Incidence of Staphylococcus aureus isolated from armpits of students of University of Abuja, Nigeria.

Ojih, Felix Eleojo [1], Okon, Okon Godwin* [1&2], Nwanna, Vivian Nkemjika[1], Udosen, Imo Joseph[3] and Abraham, Nsikak Andrew[3].

[1] *Biological Sciences Department, University of Abuja, Nigeria.*
[2] *Department of Botany and Ecological Studies, University of Uyo, Nigeria.*
[3]*Department of Microbiology, University of Uyo, Nigeria.*
**Corresponding author: okjunior4zeeb@gmail.com*

Abstract

A study of the carriage of microorganisms in armpits and the factors affecting it was carried out on 180 students of the University of Abuja. The armpits were swabbed and microbiological analyses were carried out on the swab samples. Samples were inoculated on Mannitol salt agar plates and coagulase positive Staphylococcus aureus isolates were identified. Questionnaires on gender and health related factors were given to students before collection of samples. Statistical analysis using analysis of variance was used for data analysis. Male students used toilet soap (73.75%), had their bath once daily (75%), used deodorant (23%), used sponge for body scrubbing (85%), shaved once in two weeks (65%) and 15% used powder but these did not have any significant influence on the carriage of Staphylococcus aureus (P = 0.05). Most female students used medicated soap (69%), had their bath twice daily (66%), used deodorant (73%), used sponge for body scrubbing (73%), shaved once in month (65%) and 38% used powder but these did not have any significant influence on the carriage of Staphylococcus aureus (P = 0.05). More female participants used deodorants, than the males. The Staphylococcus aureus counts in the armpits of females were lower than the counts from male armpits, which mean that the use of deodorant and powder reduced the carriage of the microorganisms.

Key Terms –armpit, deodorant, Mannitol, microorganisms and*Staphylococcus aureus*

1. Introduction

Large numbers of microorganisms live on and in the various components of the normal skin. Depending on the body location and the amount of skin moisture, the number of skin bacteria may range from only about 1000 organisms per square centimeter on the back to more than 10 million in the groin and armpit, where moisture is more plentiful. The number of microorganisms increases after a hot shower because of increased flow of secretion from the skin glands where many reside (Uzeh *et al*., 2012; Nester *et al*., 2004). In humans, the formation of body odours is mainly caused by skin glands excretions and bacterial activity (Lundstrom and Olsson, 2010). The

axilla, a skin region around the armpit usually differs from other regions of the body with respect to the presence, identity and number of sweat glands. The axillary region is of particular interest, as it contains dense aggregations of eccrine, apocrine, and sebaceous glands that nurture diverse communities of micro-biota thought to play an important role in generating individual odour (Uzeh *et al*., 2012; Taylor *et al*., 2003).Between the different types of skin glands, the human body odour is primarily the result of the apocrine sweat glands, which secrete the majority of chemical compounds needed for the skin flora to metabolize it into odorant substances (Lundstrom and Olsson, 2010). This happens mostly in the axillary (armpit) region, although the gland can also be found in the areola, anogenital region, and around the navel (Turkington and Dover, 2007). In humans the armpit regions seem more important than the genital region for body odour which may be related to human bipedalism. The genital and armpit regions also contain springy hairs which help diffuse body odours (Claus, 2007). Body odour is influenced by the actions of the skin flora, including members of *Corynebacterium*, which manufacture enzymes called lipases that break down the lipids in sweat to create smaller molecules like butyric acid. These smaller molecules smell, and give body odour its characteristic aroma (Uzeh *et al*., 2012; Buckman, 2003).Propionic acid (propionic acid) is present in many sweat samples. Propionibacteria in adolescent and adult sebaceous glands can turn its amino acids into propionic acid. Isovaleric acid (3-methyl butanoic acid) is the other source of body odour as a result of actions of the bacteria *Staphylococcus epidermidis* (Ara *et al*., 2006). A wide variety of deodorant and antiperspirant products are sold for the purpose of mitigating this odour. Factors such as food, drink, and diseases can affect body odour (Claus, 2007). The flourishing of body flora in a given area depends upon the physiological factors of temperature, moisture, and the presence of certain nutrients and inhibitory substances. Microbes of the normal resident flora are harmless and may be beneficial in their normal location in the host and in the absence of coincident abnormalities. On mucous membranes and skin, the resident flora may prevent colonization by pathogens and possible disease through bacterial interference (Brooks *et al.*, 2004). These organisms are adapted to the noninvasive mode of life defined by the limitations of the environment. However, resident microbes may produce disease if introduced into foreign locations in large numbers and if predisposing factors are present (Brooks *et al.*, 2004). They can cause skin diseases and enter the blood system creating life threatening diseases particularly in immunosuppressed people (Uzeh *et al*., 2012; Cogen *et al.*, 2008). Thus, this study was carried out to determine the prevalence and distribution of *Staphylococcus aureus* in the armpits of some students of the University of Abuja, determine the role of gender in the distribution *Staphylococcus aureus* and assess health related characteristics such as the use of soap, sponges, deodorant, frequency of bathing and shaving in the distribution *Staphylococcus aureus.*

2. Materials and Methods

2.1 Study Area

Armpit swabs were obtained from students of different departments of the University of Abuja. Permission was obtained from heads of the different departments through a letterfrom the department of Biological Sciences, Faculty of Science, University of Abuja. The material for the sample collection and overview of the study was shown and briefly describe to the students and Head of Department. Confidentiality of the result was explained to the participants.

2.2 Collection of Samples

Armpit swabs of 180 students from University of Abuja, between the age group of 17-25 years were collected before and after bath. Sterile swab sticks moistened with 1.0 ml of sterile normal saline were rubbed vigorously with rotation over the armpits of participants.Questionnaire on health related characteristics of the participants was completed before the sample collection. Information such as gender, age, use of soap, sponge, deodorant and powder, frequency of bathing and shaving of armpits hairs were required from the participants.

2.3 Sample Size

One hundred and eighty 180 samples were randomly collected from the both male and female hostels. Eighty (80) samples of male participants and one hundred (100) samples of female participants

2.4 Culture Conditions and Identification of Isolates

The swabs were inoculated immediately after rolling them over the armpits of participants onto Mannitol Salt Agar (MSA) plates. The plates were inoculated at 37^{o}C aerobically for 24 hours. The plates were examined for growth at the end of inoculation period. Typical colonies were picked aseptically with inoculating loop and purified by sub-culturing. *Staphylococcus aureus* was examined on the basis of morphology, gram stain reaction, positive catalase test, coagulase production and fermentation of Mannitol.

2.5 Macroscopic and microscopic analysis of isolates

The cultured plates were observed macroscopically for physical appearance of the colonies. These physical appearances include colour, size, shape, transparency, consistency and nature of the surface (Raygada and Levine, 2009). The isolates were also observed microscopically after Gram staining as described by Cheesbrough (2005). Using the ×100 oil immersion objective of the light microscope, isolates were identified and assigned to their genera according to the procedures outlined by Cheesbrough (2005).

2.6 Coagulase test

In this study, the slide method test was used. A drop of saline on two separate spots was placed on a grease-free slide. Then, a speck of growth of the test organism was picked and emulsified in both spots, to one spot a drop of plasma was added and to the other a drop of saline was added. Both treatments mixtures were mixed thoroughly by rocking. Coagulation was an indication of positive

test to which plasma was added. The presence of clotting indicates positive test for *Staphylococcus aureus* (Cheesbrough, 2005). This test was based on our understanding that the microorganism has the capability to produce Coagulase enzyme which causes the coagulation of human blood plasma.

2.7 Catalase test

This was carried out to determine the ability of the microorganisms to produce Catalase enzyme and degrade hydrogen peroxide (H_2O_2). A drop of 3% Hydrogen peroxide was placed on a clean glass slide. A speck of growth of each isolate was collected from the medium using a wire loop and the growth was emulsified in the drop. A positive test was indicated by bubbling and frothing (Cheesbrough, 2005). The principle behind the Catalase test is to differentiate between pathogenic and non-pathogenic *Staphylococcus.*

2.8 Sugar Fermentation

10 ml of peptone water was introduced into 4 sterile test tubes respectively. 1 g of respective carbohydrate such as glucose, Mannitol, lactose, maltose and sucrose were added into each of the test tubes that contain the peptone water and labeled accordingly. They were stirred to dissolve completely over a Bunsen burner after which 3 drops of phenol red was added into each of the test tubes. The tubes were plugged with cotton wool and sealed with foil before sterilization in autoclave at 115^{o}C for 15 minutes. After the sterilization of medium, the cultural organisms were inoculated into each of the tubes respectively and Durham's tubes were inserted in an inverted position into each of the tubes. Then, they were inserted in an inverted position into each of the tubes. They were also incubated at 37^{o}C for 24 hours. A change in the coloration of medium after 24 h from purple to yellow indicated acid production due to the fermentation of sugar by the organisms while retention of the purple color indicated a negative reaction. Gas production was shown by the presence of bubbles on the surface of the medium and on upward movement of the inverted Durham's tubes

2.9 Statistical Analysis

Data generated in the course of this study were analyzed using analysis of variance (ANOVA). The student's t-test statistical tool was used to test for significant differences between the variables with regard to the hypotheses tested. The P-value was used as decision rule for accepting or rejecting the null hypothesis. The level of significance for the test was 0.05.

3. Results

Observations on the effect of age of student participants on cultural characteristics of the bacterial isolates showed that almost all students (83.00%) had growth on Mannitol Salt Agar. On the average 61.00% pure cultures and 39.00% mixed cultures on primary isolation. The pure colonies were mostly yellow and fermented Mannitol while the pure white colonies were mucoid and non-mannitol fermenting. The mixed cultures consisted of yellow colonies (about 3mm) in diameter, tiny background white

colonies (1mm) in diameter, medium white colonies (3mm) in diameter and large white colonies (5mm) in diameter. *S. aureus* was isolated from more male (25.00%) than female (15.00) according to gender characteristics. male (22.00%) than female (13.00) according to age characteristics.There were no significant differences in most of the variables examined between male and female student subjects including deodorant usage (P = 0.0699) with more female using it, bathing habit (P = 0.2151), powder usage (0.01182), shaving pattern (P = 0.226) and sponge usage (P = 0.0344). However, there was significant difference in the use of soap (P = 0.001) among males and females. Majority of the participants used medicated soaps (49.44%), had bath twice daily (47.22%), used sponge (78.33%), shaved once a month (42.78%), while 53.33% used deodorant as shown on Table 1.

Table 1: Percentage distribution of characteristic habits of students by gender

Health related characteristics	Male n=80 (%)	Female n=100 (%)	Total n=180 (%)	P-value
Soaps Usage				
Medicated soap	23 (28.75)	66 (66.00)	89 (49.44)	1.634
Toilet soap	42 (52.50)	30 (30.00)	72 (40.00)	
Local soap	15 (18.75)	04 (4.00)	19 (10.56)	
Deodorant Usage				
Yes	18 (22.50)	78 (78.00)	96 (53.33)	0.0699
No	62 (77.50)	22 (22.00)	84 (46.67)	
Powder Usage				
Yes	15 (18.75)	35 (35.00)	50 (27.77)	0.1016
No	65 (81.25)	65 (65.00)	130 (72.22)	
Bathing Habits				
Once daily	53 (66.25)	19 (19.00)	72 (40.00)	0.2151
Twice daily	24 (30.00)	61 (61.00)	85 (47.22)	
Thrice daily	03 (3.75)	20 (20.00)	23 (12.73)	
Sponge Usage				
Yes	73 (91.25)	68 (68.00)	141 (78.33)	0.0344
No	07 (8.75)	32 (32.00)	39 (21.67)	
Shaving Pattern				
Once a week	-	04 (4.00)	04 (2.22)	0.02
Once in two weeks	44 (55.00)	31 (31.00)	75 (41.67)	
Once a month	23 (28.75)	54 (54.00)	77 (42.78)	
Once in two weeks	13 (16.25)	11 (11.00)	24 (13.33)	
***S. aureus* isolated**				
Yes	20 (25.00)	15 (15.00)	35 (19.44)	0.00619
No	60 (75.00)	85 (87.00)	145 (80.56)	
***S. aureus* carriers**				
Yes	20 (25.00)	13 (13.00)	33 (18.33)	0.2326
No	60 (75.00)	87 (87.00)	147 (81.67)	

n= Number of students subjects

Table 2 shows the influence of age on health related characteristics of participants. There were no significant differences according to age in types of soap used (P=0.2418), use of powder (P=0.1016) or sponge (P=0.1259), frequency of bathing (P=0.01585), shaving frequency (P=0.02633) and deodorant usage (P=0.2016). Majority of the participants used medicated soaps (49.44%), had bath twice daily (47.22%), used sponge (78.33%), shaved once a month (42.78%), while 53.33% used.

Table 2: Percentage distribution of characteristic habits of students by gender

Health related characteristics	**Male 18-25 yrs n=80 (%)**	**Female 17-23 yrs n=100 (%)**	**Total n=180 (%)**	**P-value**
Soaps Usage				
Medicated soap	20 (25.00)	69 (69.00)	89 (49.44)	0.7156
Toilet soap	59 (73.75)	13 (13.00)	72 (40.00)	
Local soap	01 (1.25)	18 (18.00)	19 (10.56)	
Deodorant Usage				
Yes	23 (28.75)	73 (73.00)	96 (53.33)	3.211
No	57 (71.25)	27 (31.00)	84 (46.66)	
Powder Usage				
Yes	12 (15.00)	38 (38.00)	50 (27.77)	0.01182
No	68 (85.00)	62 (62.00)	130 (72.22)	
Bathing Habits				
Once daily	60 (75.00)	12 (12.00)	72 (40.00)	0.8379
Twice daily	19 (23.75)	66 (66.00)	85 (47.22)	
Thrice daily	01 (1.25)	22 (22.00)	23 (12.77)	
Sponge Usage				
Yes	68 (85.00)	73 (73.00)	141 (56.66)	1.7736
No	12 (15.00)	27 (27.00)	39 (43.33)	
Shaving Pattern				
Once a week	03 (3.75)	01 (1.00)	04 (2.22)	0.6277
Once in two weeks	52 (65.00)	23 (23.00)	75 (41.66)	
Once a month	12 (15.00)	65 (65.00)	77 (42.77)	
Once in two weeks	08 (10.00)	16 (16.00)	24 (13.33)	
***S. aureus* isolated**				
Yes	22 (27.50)	13 (13.00)	35 (19.44)	0.4451
No	58 (72.50)	87 (87.00)	145 (80.56)	
***S. aureus* carriers**				
Yes	22 (27.50)	11 (11.00)	33 (18.33)	0.4451
No	58 (72.50)	89 (89.00)	147 (81.66)	

n= Number of students subjects

Table 3 summarizes the effect of the variables on *S. aureus* carriage in armpits of participants. Age, medicated soap usage and use of deodorants affected carriage of *S. aureus* in armpits. Most male (18-25 years) did not use medicated soap and deodorant and *S. aureus* carriage was significantly high among them. Age difference was significant (P=0.0512) with 27.50% being male carriers compared to 13.00% female carriers (Figure 3 and 4). The other variables such as gender, soaps usage, use of powder or sponge, bathing and shaving frequency had no effect on *S. aureus* carriage.

Table 3: Effects of variables on carriage of *S. aureus* on armpits of participants

Variables	Percentage of *S. aureus* carriage		P-value
Sex			
Males	20/80	(25.00)	0.1063
Females	15/100	(15.00)	
Age			
18 – 25 years	22/80	(27.50)	0.0512
17 – 23 years	13/100	(13.00)	
Soaps Usage			
Medicated Soap	50/89	(56.17)	0.1851
Toilet Soap	40/72	(55.56)	
Local Soap	10/19	(52.53)	
Deodorant Usage			
Yes	50/96	(52.08)	0.02533
No	40/84	(47.62)	
Powder Usage			
Yes	30/50	(60.00)	0.013
No	65/130	(50.00)	
Bathing Habits			
Once	30/72	(41.67)	2.005
Twice	43/85	(50.58)	
Thrice	11/23	(47.82)	
Sponge Usage			
Yes	71/141	(50.35)	0.0758
No	20/39	(51.28)	
Shaving Pattern			
Once a week	2/4	(50.00)	0.2641
Once in two weeks	37/75	(49.33)	
Once a month	39/77	(50.64)	
Once in two months	10/24	(41.67)	

n= Number of students subjects

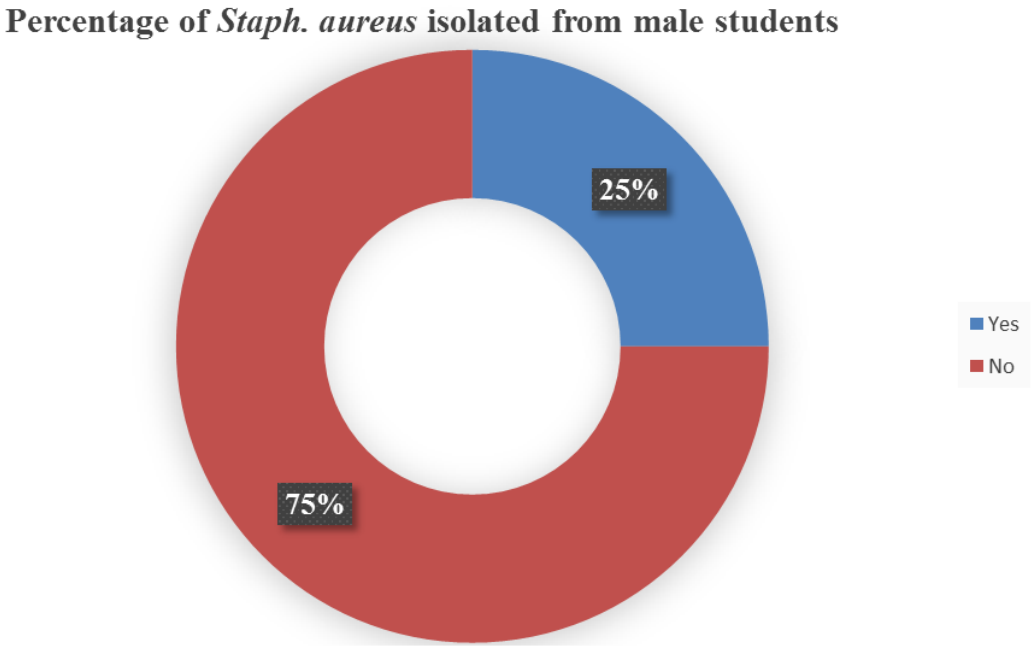

Figure 1: A pie chart showing the percentage of *S. aureus* isolated from armpit of male students

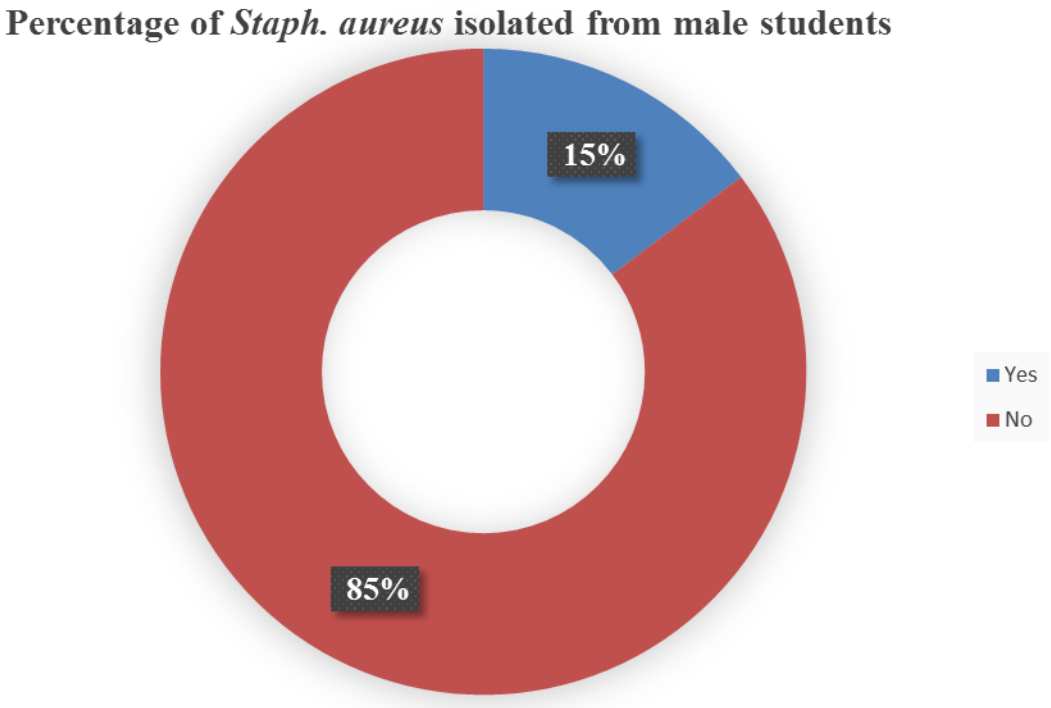

Figure 2: A pie chart showing the percentage of *S. aureus* isolated from armpit of female students

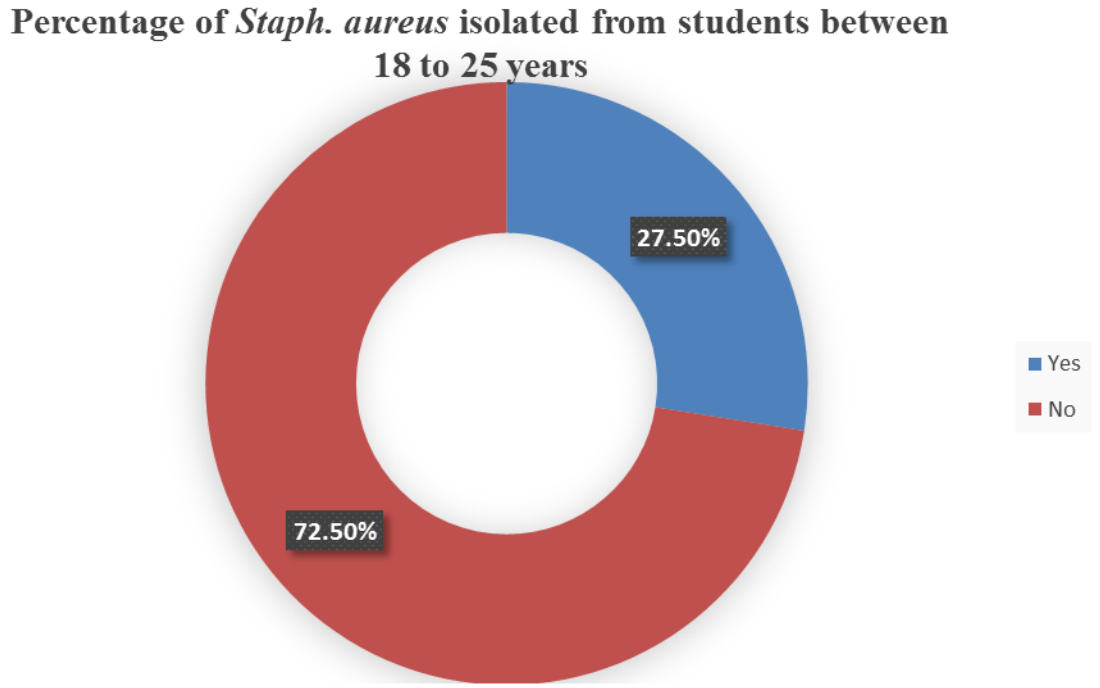

Figure 3: A pie chart showing the percentage of *S. aureus* isolated from according to age

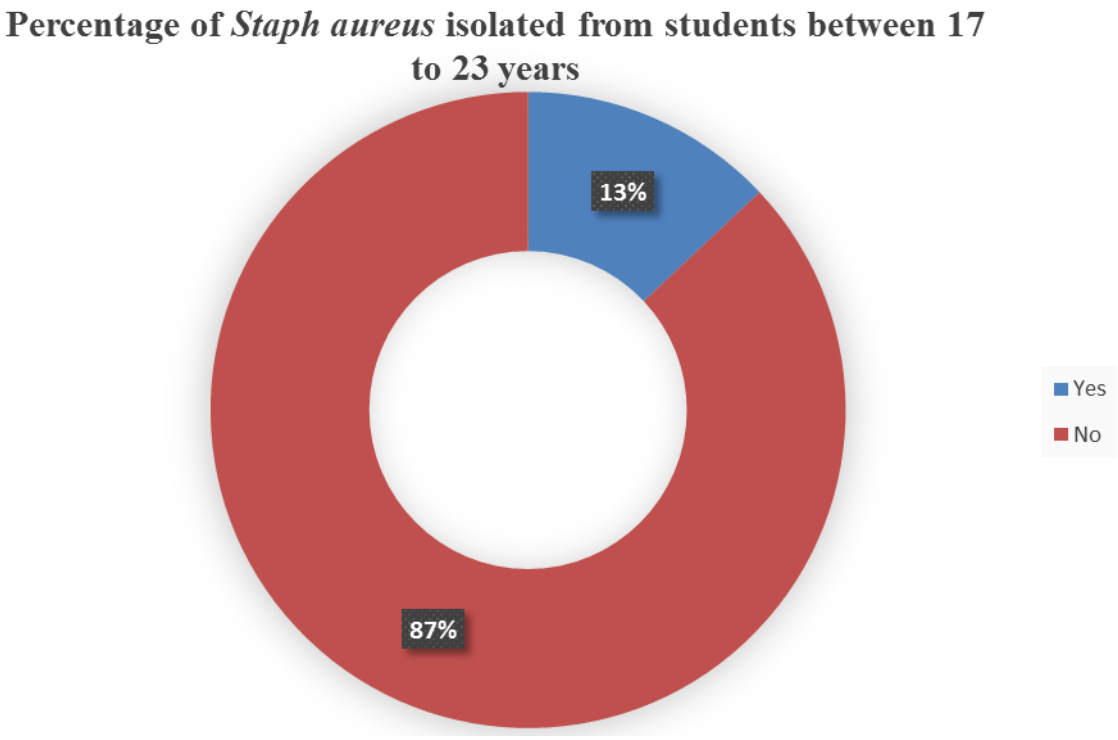

Figure 4: A pie chart showing the percentage of *S. aureus* isolated from according to age

5. Discussion

The *Staphylococcus aureus* population in the armpits of males was higher than in females. However, there was no significant difference in *Staphylococcus aureus* population before and after bath and in the use of soaps, sponges, Powder, frequency of bathing or shaving for both male and female participants. Neither profuse sweating nor washing and bathing can eliminate or significantly modify the normal resident *Staphylococcus aureus*. The number of superficial *Staphylococcus aureus* may be diminished by vigorous daily scrubbing with soap containing hexachlorophene or other disinfectants, but the flora is rapidly replenished from sebaceous and sweat glands even when contact with other skin and areas or with the environment is completely excluded (Brooks *et al.*, 2004). *Staphylococcus aureus* is a known commensal on human skin, but it is an opportunistic pathogen causing a wide range of infections among which are furuncles (boils), carbuncles, impetigo, epidermal necrosis, osteomyelitis, staphylococcal food poisoning and toxic shock syndrome. The observation in this study shows that *S. aureus* armpit carriage was 35.00% for the population studied, with 27.50% carriage among male students, ages 18-25 years and female students, ages 17-23 years is comparable to estimates by previous researches. Ibe and Wariso (2005) estimated carriage of *Staphylococcus aureus* on armpits of secondary school and university of Port Harcourt, Port Harcourt, Nigeria and most of the secondary school teenagers had less *Staphylococcus aureus* carriage on their armpits, Uzeh *et al*. (2012) worked on microbial assessment of armpits of selected students of University of Lagos, Lagos Nigeria and recorded that both male and female that used deodorants had lower bacteria counts since deodorants are known to work by suppressing the growth of microorganisms and hence armpit odors. Kloos and Jorgensen (1982) estimated *Staphylococcus aureus* carriage on the anterior nares and most areas of the skin including the armpits of apparently healthy persons to be 20.00% to 30.00%, Ibe and Wariso (2005) also reported that 30.00 to 40.00% of adults are asymptomatic carriers of *Staphylococcus aureus.* Bruer (2002) observed that *Staphylococcus aureus* can be isolated from skin of 5 to 30% of normal individuals and that persistent nasal carriage is present in 20.00% of normal adults. Stewart and Beswick (1997) estimated that about 5.00 to 10.00% of any populations are carriers. Chin (2000) also estimated 20.00% nasal carriage among population and noted that areas of the world which lack water and soaps and are filthy have higher incidence of *Staphylococcus aureus* infection.

From this study it was observed that age has a major influence on armpits carriage of *Staphylococcus aureus.* Lack of use of deodorants and medicated soap by most of the male students were clearly the health factors responsible for the higher carriage of *S. aureus.* These findings support the fact that the normal flora is acquired rapidly during and shortly after birth and changes continuously throughout life and that the organisms present at any given time reflect the age, nutrition and environment. Deodorants are known to work by

suppressing the growth of *Staphylococcus aureus* and hence armpit odours. As observation in this study, the use of soaps, sponges, powders, frequency of bathing or shaving did not affect carriage of *Staphylococcus aureus.*This agrees with the findings of Reagan *et al.* (1991) who observed that skin decontamination among hospitalized patients who were nasal carriers was not easy. They observed that armpit decolonization in 71.40% of patients was possible with six month when daily wash with antiseptic solution (biseptine with chlorhexidine) was combined with nasal de-colonization using mupirocin. Uzeh *et al.* (2004) also observed that decolonization of the skin by *Staphylococcus aureus* occurred 4 to 8 weeks of decontamination with mupirocin, chlorhexidine and potassium permanganate bath.

6. Conclusion

From this research male students of the University of Abuja had more profuse growth of *S. aureus* than the female participants who used deodorants and *S. aureu*s counts in the armpits of females were lower than the counts from male armpits. This means that the use of deodorant reduces the carriage of *Staphylococcus aureus*. Students and the general publicshould be made aware of the importance of personal hygiene by bathing frequently; paying special attention to underarms and pubic areas, using underarm deodorants, changing under wears daily and the use of good soaps.

References

[1] Ara, K., Hama, M., Akiba, S., Koike, K., Okisaka, K., Hagura, T., Kamiya, T. and Tomita, F. (2006). Foot odor due to microbial metabolism and its control. *Canadian Journal of Microbiology*, *52(4)*:357-364.

[2] Breuer, K., Haussler, S., Kapp, A. and Werfel, T. (2002). *Staphylococcus aureus* colonizing features and influence on an antibacterial treatment in adults with atopic dermatitis. *British Journal of Dermatology*, 147: 55-61.

[3] Brooks, G. F., Butel, J. S. and Morse, S. A. (2004). Normal microbial flora of the human body. Medical Microbiology. Mc Graw-Hill Companies Incooperation, New York, Pp. 196-201.

[4] Buckman, R. (2003). *Human Wildlife: The Life That Lives On Us*. The Johns Hopkins University Press, Baltimore, Pp. 93-94.

[5] Cheesbrough, M. (2005). *District Laboratory Practice in Tropical Countries*. Part 2. Cambridge University Press, Cambridge, Pp. 62-70.

[6] Chin, J. (2000). *Control of communicable diseases manual.*American Public Health Association, Washington, Pp. 460-470.

[7] Claus, W. (2007). *Body odour and body odour preferences in humans*, In: Dunbar, R. and Barret, L. (editions). The Oxford Handbook of Evolutionary Psychology, Oxford University Press. P. 226.

[8] Cogen, A. L., Nizet, V. and Gallo, R. L. (2008). Skin Microbiota: A Source of Disease or Defence. *British Journal of Dermatology*, 158(3):442-455.

[9] Ibe, S. N. and Wariso, B. A. (2005). Carriage of *Staphylococcus aureus* on armpits of secondary school and university students in Port Harcourt, Nigeria. *African Journal of Applied Zoology and Environmental Biology*, 7:125-130.

[10] Kloos, W. E. and Jorgensen, J. H. (1982). *Manual of clinical microbiology.* American Society for Microbiology. Washington, D. C. Pp. 143-153.

[11] Lundstrom, J. N. and Olsson, M. J. (2010). *Functional Neuronal Processing of Human Body Odors: Pheromones.* Academic Press, P. 4.

[12] Nester, E. W., Anderson, D. G., Roberts, C. E., Pearsall, N. N. and Hurley, D. (2004). *Skin infections.* Microbiology. Mc Graw-Hill companies Inco-operation, New York, Pp. 533-560.

[13] Raygada, J. L. and Levine, D. P. (2009). Managing CA-MRSA Infections: Current and Emerging Options. *Infections in Medicine*, 26(2).

[14] Reagan, D. R., Doebbeling, B. N., Pfaller, M. A., Sheetz, C. T., Houston, A. K., Hollis, R. J. and Wenzel, R. P. (1991). Elimination of coincident S. aureus nasal and hand carriage with intranasal application of mupirocin ointment. *Annal of Internal Medicine*, 114: 101-106.

[15] Stewart, E. S. and Beswick, T. L. S. (1997). *Bacteriology, virology and Immunology of students of medicine.* English language book society, London, Pp. 204-211.

[16] Taylor, D., Daulby, A., Grimshaw, S., James, G., Mercer, J. G. and Vaziri, S. (2003). Characterization of the microflora of the human axilla. *International Journal Cosmetics and Science*, 25:137-145

[17] Turkington, C. and Dover, J. S. (2007). *The Encyclopedia of Skin and Skin Disorders* (3rd edition). Facts on File, New York, P.36.

[18] Uzeh, R. E., Omotayo, E. A., Adesoro, O. O., Ilori, M. O. and Amund, O. O. (2012). Microbial assessment of the armpits of some selected University students in Lagos, Nigeria. *International Journal of Biological and Chemical Sciences*, 6 (6): 5022-5029.