Original Research Article

Bacterial Content of *Acne vulgaris* and Its Antimicrobial Susceptibility

ABSTRACT

Acne vulgaris is a chronic inflammatory disease associated with the pilosebaceous follicle of the skin. The development of antibiotics resistance by species of bacteria associated with this disease condition has been attributed to the indiscriminate and overuse of certain antibiotics in its treatment. This study was aimed at determining the bacterial content of Acne vulgaris and its antimicrobial susceptibility. Samples were collected under aseptic condition from fifty one (51) subjects with facial pustular and nodulocystic inflammatory lesions among Sixty (60) students of Imo state University, Owerri who participated in the study. The samples were cultured on Blood agar and Mannitol salt agar at 37°C under aerobic and anaerobic conditions for 3 to 7 days. Bacterial growths were identified by colonial morphology, gram staining and standard biochemical tests while the antibiotic susceptibility pattern of the identified organisms was determined following the enumeration of the bacterial growths. *Staphylococcus aureus* was found to be the most prevalent organism with a Mean $\log_{10}$ of CFU/ml of 5.58, while *Staphylococcus epidermidis* and *Propionibacterium acnes* have a Mean Log_{10} of CFU/ml of 5.29 and 5.33 respectively. Based on the study, it was found *Staphylococcus aureus* was sensitive to Ciprofloxacin, Gentamicin, Amoxil, Rifampicin, Chloramphenicol, Ampiclox, and Levofloxacin and was resistant to Norfloxacin, Streptomycin and Erythromycin. *Propionibacterium acnes* was found to be sensitive to Levofloxacin, Gentamycin, Rifampicin, Ciprofloxacin and Chloramphenicol and resistant to Norfloxacin, Streptomycin, Ampiclox and Erythromycin while *Staphylococcus epidermidis*

was sensitive to Rifampicin, Ciprofloxacin, Amoxil, Chloramphenicol, Ampiclox and Levofloxacin and resistant to Norfloxacin, Streptomycin and Erythromycin.

Keywords: *Acne vulgaris,* Bacteria, Skin, Antimicrobial, Acne.

1.0. INTRODUCTION

Acne vulgaris also known as acne is a chronic inflammatory multifactorial pleomorphic skin disease of the pilosebaceous follicles (PSFs) [1]. It is characterized by a variety of non-inflamed (open and closed comedones) and inflamed (macules, papules, pustules, and nodules) lesions [2]. Acne vulgaris is a long-term skin disease that occurs when hair follicles are clogged with dead skin cells and oil from the skin [3]. It is characterized by blackheads or whiteheads, pimples, oily skin, and possible scarring [4, 5, 6]. It primarily affects areas of the skin with a relatively high number of oil gland, including the face, upper part of the chest, and back [7]. The resulting appearance can lead to anxiety, reduced self-esteem and, in extreme cases, depression or thoughts of suicide [8, 9, 10].

Four major factors are involved in the pathogenesis including increased sebum production, hypercornification of the pilosebaceous duct, an abnormality of the microbial flora (especially colonization of the duct with *Propionibacterium acnes*), and the production of inflammation [11]. Microcomedones (earliest subclinical lesions) are thought to be the precursor Lesions that can then develop into non-inflamed and/or inflamed lesions. Although a common disease, the aetiology of acne is not yet fully elucidated. It carries an enormous financial and psychosocial impact. Despite extensive research on acne pathogenesis, the exact sequence of events and possible mechanisms leading to the development of a microcomedone and its transformation into an inflamed lesion has remained unclear [2].

In 2015, acne was estimated to affect 633 million people globally, making it the 8th most common disease worldwide [12]. Acne commonly occurs in adolescence and affects an estimated 80-90% of teenagers in the western world [13, 14]. Lower rates are reported in some rural societies [15, 16]. Children and adults may also be affected before and after puberty [17]. Although acne becomes less common in adulthood, it persists in nearly half of

affected people into their twenties and thirties and a smaller group continue to have difficulties into their forties [4].

Acne is not an infectious disease, but three major organisms were isolated from the surface of the skin and the pilosebaceous duct of patients with acne including *Propionibacterium acne*, *Staphylococcus epidermidis* and *Malassezia furfur* [11]. Depending on the severity of disease, the acne patients receive topical or systemic therapy, or a combination [18, 19].

Many individuals have made researches on this topic, but only a few have clearly accounted the involvement of bacteria in *Acne vulgaris*. Again, most people in our contemporary society are fully not aware of the involvement of certain bacteria in *Acne vulgaris* and at some point have abused drugs in the course of trying to treat this disease condition. Thus, this work is geared towards educating the society about the involvement of some particular bacterial organisms in *Acne vulgaris* and to add to the already existing knowledge about *Acne vulgaris*. Therefore, this work was carried out to determine the bacterial content of *Acne vulgaris* and its antimicrobial susceptibility.

2.0. MATERIALS AND METHODS

Study Area

This study was carried out in Imo State University. Imo State University is located in Owerri, which is the capital city of Imo State in Nigeria. Owerri consists of three Local Government Areas including Owerri Municipal, Owerri North and Owerri West. It has an estimated population of about 1,401,873 as of 2006 census [20] and is approximately 100 square kilometres (40 sq mi) in area. Owerri has a tropical wet climate according to the Köppen-Geiger system and the average temperature is 26.4 °C.

Study Population

Following approval by the ethics committee and obtaining informed consents, a total of 60 subjects (aged between 15 and 35 years) who are students from Imo state University, Owerri participated in this study. The exclusion criteria for this study were manifestation of any other skin disease such as eczema, ringworms or any form of dermatitis, and those on any form of antibiotics.

Sample Collection and Processing

The collection of the samples was carried out under aseptic condition. Following careful examination of the subjects, the pustular and nodulocystic inflammatory acne lesions on the face of the subjects with acne vulgaris were collected after disinfecting the surface with a cotton wool soaked in 75% alcohol using sterile disposable swab. Each of the samples was labeled appropriately with the individuals name, age, sex, and date of collection. The collected samples were inserted into a normal saline and sent to the microbiological laboratory where bacteriological analysis was conducted.

Cultivation and Identification of the Organisms

The method used for culture was according to Hassan *et al*., [21]. The samples were cultured on blood agar and Mannitol salt agar at 37°C under aerobic and anaerobic conditions for 3 to 7 days. Enumeration of the bacterial growths on the individual samples was done following the identification of the bacteria by their colonial morphology, Gram staining, and standard biochemical tests.

Determination of Antibiogram

Antimicrobial susceptibility test was performed on the bacteria isolates using disc diffusion method and the zone of inhibition of the antibiotics to the isolated organisms was measured by placing a transparent rule on the underside of the plate and measuring the diameter of the clear zone around the susceptibility disc.

Data Analysis

The data obtained from this study were analyzed statistically using frequency distribution tables and simple percentages and the results were represented graphically using pie charts and bar charts.

3.0. RESULTS

Sixty (60) subjects from Imo State University participated in this study. Of the sixty subjects, Fifty one (51) persons of whom samples were collected from after careful facial examination, were observed to have pustular and nodulocystic inflammatory acne lesions while Nine (9) persons were without acne. The subjects fall within the age group 15 – 20 years, 21 – 25years, 26 – 30 years and 31 – 35 years. The numbers of subjects from these respective age groups were 3, 45, 9 and 3. Of these numbers 2, 40, 6 and 3 were the number of the subjects found to have acne while 1, 5, 3 and 0 were the number of subjects found to be without acne. The prevalence of *Acne vulgaris* based on these age groups was therefore gotten to be 3%, 67%, 10% and 5%.

The number of the male subjects was 51, 41 were the number with acne while 7 was the number of those without acne. The numbers of females were 9, 7 were with acne while 2 were without acne. Thus the prevalence of acne was gotten to be 73.3% for males and 11.7% for females.

The Mean of $\log_{10}$ of the colony forming unit of the isolated organisms were derived from the respective viable count and colony forming unit of the samples in which the organisms are distributed, after the organisms have been morphologically and biochemically identified. *Staphylococcus aureus*, have a Mean $\log_{10}$ of the colony forming unit of 5.58 while *Staphylococcus epidermidis* and *Propionibacterium acnes* have Mean $\log_{10}$ of the colony forming units of 5.29 and 5.33.

From the antimicrobial susceptibility, *Staphylococcus aureus* was sensitive to Ciprofloxacin, Gentamicin, Amoxil, Rifampicin, Chloramphenicol, Ampiclox, and Levofloxacin and resistant to Norfloxacin, Streptomycin and Erythromycin. *Propionibacterium acnes* was sensitive to Levofloxacin, Gentamycin, Rifampicin, Ciprofloxacin and Chloramphenicol and resistant to Norfloxacin, Streptomycin, Ampiclox and Erythromycin while *Staphylococcus epidermidis* was sensitive to Rifampicin, Ciprofloxacin, Amoxil, Chloramphenicol, Ampiclox and Levofloxacin and resistance to Norfloxacin, Streptomycin and Erythromycin.

Figure 1 shows a pie chart showing the prevalence rate of *Acne vulgaris* among the subjects age group. From the pie chart, 15 – 20 years, 21 – 25 years, 26 – 30 years, and 31 – 35 years, were the age groups of the subjects from which the samples were collected. The prevalence of *Acne vulgaris* among these age groups was gotten to be 3%, 67%, 10% and 5%, respectively (See Figure 1).

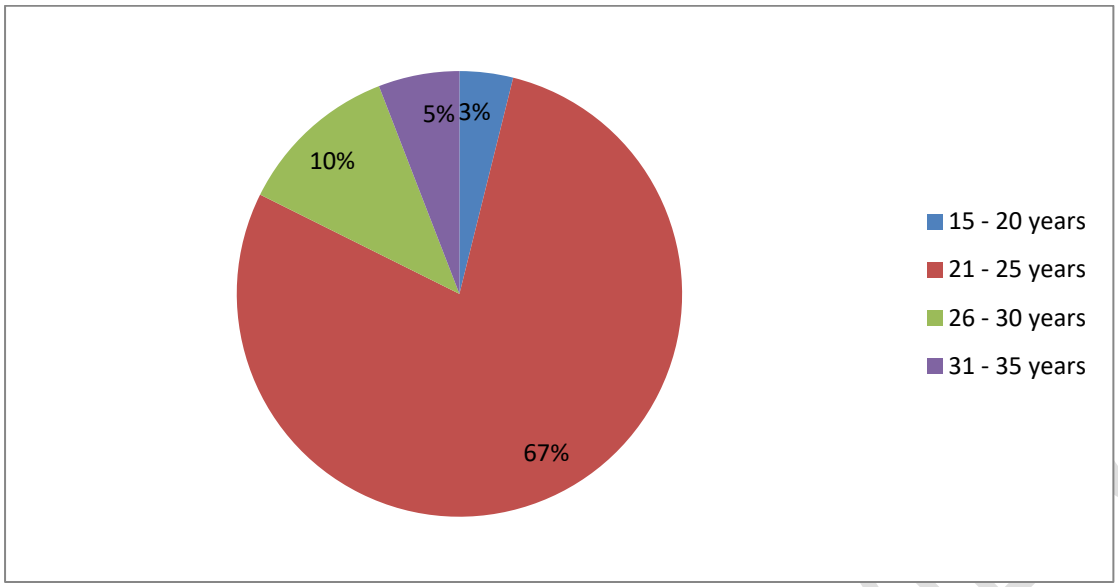


Figure 1: A pie chart showing the prevalence rate of *Acne vulgaris* among subjects age group.

Figure 2 shows a pie chart showing the prevalence of *Acne vulgaris* among the subjects sex. From the pie chart, the male subjects in the study area were found to have 73.3% prevalence rate of the disease (*Acne vulgaris*) while the female subjects were found to have 11.7% prevalence of the disease.

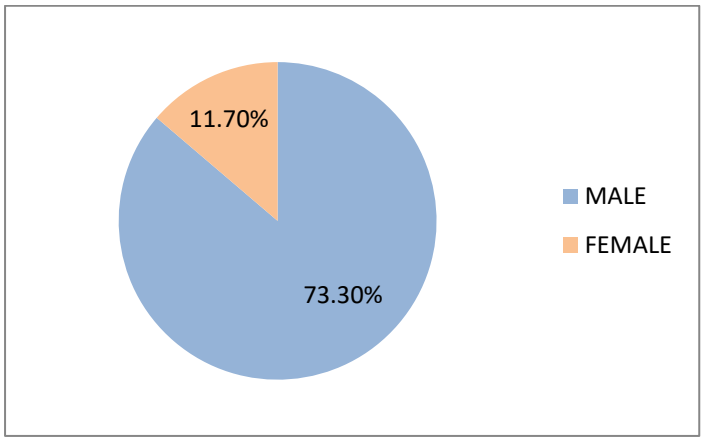


Figure 2: A pie chart showing the prevalence rate of *Acne vulgaris* among the subjects sex.

Figure 3: Shows a bar chart with Mean Log_{10} of CFU/ml of the Isolated Organisms plotted against the sample distributions. From the bar chart, *Staphylococcus aureus* was found to be distributed within sample 1 – 21 and have a Mean Log_{10} of CFU/ml of 5.58 while *Staphylococccus epidermidis* and *Propionibacterium acnes* were distributed within samples 22 – 32 and 33 – 51 and have a Mean Log_{10} of CFU/ml of 5.29 and 5.33 respectively.

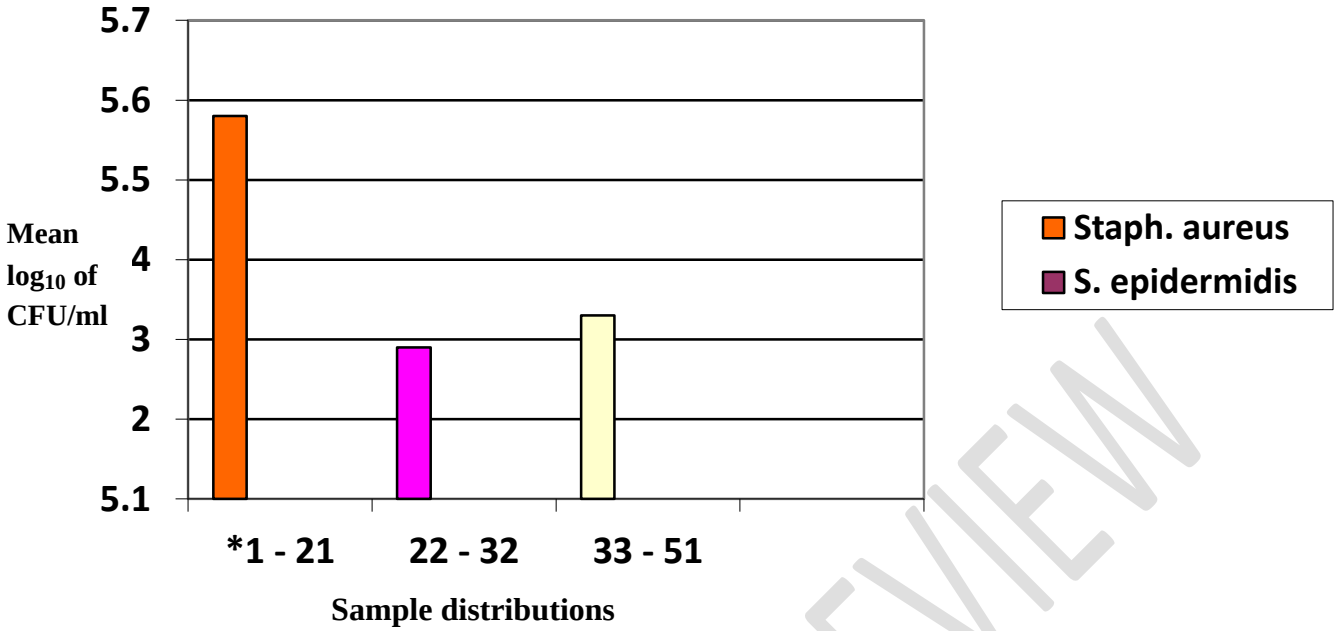
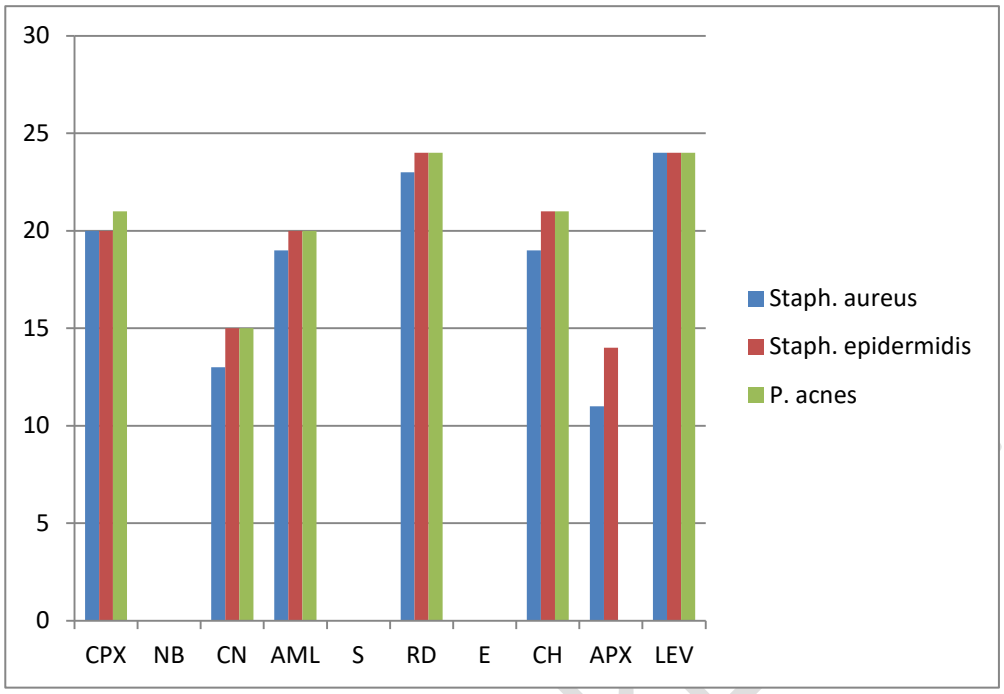


Figure 3: A bar chart with Mean Log_{10} of CFU/ml of the Isolated Organisms plotted against the sample distributions.

Figure 4: Shows a bar chart with the zone of inhibition (mm) of the antibiotics to the isolated organisms plotted against the antibiotics. From the bar chart, *Staphylococcus aureus* was found to be sensitive to Ciprofloxacin, Gentamicin, Amoxil, Rifampicin, Chloramphenicol, Ampiclox, and Levofloxacin and resistance to Norfloxacin, Streptomycin and Erythromycin. *Propionibacterium acnes* was found to be sensitive to Levofloxacin, Gentamycin, Rifampicin, Ciprofloxacin and Chloramphenicol and resistance to Norfloxacin, Streptomycin, Ampiclox and Erythromycin while *Staphylococcus epidermidis* was sensitive to Rifampicin, Ciprofloxacin, Amoxil, Chloramphenicol, Ampiclox and Levofloxacin and resistance to Norfloxacin, Streptomycin and Erythromycin .



Key notes: CPX = Ciprofloxacin, NB = Norfloxacin, CN = Gentamicin, AML = Amoxil, S = Streptomycin, RD = Rifampicin, E = Erythromycin, CH = Chloramphenicol, APX = Ampiclox, LEV = Levofloxacin.

Figure 4: A bar chart with the zone of inhibition (mm) of the antibiotics to the isolated organisms plotted against the antibiotics.

4.0. DISCUSSION

Acne vulgaris is a non - infectious skin disease condition that occurs more commonly in adolescence (young adults) and less commonly in adults [13, 4, 11]. From the study, it was found that the age group 21 – 25 years had the highest prevalence of the disease which is

67%, followed by the age group 26 – 30 years which had the prevalence to be 10% with the least being the age group 31 – 35 years and 15 – 20 years which had their prevalence to be 5% and 3% respectively. This was in agreement with Kong and Tey [22] and Holzman and Shakery [23] who found acne as being remarkable between the ages of 21 – 25 years. This however is due to an increase in Hormonal activity, such as the sex hormones called androgens, the growth hormone (GH) and insulin-like growth factor 1 (IGF-1), within this age group, which causes the skin follicle glands to grow larger and make oilier sebum which thus contributes in the formation of acne. Still from the study, it was found that the males were the sex most affected by the disease with a prevalence rate of 73.7% while the females were the least affected with a prevalence rate of 11.7%. This is in contrast to Ikaraoha *et al* [8], who found the occurrence of acne to be higher in females (65.0%) compared to the males (35.0%). The reason for having the prevalence rate of the disease to be higher in males than in females is due to the fact that most female students of the University uses some cosmetic creams and makeups such (as skineal, visita and zaron, e.t.c), which contains ingredients that have anti – inflammatory and antibacterial capacity and thus can inhibit the occurrence of acne.

As regards to the prevalence of the organisms isolated, it was found that *Staphylococcus aureus* was the most prevalent organism with a Mean $\log_{10}$ of CFU/ml of 5.58, while *Staphylococcus epidermidis* and *Propionibacterium acnes* have a Mean Log_{10} of CFU/ml of 5.29 and 5.33 respectively. This suggests that *Staphylococcus aureus* could be the leading cause of *Acne vulgaris*. This was in agreement with Dhillon and Krati [24] who found *Staphylococcus aureus* as their most common bacteria isolates in acne samples. But it's in contrast to Nakase *et al* [25], who implicated *Staphylococcus epidermidis* and *Propionibacterium acnes* as bacteria frequently isolated from lesions caused by *Acne*

vulgaris. This discrepancy in the distribution of the bacteria in acne may be explained to be as a result of differences in the geographical regions which may affect the bacteria involved in *Acne vulgaris*.

As regards to the antibiotic susceptibility pattern of the isolated organisms, *Staphylococcus aureus* was found to be sensitive to Ciprofloxacin, Gentamicin, Amoxil, Rifampicin, Chloramphenicol, Ampiclox, and Levofloxacin and resistance to Norfloxacin, Streptomycin and Erythromycin. *Propionibacterium acnes* was found to be sensitive to Levofloxacin, Gentamycin, Rifampicin, Ciprofloxacin and Chloramphenicol and resistance to Norfloxacin, Streptomycin, Ampiclox and Erythromycin while *Staphylococcus epidermidis* was sensitive to Rifampicin, Ciprofloxacin, Amoxil, Chloramphenicol, Ampiclox and Levofloxacin and resistance to Norfloxacin, Streptomycin and Erythromycin .

This was in agreement with Tafin *et al* [26] and Dhillon and Krati [24], who found Rifampicin to be an effective antibiotic against *Propionibacterium acnes* and *Staphylococci strains*. It was also in agreement with Cafisco *et al* [27] and Hassan *et al* [11], who found Levofloxacin as to be effective antibiotic against coagulase positive and negative Staphylococci species and *Propionibacterium acnes*.

The development of resistance to some of these antibiotics could be inferred to be due to the indiscriminate and overuse of these antibiotic drugs in the treatment of acne.

CONCLUSION

The findings of this study have shown the species of *Staphylococci* (*Staphylococcus aureus* and *Staphylococcus epidermidis*) and *Propionibacterium acnes* to be associated with *Acne vulgaris*. From the antimicrobial susceptibility test, it was also revealed that each of the organisms had one or more classes of antibiotics they were sensitive or resistant to.

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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