Bacterial identification of acne vulgaris

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ABSTRACT

Background: Acne vulgaris (AV) is a chronic inflammation of the pilosebaceous unit with clinical polymorphic lesion consist of non-inflammatory (open and closed comedones) and inflammatory lesions (papules, pustules, and nodules) with varying degree of inflammation and depth. Earlier studies showed that other bacteria might also found and played a role in acne pathogenesis besides *Cutibacterium acnes (C. acnes)*.

Patients and Methods: This descriptive observational study used cross-sectional method. Samples were collected from 40 subjects with AV. We took the samples from non-inflammatory (closed comedones) and inflammatory lesions (pustule) in each subject, followed by Gram staining, aerobic and anaerobic bacterial culture, and bacterial identification. This research has been approved by

the Ethical Committee, Faculty of Medicine, University of Sumatera Utara.

Results: There were 12 bacterial species that were identified from 80 samples. We identified *Cutibacterium acnes* (21,2%) in anaerobic culture. While in aerobic culture, we identified *Staphylococcus epidermidis* (47,5%), *Staphylococcus hominis* (17,5%), *Staphylococcus aureus* (8,7%), *Staphylococcus haemolyticus* (8,7%), *Leuconostoc mesentroides* (6,2%), *Micrococcus luteus* (3,7%), *Kocuria varians* (2,5%), *Staphylococcus vitulinus* (1,2%), *Staphylococcus cohnii* (1,2%), *Staphylococcus arlettae* (1,2%) and *Dermacoccus nishinomyaensis* (1,2%).

Conclusion: The two most common bacteria in acne vulgaris are *Staphylococcus epidermidis* and *Cutibacterium acnes*

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INTRODUCTION

Acne vulgaris (AV) is a chronic inflammation of the pilosebaceous unit with clinical polymorphic lesion consist of non-inflammatory (open and closed comedones) and inflammatory lesions (papules, pustules, and nodules) with varying degree of inflammation. AV commonly is self-limiting disease and often found in the adolescence period.[1-3] Although AV is a self-limiting disease, it can cause sequels such as scar tissue and pigmentary changes that can persist for a lifetime and decrease the Patient's quality of life and cause the psychological disorder.[1,2,4,5]

The pathogenesis of AV is multifactorial, consists of hyperproliferation of infundibulum, excess sebum production, inflammation, and colonisation of *Propionibacterium acnes*.[1] Previous studies reported that other bacteria found and played a role in the pathogenesis of AV besides *Propionibacterium acnes*.[6] Genomic and metagenomic investigations recently led to change in Propionibacterium acnes' denomination to *Cutibacterium acnes* (*C. acnes*).[7]

Various studies demonstrated that other bacteria, namely *Staphylococcus epidermidis*, *Staphylococcus aureus*, *Micrococcus spp.* were related to the

pathogenesis of AV other than *Propionibacterium acnes*.[8] However, the role of these microorganisms in pathogenesis AV is still controversial.[9,10]

This study was undertaken to identify bacterial anaerobically and aerobically patterns from the non-inflammatory lesion (closed comedones) and the inflammatory lesion (pustule) of AV patients.

METHODS

This study has been approved from the Ethics Commission in Faculty of Medicine University of Sumatera Utara/Adam Malik Hospital number 807/TGL/KEPK FK USU-RSUP HAM/2019. This study was a descriptive cross-sectional study involving 40 acne vulgaris subjects of Dermatology and Venereology outpatient Clinic University of Sumatera Utara Hospital from October till December 2019. Acne vulgaris diagnosed based on history taking and physical examinations. Inclusion criteria were patients who have been diagnosed with acne vulgaris, age more than 18 years, and signed informed consent. The exclusion criteria included pregnancy and breastfeeding, received a systemic antibiotic in the last two weeks, and topical antibiotic in the previous one weeks.

Patient demographic data included age and gender taken from medical records history. The researcher recorded patient demographic data included age, gender, phone number, and address. Then, the Patient signed informed consent. Sample collected from each non-inflammatory (closed comedones) and inflammatory lesions (pustule) in each subject. Specimens were taken using a sterilized comedones extractor. Then, using a sterilized swab moistened with nutrient broth. The specimens were immediately split into anaerobic and aerobic conditional treatments in jars containing blood agar and Brucella blood agar. Impression smears were taken on a clean slide for Grams staining. The samples were immediately inoculated on, then incubated both anaerobically and aerobically at 37°C for 24 – 48 hours. An AnaeroGen® Compact sachet was placed into each anaerobic jar for the isolation of anaerobic bacteria. No sachets were used for the isolation of aerobic bacteria. Bacterial identification performed with Vitek® 2 compact (Biomerioux, France).

Data that has been collected then processed using statistical package for the social sciences (SPSS) version 23.0 with descriptive statistical analysis for age, gender, anaerobic and aerobic bacteria distribution.

RESULTS

The Patient's demographic characteristics are shown in Table 1. Most patients were in the age group 18-25 years old with 29 subjects (72,5%), the age group 26-35 years old with 11 subjects (27,5%). Based on gender, the female gender was dominant (65%) compare to the male gender (35%).

The bacterial distribution of subjects describes in Table 2. Twelve bacterial species were identified from 80 samples. We identified *Cutibacterium acnes* in 17 samples (21,2%) in anaerobic culture. While in aerobic culture, we identified *Staphylococcus epidermidis (S. epidermidis)* in 38 samples (47,5%), *Staphylococcus hominis (S. hominis)* in 14 samples (17,5%), *Staphylococcus aureus (S. aureus)* in 7 samples (8,7%), *Staphylococcus haemolyticus (S. haemolyticus)* in 7 samples (8,7%), *Leuconostoc mesenteries* in 5 samples (6,2%), *Micrococcus luteus* in 3 samples (3,7%), *Kocuria varians* in 2 samples (2,5%), *Staphylococcus vitulinus (S. vitulinus)* in 1 sample (1,2%), *Staphylococcus cohnii (S. cohnii)* in 1 sample (1,2%), *Staphylococcus arlettae (S. arlettae)* in 1 sample (1,2%) and *Dermacoccus nishinomyaensis* in 1 sample (1,2%)

The most common bacteria found in anaerobic culture was *Cutibacterium acnes* (21,2%), wherein aerobic culture was *Staphylococcus epidermidis* (47,5%). The most common mixed growth bacteria found in AV lesions was *Cutibacterium acnes* concomitant with *Staphylococcus epidermidis* (13,7%).

DISCUSSION

Total of 40 AV patients were involved in this study with a total of 80 samples. Acne vulgaris mostly found in the age group 18-25 years old, with primarily female gender affected. Skroza et al. also reported that 12 – 25 years old was the majority age group of AV patients.[11] Eyaboglu et al. also reported in their study that females were more affected by AV than males.[12]

In this study, we found 12 bacterial species from 80 samples. This study supported previous studies' results because not only *Cutibacterium acnes*, but other bacteria were also found from AV lesions. The two most common bacteria are *Staphylococcus epidermidis* (47,5%) and *Cutibacterium acnes* (21,2%). Overall, we found *Cutibacterium acnes, Staphylococcus epidermidis, Staphylococcus hominis,*

Table 1. Demographic characteristics of subjects

Characteristics	Frequency (n=40)	
	number (n)	percentage (%)
Gender		
Male	14	35
Female	26	65
Age		
18-25 years old	29	72,5
26-35 years old	11	27,5
Total	40	100

Table 2. Bacterial distribution of acne vulgaris

Bacteria	Frequency (n=80)	
	number (n)	percentage (%)
Anaerobic bacteria		
C. acnes	17	21,2
Aerobic bacteria		
S. epidermidis	38	47,5
S. hominis	14	17,5
S. aureus	7	8,7
S. haemolyticus	7	8,7
Leuconostoc mesentroides	5	6,2
Micrococcus luteus	3	3,7
Kocuria varians	2	2,5
S. vitulinus	1	1,2
S. cohnii	1	1,2
S. arlettae	1	1,2
Dermacoccus nishinomyaensis	1	1,2
Mixed growth		
C. acnes and *S. epidermidis*	11	13,7
C. acnes and *S. hominis*	3	3,7
C. acnes and *S. aureus*	2	2,5
C. acnes and *S. haemolyticus*	1	1,2
Total	80	100

Staphylococcus aureus, Staphylococcus haemolyticus, Leuconostoc mesentroides, Micrococcus luteus, Kocuria varians, Staphylococcus vitulinus, Staphylococcus cohnii, Staphylococcus arlettae and *Dermacoccus nishinomyaensis.*

This study has a similar finding with a study conducted by Moon et al. found that the most common bacteria in AV lesion was *Staphylococcus epidermidis* (36%) followed by *Propionibacterium acnes* (30%).[13] Sylvia et al. also reported that the most common anaerobic bacteria in AV lesion was *Propionibacterium acnes* (46,2%), while the most common aerobic bacteria were *Staphylococcus epidermidis* (47,6%).[14] Another study also reported similar finding conducted by Srikanth et al. where the most common anaerobic bacteria in AV lesion was *Propionibacterium acnes.* In contrast, the most common aerobic bacteria was *Staphylococcus epidermidis.*[15] Another study conducted by Syahrial et al. also identified bacteria in comedones lesion, where the most frequent bacteria found was *Propionibacterium acnes* (37,2%), followed by *Staphylococcus epidermidis* (30,2%).[16]

In this study, the two most common bacteria found were *Cutibacterium acnes* concomitant with *Staphylococcus epidermidis.* This similar finding also found in the study of Biswal et al. from all AV samples, 13.75% of samples showed mixed growth (aerobic and anaerobic growth), and the most common mixed growth was *Propionibacterium acnes* and *Staphylococcus epidermidis.*[17]

Acne vulgaris is one of the most common skin diseases, predominantly seen in adolescence and also a multifactorial disease in which *Cutibacterium acnes* is thought to play an essential role in the pathogenesis of inflamed lesions.[7] *Propionibacterium acnes* is a gram-positive and anaerobic bacteria that colonies in the human skin's sebaceous glands and hair follicles.[18] The pathogenesis of AV is based on multiple factors, such as increased sebum production, *Propionibacterium acnes* proliferation, and inflammation.[18]

The role of bacteria other than *Propionibacterium acnes* in the pathogenesis of AV is still controversial.[19] Lipase (geh1 gene) and delta hemolysin (hld gene) are two virulence factors produced by *Staphylococcus epidermidis* in AV development that impacts acne inflammation.[20] *Staphylococcus epidermidis* also secreted exopolysaccharide intercellular adhesin (PIA), which is responsible for biofilm formation and protects them against significant human innate host components defense. This biofilm provides favorable anaerobic conditions to grow *Propionibacterium acnes.*[19,21-23]

CONCLUSION

This study concluded that the two most common bacteria identified in acne vulgaris are *Staphylococcus epidermidis* and *Cutibacterium acnes.*

ABBREVIATIONS

AV, acne vulgaris; *C. acnes, Cutibacterium acnes;* geh1 gene, lipase; hld gene, hemolysin; *P. acnes, Propionibacterium acnes*; PIA, exopolysaccharide intercellular adhesin; *S. aureus, Staphylococcus aureus; S. epidermidis, Staphylococcus epidermidis*; *S. haemolyticus, Staphylococcus haemolyticus*; *S. hominis, Staphylococcus hominis*; SPSS, statistical package for the social sciences.

AUTHOR CONTRIBUTION

All authors have contributed to all processes in this research, including preparation, data gathering, and analysis, drafting, and approval for publication of this manuscript.

CONFLICT OF INTEREST

The authors declare no conflict of interest regarding the publication of this article.

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