema/Ndoni Local Government Area with an estimated population of about 200,000 people (Demogrophia, 2016). Bori is a city in Khana Local Government Area, Rivers State. It is the traditional headquarters of the Ogoni people and the second-largest city in Rivers State after Port Harcourt (Hamilton, 2003).

Sample Collection

A total of one hundred and eighty samples (180) were collected by rotating a moist swab over the surface of the cutting edge of the clipper, and surfaces of combs and brush (Michael *et al.*, 2016). Samples were collected twice a month for two months. Samples were then put in sterile tubes containing 1mililitre of sterile distilled water to avoid drying and transported to the laboratory. The name, source, and location were noted on the swab sticks and brought to the laboratory under sterile/aseptic conditions for microbiological analysis.

Enumeration, Isolation and Characterization of Bacterial Isolates

Swab samples were dipped in 10ml of sterile normal saline and subsequently diluted into test tubes containing sterilized 9ml of normal saline to make 10^{-1} to 10^{-3} dilutions. An aliquot (0.1ml) of the dilutions were inoculated onto Nutrient agar and Mannitol salt agar plates in duplicates. Plates were incubated at 37^{o}C for 48hours according to the method of Elis *et al.*, (2007). Pure cultures of bacterial isolates were obtained by aseptically inoculating representative colonies of different morphological types which appear on the culture plates onto freshly prepared Nutrient agar plates and incubated at 37^{o}C for 24hours.The isolates were identified based on biochemical, morphology, and cultural characteristics (Wemedo and Robinson, 2018).

Antimicrobial Susceptibility Testing

The Kirby Bauer disc diffusion method as described by Wemedo and Robinson (2018) was adopted. In this method, the twenty four (24) hours test isolate was first standardized by emulsifying the test isolate in 4ml normal saline and matching the turbidity with 0.5 McFarland standard. After which a sterile cotton swab was dipped into the test tube and spread over the entire surface of the prepared Mueller-Hinton agar (Oxoid, Cambridge, UK) plates. The plates were allowed to dry. Using sterile forceps, the sensitivity disc was placed on the inoculated plates, incubated for 24hours at 37^{o}C. Zones of inhibition were measured using a graduated ruler and results were interpreted based on the guidelines of the clinical laboratory standards institute (CLSI, 2013) to determine which isolates were susceptible, intermediate, or resistant.

RESULTS

Results of the total heterotrophic bacterial load of the saloon equipment in the various locations is presented in Table 1. The mean ranges of the total heterotrophic bacterial load for the clipper, brush and combs were 17.62 ± 2.75^{a} to 24.61 ± 11.27^{a}; 29.90 ± 7.85^{b} to 40.95 ± 14.49^{a}; and 24.95 ± 3.70^{a} to 40.38 ± 12.02^{b} ($x10^{3}$CFU/cm^{2}) respectively. More so, the total heterotrophic bacterial load of the clippers in Port Harcourt barbers' shop were higher than the counts recorded for Omoku and Bori. Despite this variation, there was no significant difference ($P\leq0.05$). The

total heterotrophic bacterial load of the brushes showed that the counts recorded in Port Harcourt and Omoku barbers' shop were significantly higher than those recorded for Bori barbers' shop while the counts recorded in the combs showed that the total heterotrophic bacterial load of the combs recorded in Port Harcourt were significantly higher than those recorded in Bori and Omoku barber's shop.

Results of the *Staphylococcal* count of the saloon equipment in the various locations is presented in Table 2. The mean ranges of the *Staphylococcal* counts for the clipper, brush and comb were 11.67 ± 2.86^{a} to 16.66 ± 9.35^{a}, 31.26 ± 13.70^{a} to 39.69 ± 20.61^{b}, 13.39 ± 2.67^{a} to 23.22 ± 16.92^{b} ($x10^{2}cfu/cm^{2}$) respectively. The *Staphylococcal* counts of clippers and combs in Port Harcourt barbers' shop was higher than those reported in the Bori and Omoku barbers' shop but despite this disparity in *Staphylococcal* counts of the clippers, there was no significant difference while there was a significant difference ($P\leq0.05$) amongst the *Staphylococcal* counts recorded for the combs with counts recorded in Port Harcourt and Bori barbers' shop being significantly higher than those recorded for Omoku barber's shop.

Table 1: Total Heterotrophic Bacteria Count ($x10^{3}cfu/cm^{2}$) of Saloon Equipment.

Location	Clipper	Brush	Comb
Bori	17.62 ± 2.75^{a}	29.90 ± 7.85^{b}	24.95 ± 3.70^{a}
Omoku	23.65 ± 9.74^{a}	40.33 ± 3.98^{a}	29.93 ± 7.40^{a}
Port Harcourt	24.61 ± 11.27^{a}	40.95 ± 14.49^{a}	40.38 ± 12.02^{b}

*Means with same superscript across the column shows no significant difference($p\geq0.05$)

Table 2: Total *Staphylococcal* Count ($x10^{3}cfu/cm^{2}$) of Saloon Equipment.

Location	Clipper	Brush	Comb
Bori	14.12 ± 11.66^{a}	33.05 ± 22.84^{a}	20.64 ± 18.05^{b}
Omoku	11.67 ± 2.86^{a}	39.69 ± 20.61^{b}	13.39 ± 2.67^{a}
Port Harcourt	16.66 ± 9.35^{a}	31.26 ± 13.70^{a}	23.22 ± 16.92^{b}

*Means with same superscript across the column shows no significant difference at $p\geq0.05$

A total of seventy-seven bacterial isolates belonging to six genera were characterized from the various samples. The bacterial isolates include; *Acinetobacter baumannii*, *Arthrobacter mysorens*, *Bacillus cereus*, *Micrococcus luteus*, *Serratia marcescens*, and *Staphylococcus aureus* Results of the percentage occurrence of the bacterial isolates is presented in Figure 1. The percentage occurrence of the different saloon equipment in Bori showed that for brush, *S.aureus* had the highest percentage (62%), for clipper, *Serratia marcescens* had the highest percentage (66.7%), and for comb *Arthrobacter mysorens* had (28.6%). The percentage occurrence of the

different saloon equipment in Omoku showed that for brush, *Serratia marcescens* had the highest percentage (100%), for clipper, *Bacillus cereus* had the highest percentage (60%), and for comb *Micrococcus luteus* had (33.3%). Furthermore, the percentage occurrence of bacterial isolates from the different saloon equipment in Port Harcourt showed that *Serratia marcescens* had the highest percentage for brush (100%), for clipper, *Bacillus cereus* had the highest percentage (33.4%), and for comb *S.aureus* had (18.2%) as the highest percentage.

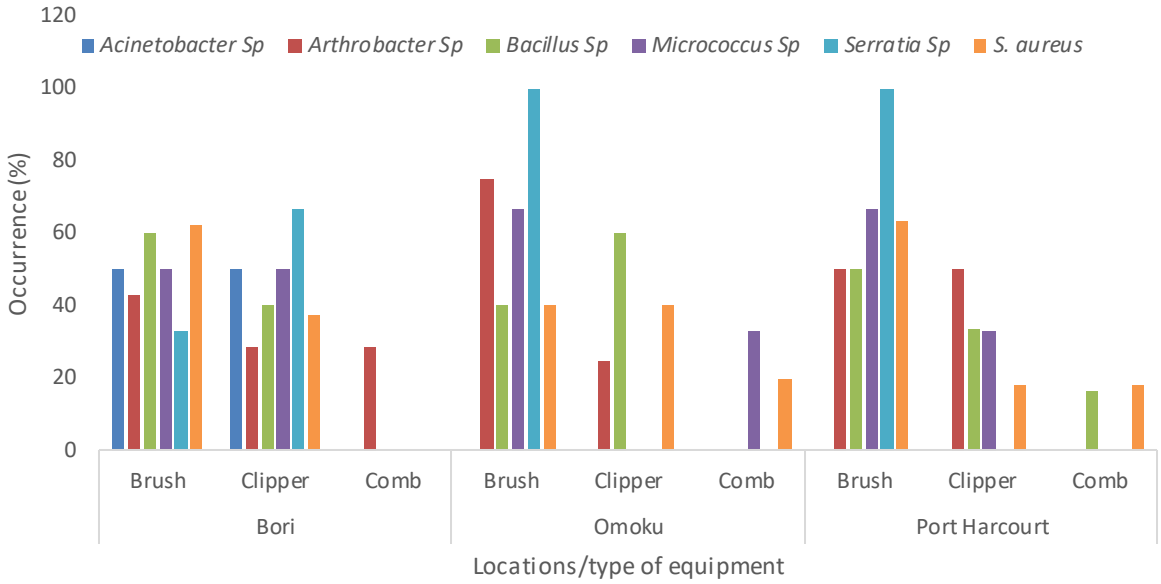

Figure: 1: Percentage Occurrence for Different Bacteria from Saloon Equipment from the Different Locations

The summary of the susceptibility pattern of the bacteria isolates are shown in Table 3. The table showed that *Staphylococcus* spp. being the most resistant isolate (four antibiotics), while *Micrococcus luteus* and *Serratia marcescens*. resisted three and five antibiotics respectively. *Acinetobacter baumannii*, *Arthrobacter mysorens* and *Bacillus cereus* had no resistant isolates.

Table 3: Multiple Antibiotic Resistance and Percentage Resistance of Bacterial Isolates

MAR	*Acinetobacter*	*Arthrobacter*	*Bacillus*	*Micrococcus*	*Serratia*	*S.*

	baumannii (N=2)	*mysorens* (N=13)	*cereus* (N=16)	*luteus* (N=10)	*marcescens* (N=7)	*aureus* (N= 29)
0.00	2(100)	13(100)	16(100)	7(70)	2(28.6)	24(82.8)
0.1	0(0.00)	0(0.00)	0(0.00)	1(10)	2(28.6)	2(6.9)
0.2	0(0.00)	0(0.00)	0(0.00)	(0.00)	2(28.6)	2(6.9)
0.3	0(0.00)	0(0.00)	0(0.00)	2(20)	1(14.2)	0(0.00)
0.4	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	1(3.4)

N= number of isolates

MAR index = a/b…Equation 1

Where, a = total number of antibiotics in which the organism showed resistance

b = total number of antibiotics in which the isolated were evaluated against

Percentage resistance = $\frac{\text{number of isolates resisting a particular antibiotic}}{\text{total number of the isolates}} \times 100$ …. Equation 2

(Barika and Akani, 2019)

Discussion

The mean total heterotrophic bacterial counts of saloon equipment from Barbers' shop in Bori showed that brushes had the highest counts followed by the clippers while the combs had the least counts of total heterotrophic bacteria. The brushes also had the highest counts in the total heterotrophic fungi and total *Staphylococcal* counts compared with counts obtained for clippers and combs in the barbers' shop located in Bori. In the barbers' shops located in Bori, the least heterotrophic fungal and *Staphylococcal* counts were recorded from clippers while combs had the second highest counts in the aforementioned microbial populations. Statistical analysis showed that there were statistical differences in the total heterotrophic bacterial and total heterotrophic fungal counts recorded amongst the saloon equipment with counts recorded for brushes being significantly ($P<0.05$) higher than those recorded for clippers. While the disparity in *Staphylococcal* counts in the respective saloon equipment was not enough to show any significant differences ($P> 0.05$) in barbers shops located in Bori. The heterotrophic bacterial counts recorded for brushes in barbers' shops located in both Port Harcourt and Omoku were higher than counts obtained for clippers and combs of the respective barbers' shops. Despite the varied counts in microbial population recorded for the saloon equipment in barbers shop located in Port Harcourt, there was no significant differences ($P> 0.05$) amongst the counts for total heterotrophic bacteria, total heterotrophic fungi and *Staphylococcal* counts. This is in contrast with findings obtained from barbers' shops located in Omoku which showed that the total heterotrophic bacterial, total fungal and *Staphylococcal* counts of brushes were significantly higher ($P<0.05$) than counts obtained for clippers and brushes.

Although the microbial counts recorded in this study were very high, the counts recorded in a previous study were higher than those recorded in this study. The mean ranges of the total

heterotrophic bacterial load for combs and brushes recorded by Stanley *et al.* (2019) were 1.8×10^6 to$3.7x10^6$ CFU/cm^2 and $1.6x10^6$ to $3.4x10^6$ CFU/ cm^2 which are higher than the mean ranges recorded in the current study for saloon equipment. However, their findings which showed that brushes had higher heterotrophic bacterial counts especially in some of the barbers' shop agreed with our findings. Also, the total heterotrophic fungal counts in this study are lower than values reported in a previous study (Mbajiuka *et al.*, 2014; Stanley *et al.*, 2019). More so, the total heterotrophic bacterial counts in this study were lower than the values reported by (Mbajiuka *et al.*, 2014). The high bacterial counts recorded in the brushes across the barbers' shop could be attributed to over use or inadequate treatment or washing of brushes. Other factors that could have orchestrated high microbial load in the brushes amongst other saloon equipment is the number of the saloon equipment available per haircut and the type of treatment used on the saloon equipment. The bacterial isolates especially *Staphylococcus* sp and *Bacillus* sp isolated in this study were also reportedly isolated in Wukari, Taraba State, Nigeria, from saloon equipment used by barbers (Ebuara *et al.*, 2020) and in other previous studies (Dadashi and Dehghanzadeh, 2016; Mbajiuka *et al.*, 2014; Stanley *et al.*, 2019). The differences between the current study and these previous studies are the isolation of *Acinetobacter baumannii* and *Arthrobacter mysorens* (Ebuara *et al.*, 2020; Gahongayire *et al.*, 2020; Stanley *et al.*, 2019). *Micrococcus luteus* and *Serratia marcescens* which were isolated in this study were reportedly isolated from saloon equipment in previous studies which agreed with the current findings (Gahongayire *et al.*, 2020; Stanley *et al.*, 2019). Another difference between the current study and previous study is the isolation of *Citrobacter* spp., *Proteus* spp. and *Shigella* spp. by Stanley *et al.* (2019).

The prevalence and distribution of bacterial isolates in saloon equipment showed that *Staphylococcus aureus* was very predominant and ranked as the most frequently isolated bacteria across all barbers' shop in the respective saloon equipment. The finding agreed with findings of Enemuor *et al.* (2013) and Dadashi and Dehghanzadeh (2016) who reported higher distribution of *S. aureus* from hairdressing and beauty salons tools. However, the findings in the current study is contrary to reports by Gahongayire *et al.* (2020) who reported *S. epidermidis* as the most prevalent bacterial isolate. Also, the findings in this study agreed with Stanley *et al.*,(2019) who reported that *S. aureus* was the predominant bacterial isolates in saloon equipment. The contamination of all saloon equipment with brush having the highest contamination is contrary to Omoruyi and Idemudia (2011), who reported prevalence of 91.67% and 83.3% of *Staphylococcus* and *Bacillus* sp, respectively, from clippers used in barbers' shop in Benin City, Nigeria. The findings in the current study corroborates that of Naz *et al.* (2012) who reported 100%, 100%, and 88% of *S. aureus* contamination in sponge, brush, and wax, respectively, from cosmetic tools used in beauty saloons from different areas of Lahore, Pakistan. *Staphylococcus* sp are known to be normal skin flora, thus, there high predominance in saloon equipment could be attributed to contamination from handlers unlike *Acinetobacter baumannii*, *Arthrobacter mysorens*, *Bacillus cereus*, *Micrococcus luteus* and *Serratia marcescens* which are commonly found in the soil (Prescott *et al.*, 2011). The high bacterial distribution observed in the brushes amongst other saloon equipment could be attributed to lack of standard sterilization procedures for brushes unlike the clippers which are sterilized (though inadequately since it is not able to rid it of microbial contamination). This observation were also reported in a previous study (Ebuara *et al.*, 2020). *Staphylococcus* spp.. are able to cause various diseases in humans such as skin abscess, impetigo contagiosa, scaled-skin disease syndrome, and it is the most commonly

identified agent that is responsible for skin and soft tissue infection (Enemuor *et al.,* 2013; Helaskoski *et al.,* 2014).

Generally, the antibiotics susceptibility pattern of the bacterial isolates in the current study showed multi-drug resistance as some of these isolates such as *Acinetobacter baumannii*, *Arthrobacter mysorens*, *Bacillus cereus*, *Micrococcus luteus*, *Serratia marcescens* and *S. aureus* were resistant to more than two antibiotics. These could be attributed amongst other factors to the possession of resistance genes. Previous studies have reported that due to environmental factors, heavy antimicrobial use and other undefined factors, antimicrobial susceptibility could vary in respective locations (Dora, 2011). In the current study, *Staphylococcus* sp were found to be resistant to four different antibiotics especially Ofloxacin, Ciprofloxacin and Gentamycin. Thus, resistance of the isolate to Ciprofloxacin and Ofloxacin agreed with the work of Eribo *et al.*(2017) who reported that *Staphylococcal* isolates isolated from barbers shop exhibited resistance to cephazolin, ofloxacin, ciprofloxacin, sulfamethoxazole and novobiocin.The resistance of antibiotics observed in this study could be attributed to continuous usage or exposure of these antibiotics to the bacterial isolates. The resistance observed in the aminoglycoside like the Gentamycin could be mediated by preventing the drugs from reaching the ribosomes which is their target site and this is usually achieved in two ways: by altering the cell envelope which inhibits the uptake of the drug and via the modification of the drug by inactivating enzymes (Haifei *et al.,* 2012)

Conclusion

In conclusion, the evaluation of saloon equipment for bacterial contamination and antibiotics susceptibility have revealed that the bacterial load are very high and sterilization methods used on this equipment may not be effective in inhibiting the presence of these bacterial population. More so, the bacterial types identified in the study could be normal or transient flora from the persons who utilize these materials. Thus, the transfer of bacterial types from one person to another could predispose people who go for hair cuts especially those who are susceptible to skin or other forms of infections. Although the level of resistance of the bacterial isolates to these antibiotics is alarming, ciprofloxacin and ofloxacin have shown to be the drug of choice.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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