

Bacteriological study of Acne Vulgaris in Cairo Egypt

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ABSTRACT

Background: acne vulgaris is chronic inflammatory dermatosis of the pilosebaceous units which is characterized by open and/or closed comedones (blackheads and whiteheads) and inflammatory lesions including papules, pustules, or nodules, in some cases, is accompanied by scarring. Acne is multifactorial disease including increased sebum production, hypercornification of pilosebaceous duct, an abnormality of microbial flora especially colonization of the duct with propionibacterium acne and the production of inflammation. P.acne has been implicated in the pathogenesis of acne for more than 100 years.

Objective: the present work was done to determine bacteria involved in acne vulgaris among Egyptian acne patients.

Patients and Methods: this study was conducted on one hundred patients with inflammatory acne vulgaris recruited from the Dermatology outpatient clinic of Al-Hussien hospital.

Results: in this study, S.epidermidis was higher in normal skin than in acne lesion. But P.acne was higher in acne lesion than in normal skin with significant statistical difference. There was a significant relation between type of organisms isolated and severity of acne lesions in both aerobic and anaerobic state. There was a significant relation between positive acne family history and liability for post acne scar. Even though there was no significant relation between hermonal therapy and severity of acne lesions. We noticed that the majority of female patients with POP history had moderate acne lesions. In this study, the high percentage of S.epidermidis was observed in moderate acne lesion and in normal skin in aeorobic state but high percentage of P.acne was found in moderate acne lesions in anaerobic state. However, P.acne can not be solely held responsible for the initiation of inflammation in inflamed acne lesions or for the variation in its severity.

Conclusion: the high percentage of no growth result in this study directs us that the antibiotic treatments are not the most useful treatment for acne and there are other effective methods for treatment whether topical as retenoids or ablative as Lasers. Patient with family history of acne lesion especially first relative (mother) we must keep in mind that they are more susceptible for post acne scar so we should start with the perfect treatment as early as possible to prevent the post acne sequels.

Keywords: 1,25 dihydroxy vitamin D3, Dihydrotestosterone, Propionibacterium acnes, Pilosebaceous units

INTRODUCTION

Acne vulgaris is a chronic inflammatory dermatosis of the pilosebaceous units which is characterized by open and/or closed comedones (blackheads and whiteheads) and inflammatory lesions including papules, pustules, or nodules, in some cases, is accompanied by scarring ⁽¹⁾.

Reports vary, but incidence rates suggest that acne is more frequent and severe during adolescence (70% to 87% of the adolescent population), although onset may be delayed until the age of 30 or 40. The frequency of acne in the adolescent population increases with age and frequently, but not always, resolves in adulthood ⁽²⁾.

The etiology of acne appears to be complicated and is not completely understood. The pilosebaceous unit is the target organ in acne. This explains the distribution of acne in regions such as face, neck, and the upper parts of the chest and back, specific areas that have a high density of sebaceous glands ⁽²⁾.

The pilosebaceous unit possesses sebocytes that synthesize and accumulate lipids and express a

variety of cytokines and are implicated in inflammatory and immune responses. The pathogenesis of acne includes hormonal, inflammatory, and immunologic mechanisms. Several major pathophysiologic factors (mainly endogenous) influence the development of acne lesions ⁽²⁾: Hyperplasia of sebaceous glands with increased sebum production (seborrhea) as a reaction to androgenic hormone stimuli, modification of the quality of sebum lipids, regulation of skin steroidogenesis, disturbance of growth and differentiation of sebaceous follicles with ductal hypercornification and formation of plugs of cornified cells in the dilated pilosebaceous lumen, hypercolonization of the follicle from Propionibacterium acnes (p.acne). Rupture of the obstructed pilosebaceous duct leading to inflammation.

Acne treatment options are vast and include a number of non-hormonal and hormonal therapies. When deciding on the acne treatment regimen, one should take into account individual patient factors including disease state, predominant lesion type and severity, pre-existing medical conditions, desired treatment mode topical vs. oral, and endocrine treatment ⁽³⁾.

Although acne is not infectious disease three major organisms have been isolated from the pilosebaceous ducts of acne patients including *Staphylococcus epidermidis*, *Malassezia furfur* and *Propionibacterium acnes*. Among these different organisms recovered from pilosebaceous units, *P. acnes* is the most important one in the pathogenesis⁽⁴⁾.

Antibiotics have been used more than 40 years to treat acne vulgaris. *P. acnes* antibiotic resistance was reported in USA in 1979 and then in other countries in the world. Successful and early treatment of acne is very important and delay or failure in treatment causes probably reversible sequelae⁽⁴⁾.

AIM OF THE WORK

The present work is done to determine bacteria involved in acne vulgaris patients in Cairo Egypt.

METHODOLOGY

This study was conducted on one hundred patients (33 males and 67 females) with inflammatory acne and normal skin from the same patients, attending the outpatient clinic of Dermatology of Al-Hussien hospital. Their age ranged from 18 to 35 years. Cases collected from January 2017 to January 2018. The duration of the disease ranged from two months to six years.

Patients (n=100):

Inclusion criteria: The patients with acne vulgaris in the age group of (18-35 years). Patients with inflammatory acne lesion (papules, pustules and nodulocystic), the patient must have pus lesion. Patients don't receive topical or systemic treatments for acne at least one month prior to the study.

Exclusion criteria: Patients with other forms of acne as: comedonal acne, acne coglobata and acne fulminans, pregnancy and lactation, systemic diseases: liver cell failure and renal cell failure.

The patients were informed about the nature of investigation. A written consent was taken from each patient.

Methods

All the patients were subjected to the following:

1- Detailed medical history: Personal history regarding name, age, sex as well as occupation. History of present illness regarding the onset, course, duration and progression of the

disease. History of drug intake for acne (topical or systemic). History of menstrual irregularities (in the form of amenorrhea more than three months duration or menstruation more than 7 days from a standard 28-day cycle over three consecutive cycles), presence of hirsutism, androgenic alopecia. History of hormonal treatment. Family history for the presence of acne and its severity. **The study was approved by the Ethics Board of Ain Shams University and an informed written consent was taken from each participant in the study.**

2- Dermatological Examination:

included: The type of acne lesion: papules, pustules, nodules, cysts. Distribution of lesion: face, back, chest, and upper limbs. Degree of severity: There were 16 cases presented with mild acne, 84 cases presented with moderate acne and 2 cases with severe acne. Acne severity is rated as regard the Combined Acne Severity classification that classifies acne according to the number and type of lesions, into: **Mild acne:** fewer than 20 comedones, or fewer than 15 inflammatory lesions or a total lesion count lower than 30. **Moderate acne:** 20-100 comedones, or 15-50 inflammatory lesions or a total lesion count of 30-125. **Severe acne:** More than 5 cysts or comedones count greater than 100, or a total inflammatory count greater than 50, or a total lesion count greater than 125.

Sampling and culture: The samples were taken from pustular lesions of 100 acne patients. The skin was first cleansed with 70% ethanol and leaves to dry. The material was extracted from pustular lesions took up by sterile swab. Samples were transported to the microbiology laboratory immediately. In the laboratory, each specimen was inoculated on two plates of Blood agar, one of which was incubated in aerobic condition at 37°C for 48h and the other one was incubated in anaerobic jar for 2-7 days at 37°C.

Identification of isolates: After the bacteria cultured have been obtained in pure growth, the colonies were identified according to the standard bacteriological methods. *P. acnes* colonies were identified by Gram stain and specific tests: catalase test, indole test and gelatin test. *Staphylococcus aureus* colonies were identified by Gram stain and the following specific tests: Catalase test, Coagulase test (slide test and tube test), Gelatin liquefaction test and Mannite fermentation.

Statistical analysis

The collected data were tabulated and analyzed by the suitable statistical methods using computer program SPSS (Statistical package for social science) version 16 to obtain:

1- Descriptive data

Descriptive statistics were calculated for the data in the form of: 1. Mean. 2. Standard deviation (\pm SD).

2- Analytical statistics

In the statistical comparison between the different groups, the significance of difference was

tested using one of the following tests: 1- Student's *t*-test: used to compare between mean of two groups of numerical (parametric) data. 2- Test of proportion z-test: used to compare between the two percentages of the variables of the study groups.

The level of significance: *P* value <0.05 was considered statistically significant (S), *P* value <0.001 and *P*<0.01 was considered highly significant (HS) and *p* value >0.05 was considered insignificant.

RESULTS

Table (1): Statistical data analysis of general sheet items

		Count	%
Sex	Male	33	33.0%
	Female	67	67.0%
Smoking	Positive	47	47.0%
	Negative	53	53.0%
History of hormonal therapy	POP	28	28.0%
	Non	39	39.0%
History of acne treatment	topical and systemic	27	27.0%
	Topical	43	43.0%
	Non	30	30.0%
Site	back & chest	18	18.0%
	Face	51	51.0%
	face & other sites	31	31.0%
post acne scar	Positive	33	33.0%
	Negative	67	67.0%
History of family acne	Positive	34	34.0%
	Negative	66	66.0%
History of menstrual irregularity	Yes	19	28.4%
	No	48	71.6%
type of acne severity	Mild	16	16.0%
	Moderate	82	82.0%
	Sever	2	2.0%

Table (2): Mean and Median of age and duration of disease

	Mean	Standard Deviation	Median	Minimum	Maximum
Age	23.36	4.84	22.00	18.00	35.00
Duration (months)	29.27	20.46	24.00	2.00	96.00

Table (3): Culture results of both normal skin and acne lesions in aerobic and anerobic states.

	Staph epidermidis		Staph aureus		Propionibacterium acne		Micrococcus		No growth	
	Count	Row N %	Count	Row N %	Count	Row N %	Count	Row N %	Count	Row N %
Type of organism in acne lesion aerobic	39	39.0%	14	14.0%	0	.0%	4	4.0%	43	43.0%
type of organism in acne lesion anaerobic	23	23.0%	10	10.0%	56	56.0%	0	.0%	11	11.0%
Type of organism in normal skin aerobic	42	42.0%	0	.0%	0	.0%	4	4.0%	54	54.0%
Type of organism in normal skin anaerobic	44	44.0%	0	.0%	25	25.0%	0	.0%	31	31.0%

DISCUSSION

Acne vulgaris (acne) is the most common skin disorder seen in ambulatory dermatology practices regardless of gender, skin color or ethnicity ⁽⁵⁾.

Acne Vulgaris is a common, chronic, inflammatory, facial skin disorder that can affect individuals from any race, ethnicity, or cultural background, the onset of AV is usually shortly before or during early adolescence; however, some cases start in latter childhood with the subset of preadolescent acne defined within the ages of 7 and 11 years ⁽⁶⁾.

Acne is estimated to affect 9.4% of the global population, making it the eighth most prevalent disease worldwide. Epidemiological studies have demonstrated that acne is most common in postpubescent teens, with boys most frequently affected, particularly with more severe forms of the disease ⁽⁷⁾.

Acne vulgaris is a common dermatological disorder that predominantly affects teenagers, but can also affect preadolescents and post-teen individuals ⁽⁸⁾.

Acne vulgaris is a disorder of the pilo-sebaceous unit caused primarily by increased sebum secretion and follicular plugging. Acne variants can affect people of all age groups, but it is most prevalent in adolescence. Over 90% of males and 80% of females have experienced acne by the age of 21 ⁽⁹⁾.

Etiology of acne vulgaris is multifactorial, with several pathophysiologic associations identified, including hormonal function, increased sebum production, follicular hyperkeratinization, proliferation of *Propionibacterium acnes* (*P acnes*), and various cascades of inflammation ⁽⁵⁾.

Acne is a chronic inflammatory disease of the pilosebaceous unit resulting from androgen-induced increased sebum production, altered keratinisation, inflammation, and bacterial colonisation of hair follicles on the face, neck, chest, and back by *Propionibacterium acnes*. Although early colonisation with *P acnes* and family history might have important roles in the disease, exactly what triggers acne and how treatment affects the course of the disease remain unclear ⁽¹⁰⁾.

Hormones play a central role in the stimulation of sebaceous glands and development of acne. Sebaceous gland size and metabolic rate are directly stimulated by dihydrotestosterone, a derivative of testosterone (an androgenic/sex hormone) ⁽⁹⁾.

There is no ideal treatment for acne, although a suitable regimen for reducing lesions can be found for most patients. Good quality evidence on comparative effectiveness of common topical and systemic acne therapies is scarce ⁽¹⁰⁾.

The mean of our patients' age was (23.36 ± 4.84 years.) all of them were above 18 years old. There were 67 females and 33 males with female predominance (67%) this comes in accordance with

Bagatin *et al.* ⁽¹¹⁾ where 452 adolescents aged between 10 and 17 years are included in his study 62.4 % of them were females although it did not reveal a significant association with type of acne severity ($p > 0.05$). Another study by *Ismail et al.* ⁽¹²⁾ who found the same in his study on included 510 patients (173 males and 337 females) with a male: female ratio of 0.41:1; but it was against *Al Kubaisy et al.* ⁽¹³⁾ who made a cross-sectional study conducted in the Syrian International University for Science and Technology, a sample of 500 students was chosen and found that males have higher prevalence of acne compared to females (42.9% versus 23.6%, $P < 0.0001$) may be due to increased sebum production. Also *Tan and Bhate* ⁽⁷⁾ study results revealed that male patients were more frequently affected, particularly with more severe forms of acne. This doesn't mean that acne is more common in female but it means that girls are more aware about their facial appearance than boys, and they seek for therapy ⁽¹²⁾.

In the present study the mean duration of acne in the study population was 29.27 months, similarly *Khodaeiani et al.* ⁽¹⁴⁾ results on 78 young adults (age range: 18–30 years) and found that the mean duration of acne in the study population was 3.03 ± 1.54 years. Another study by *Al-Ameer et al.* ⁽¹⁵⁾ who reported that the duration of the disease at presentation ranged from 6 months to 7 years, with a mean of 35.3 ± 25.2 months for males (median, 24.0 months) and 40.2 ± 25.1 months for females (median, 36.0 months). There was no significant difference between the genders ($P = 0.20$). This comes in accordance with the conclusion of acne review done by *Dawson & Dellavalle* ⁽¹⁶⁾ who reported that nearly 90% of teenagers in his study have acne, and half of them continue to experience symptoms as adults.

Most *acne* patients present with localised lesions on the face, neck and truncal areas. However, *acne vulgaris* can affect all areas of the body except the palms of the hands and soles of the feet, as these regions do not have sebaceous glands ⁽¹⁷⁾.

Regarding our clinical examination of distribution sites of acne, our results showed that 51 % of our patients had a facial distribution of acne, 18 % had back and chest acne. This comes in accordance with *Al- Kubaisy et al.* ⁽¹³⁾ study reports as regard to acne location, face is the commonest site for acne. The study also found a significant

difference in acne location ($P < 0.001$) in which both males (96.7%) and females (92%) demonstrated the highest rate of acne on the face followed by the shoulders. Similarly *Bagatin et al.* ⁽¹¹⁾ study reported that the most commonly affected site was the face (97.5%) or the face and trunk (2.3%).

Our results reported that 34 % of our patients had positive family history of acne. This was in agreement with *Bagatin et al.* ⁽¹¹⁾ who found about 50% of his patients reported family history for acne in mother or father but it was of no statistical significant association regarding family history in fathers ($p = 0.052$), mothers ($p = 0.085$). Another study by *Zouboulis & Bettoli* ⁽¹⁷⁾ concluded that the severity of acne in adolescence is associated with a positive history of severe acne in first-degree relatives, especially the mother. The risk of inflammatory acne, although slight, was greater among adolescents whose siblings had the disease, which is consistent with the literature and probably related to genetic factors. The same pattern was observed in relation to lower educational levels of parents, which may mean less concern with the disease or greater difficulty in accessing early treatment. Another epidemiological study conducted in French schools by *Dréno* ⁽¹⁸⁾ comprising 913 adolescents aged 11-18, reported that 16% of those with acne had positive family histories for acne in their fathers, versus 8% of those without acne.

Our study results showed that 19 of the females in the study (28.4%) had history of menstrual irregularity. this was in agreement with *Al- Kubaisy et al.* ⁽¹³⁾ results that reported about 80 % of the females in the study stated that hyperandrogenicity during the menstrual cycle may have contributed to the acne formation.

In the present study 33 % of patients had positive acne scar, (55.9 %) of patients with family history of acne have the liability of having post acne scar rather than those with negative family history of acne, on the other hand about (21.2 %) of patient with negative family history of acne have the liability of having post acne scar ($P = 0.001$). This comes in accordance with *Williams et al.* ⁽¹⁰⁾ who found that facial scarring due to acne affects up to 20% of teenagers in his study.

The present study revealed that the results of isolated organisms from normal skin of 100 acne

patients were: *S.epidermidis* in 44 cases (44%), *P.acne* in 25 cases (25%) and 31 cases (31%) were found to be with sterile culture.

The current study reported that the results of isolated organisms from pustular lesion of 100 acne patient in aerobic culture were: *S.aureus* in 14 cases (14%), *S.epidermidis* in 39 cases (39%) and micrococcus in 4 cases (4%) and 43 cases (43%) were found to be with sterile culture.

In anaerobic bacterial culture of pustular skin lesion were: *S.aureus* in 10 cases (10%), *S.epidermidis* in 23 cases (23%) and *P.acne* in 56 cases (56%), 11 cases (11%) were found to be with sterile culture. Staphylococci recovered may be derived from contaminating skin flora. This was similar to *Well* ⁽⁹⁾ review who concluded that *Propionibacterium acnes* (*P. acnes*) is the predominant bacteria associated with acne. It is considered part of the normal skin flora and is an inhabitant of the pilosebaceous follicle. However, the role of *P. acnes* in acne vulgaris is significant since the bacteria greatly contributes to the inflammation and irritation associated with acne.

In the present study there was a highly significant statistical difference between the type of organism in acne lesion and normal skin at aerobic & anaerobic state ($P = 0.001$ & 0.001) respectively. In aerobic status *S.epidermidis* was the most frequent bacteria isolated from the pustular skin lesions in our patients (39 %), *S.aureus* accounts for (14%) and (43%) had no growth, while *P. acne* was predominant in the anaerobic state presenting (56%) of our patients, 23% for *S.epidermidis* & (11%) had no growth.

This comes in agreement with *Silverberg and Silverberg* ⁽¹⁹⁾ who hypothesized that *Propionibacterium acnes* may play a role in the pathogenesis of other disorders, particularly those involving common areas where *P. acnes* is found such as the head and neck and gastrointestinal system.

Our results revealed that there was significant statistical difference between the females in the study who are taking POP and those who didn't use it ($P = 0.049$) about 19 female patients (67.9 %) with history of POP taking and 63 patient (87.5%) did not use POP have moderate acne lesions, while 8 of them with history of POP taking (28.6%) and also 8 patient (11.1%) did not use POP have mild acne and only one patient

which taking POP (3.6%) And one patient did not take POP (1.4%) have severe acne. This was relatively high percentage in comparison with the study results reported by *Bagatin et al.* ⁽¹¹⁾ who reported the use of oral contraceptives was reported by only four adolescents (0.9%).

Oral contraception, used properly, can be an effective method in treating acne vulgaris among female patients in their late adolescence ⁽²⁰⁾.

The progestin norgestimate has been presented as a reasonable alternative to drospirenone in treating acne vulgaris for those with a higher risk of blood clots, although it is slightly less effective in the management of acne ⁽²⁰⁾.

Limitations of this study include the sample size and characteristics of the selected population.

CONCLUSION

The high percentage of no growth result in this study directs us that the antibiotic treatments are not the most useful treatment for acne and there are other effective methods for treatment whether topical as retinoids or ablative as Lasers. Patient with family history of acne lesion especially first relative (mother) we must keep in mind that they are more susceptible for post acne scar so we should start with the perfect treatment as early as possible to prevent the post acne sequels.

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