

Study about the Relationship of Some Aerobic Anaerobic Bacteria of Acne and its Resistance to Some of Plant Extracts

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Abstract: *Acne vulgaris* is a common chronic skin disease involving blockage and/ or inflammation of pilosebaceous unit (hair follicle) and their accompanying sebaceous glands. Acne develops as results of an interplay of the following four factors one of them the presence and activity of commensal bacteria *Propionibacterium acnes*. **Methods:** (438) samples were collected from (312) infected patients with acnes from both sexes (132 males) and (180 female) for the period from January 2015 to February 2016 where the comedone samples were (116) and pustules samples (322) samples. Samples were distributed in different agricultural medium where identified their depending upon appearance characteristics and biochemistry tests. (12) plant extracts were used, testing microbe sensitivity of plant extracts and all the results were analyzed by using variance analysis (ANOVA). Moral valued at level ($P < 0.05$). **Results:** Clinical results showed that repetition of infections at females was more than the male. Domination of (*P. acnes*) bacteria in comedone while domination of (*S. epidermidis*) bacteria in pustules. it was noticed that the extract of *Quercus Robur L.* was the most effective in killing all micro-organism under consideration comparing with extracts of used plants. **Conclusion:** The study revealed that *Quercus Robur L.* has fatal effectiveness at rate of (100%) to all positive and negative isolates to gram stain comparing with other plant extracts. We recommend using new medical herbs not used before for the treatment of acne which must have fatal effect upon bacteria (Antibacterial Activity).

Keywords: Acne vulgaris, Plant extracts, clinical findings, Bacteriological findings

1. Introduction

Acne vulgaris is a common chronic skin disease involving blockage and /or inflammation of pilose because unit (hair follicle and their accompanying sebaceous gland). Acne can present as Non-inflammatory inflammatory lesions or a mixture of both. Affecting mostly the face, but also the back and chest (1). Acne develops as result of an interplay of the following four factors:

- 1) Follicular epidermal hyper-proliferation with subsequent plugging of the follicle.
- 2) Excess sebum production.
- 3) The presence and activity of the commensal bacteria propion bacterium acnes and,
- 4) Inflammation (3).

Management

Treatment of acne vulgaris should be directed towards the known pathogenic factors including follicular, hyper-proliferation, Excess sebum production acnes and inflammation. The most appropriate treatment is based on grade and severity of the acne. The following medications are used in treatment of propion bacterium acnes vulgar :-

- 1) Retinoid – like agents (isotretinoin).
- 2) Antibiotic (doxycycline).
- 3) Acne products (azelaic acid).
- 4) Herbal therapy : There are several herbal and folk remedies available for curing acne or pustule like (*Quercus alba*) (tannin containing plants including white oak bark (2), Thyme oil acne). Acne patients have become extremely interested in thyme oil as a tropical treatment (4).

Methods

(438) samples were collected from (312) patients suffer from acne from both sex (132 male, 180 female) for a period from

January 2015 till February 2016 with different ages around (15-35 years) in which samples preparation were from (116) sample taken by sterilized comedone extractor after rubbing the face skin three times with spirit (70%). While the number of the pustule s was (322) samples were taken by pustule s tingling by sterilized disposable lancet after rubbing the face skin twice or three times with spirit (70%) with cotton swab. Both samples were put in small glass bottles with screw heads (capped bottles) called (pigot tube) filled with Thioglycolate, then transfer the samples by cooled containers to the laboratory. **Sample Implant.** Comedones samples were implanted after breaking them by sterilized glass balls (glass beets) were put inside glass bottle by vortex mixture upon blood agar and MacConkey agar (separation medium) by using planning through sterilized standard implanting transport (loopful). While the pustule s samples were implanted directly from transfer medium upon enriched medium of blood and MacConkey agar. This process carried out in case of aerobic implant. In case of anaerobic, both samples were implanted (comedone and pustule s) on the (Thioglycolate) were put already in well capped glass bottles (screw capped bottles) after adding (1%) (Tween 80) in order to motivate the bacteria (*P. acnes*). The bottles will be hugged for the period of (5-7 days). Then transfer (loopful) from implanted to blood agar which was put in anaerobic refreshment (anaerobic jar) for (2-3) days for separation of *P. acnes*. Also conduct anaerobic implant upon Brewer and anaerobic hugging in an anaerobic jars kit) for a period of (5-7) days.

Identification of the Isolate

Identification of isolate depend upon appearance characteristics of the implanted item against implanting medium including (size, color, edges and heights of the colonies and conducting bio-chemical tests according to (Bergey's Manual of determinative) (Bacteriology -1994).

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Preparation of Plant Extracts. (12) plant extract were used in the experiment in which preparation of water and alcohol extracts were made according to type of the plants, and type of the separated part as in the table. We tested the sensitivity of microbes towards plant extracts by using Muller Hinton agar and process (well methods) in which we prepared germ suspension and vaccinate it with hard medium. Then preparing a well of (5 mm) and putting (100) micro-liter from the extract in each well at (37 °) for (18-24) hours, then measuring places of inhibition of all isolates. All clinical results subjected to statistical analysis by using analysis of variance (Anova). The moral was valued at the level ($R < 0.05$).

2. Results

Clinical results of the current study revealed which included (312) patients infected with acne, their ages ranging between (19-39) years in which the infections of the males was (132) at the percentage of (42.30%), while at the female was (180) infections at the percentage of (57.69%), (table-1).

Table 1: The relation of acne occurring with age group for both sexes

Age groups	Male%	Female%	Total number
12-17	52 (39.39)	82 (45.55)	134 (45.55)
18-23	64 (48.48)	75 (41.66)	139 (44.55)
24-29	12 (9.09)	19 (10.55)	31 (9.93)
30-35	4 (3.03)	4 (2.22)	8 (2.56)
Total	132 (42.30)	180 (57.69)	312

Microbiology Results

(438) samples were implanted infected by acne including (116) comedones and (132) pustules. After separation and identification of all germ isolates according to the gender and type depending upon appearance characteristics for both types, and biochemical tests, the results showed the lesion of both types illustrated in figures (2,3) which refers to identified isolates germ from (116) samples (comedone) and (322) pustule.

Table 2: Types of isolates bacteria from (116) comedone

Isolates bacteria	Percentage
<i>P. acnes</i>	68.96
<i>S. epidermidis</i>	65.51
<i>S. aureus</i>	8.62

Table 3: Types of isolates bacteria from (322) pustule

Isolates bacteria	Percentage
<i>S. epidermidis</i>	71/32
<i>P. acnes</i>	37.26
<i>P. aeruginosa</i>	11.49
<i>S. aureus</i>	9.93
<i>E. coli</i>	7.14

It was noticed that the domination of isolation of bacterial microscope organism (*P. acnes*) at percentage of (68.96%) followed by bacterial type of (*S. epidermidis*) at isolate percentage of (65.51%), then the third grade comes negative bacteria of gram stain. (table. 4, 5).

Test of microbe sensitivity towards plant extracts by using (Muller-Hinton Agar)

Test carried out to check the sensitivity of all bacterial isolates for (12) plant extracts and the results illustrated in the table with concentration of (10%) for all extracts in which the results showed that *QuercusRobur* has fatal effect (100%) to all bacterial isolates under study.

Table 4: Sensitivity types of plant extracts of (306) *S. epidermidis* isolate

S	Name of extracted plant	%
1	<i>QuercusRobur L.</i>	100
2	<i>Thmus vulgaris</i>	93.13
3	<i>PlantagoLaceolata</i>	91.71
4	<i>Lawsoniainermis</i>	87.25
5	<i>Fumariaoffixinalis</i>	86.92
6	<i>apiumgraveolens</i>	83
7	<i>Linumusitatissimum</i>	81.04
8	<i>Eugenia aryophyllus</i>	78.10
9	<i>Zingiberofficinale</i>	0
10	<i>Pimpinellaanisum</i>	0
11	<i>Lepidiumsativum L</i>	0
12	<i>Trigonella – foenumgraecum</i>	0

Table 5: Sensitivity types of plant extracts of (180) *P. acne* isolate

S	Name of extracted plant	%
1	<i>QuercusRobur L.</i>	100
2	<i>Thmus vulgaris</i>	95.50
3	<i>Eugenia aryophyllus</i>	93.33
4	<i>PlantagoLaceolata</i>	93.20
5	<i>Lawsoniainermis</i>	90
6	<i>Fumariaoffixinalis</i>	90
7	<i>Apiumgraveolens</i>	83.33
8	<i>Linumusitatissimum</i>	73.33
9	<i>Zingiberofficinale</i>	0
10	<i>Pimpinellaanisum</i>	0
11	<i>Lepidiumsativum L</i>	0
12	<i>Trigonella – foenumgraecum</i>	0

Table 6: Sensitivity types of plant extracts of (23) *E. coli* isolate

S	Name of extracted plant	%
1	<i>QuercusRobur L.</i>	100
2	<i>PlantagoLaceolata</i>	95.56
3	<i>Thmus vulgaris</i>	91.30
4	<i>Lawsoniainermis</i>	91.30
5	<i>Fumariaoffixinalis</i>	86.95
6	<i>apiumgraveolens</i>	82.60
7	<i>Linumusitatissimum</i>	78.26
8	<i>Eugenia aryophyllus</i>	73.91
9	<i>Zingiberofficinale</i>	0
10	<i>Pimpinellaanisum</i>	0
11	<i>Lepidiumsativum L</i>	0
12	<i>Trigonella – foenumgraecum</i>	0

It showed also variation of plant extracts with concentration of (10%) *Thmus vulgaris*, *PlantagoLaceolata*, *Lawsoniainermis*, *apiumgraveolens*, *Fumariaoffixinalis*, *Eugenia aryophyllus*, *Linumusitatissimum* in their effect upon all bacterial isolates under consideration, with the absence the effects of (*Zingiberofficinale*, *Pimpinellaanisum*, *Lepidiumsativum L.*, *Trigonella – foenumgraecum*) upon bacterial isolates which did not show any antibacterial efficacy. (table -4, 5 and 6).

3. Discussion

Clinical Results

It was noticed that clinical results under study the repletion of infection with the female more than male especially in the age between (12-17) due to sexual maturity stage and appearance of adolescence much earlier than male. Appearance of acne is the beginning of sexual maturity stage for both sexes (6).

These results agreed with international studies (7&8), and local study (9, 10), but differ with local studies (11). While conducting statistical analysis, we found there were moral differences at ($P < 0.003$) between male and female regarding the relationship of the disease with both sexes and their age categories for each.

Microbiology Results

Domination of the type (*P. acnes*) refers to oxygen pressures in comedone which helps to create suitable anaerobic environments of this type (12). Domination of the type (*S. epidermidis*) in pustules refers to high oxygen pressures and change of Hydrogen basis of the medium which lead to create suitable environment of above-mentioned type and non-suitability of other type (*P. acnes*).

Also the swept of white blood cells (Neutrophil) in infected area will lead to lessen of (*P. acnes*) which excreted enzyme of chemical attractive characterized by its low molecular of neural cells which lead to preferring the neural cells (*P. acnes*) more than other types (13).

Separation of negative bacteria of gram color from both (*P. aeruginosa*, *E. coli*) at total percentage of (17.07%) agreed with a study in this regard (14) whereas the existence of these organism very few in natural conditions, but possibility of its existence refers to world –wide using of antibiotic and long period of medication with lead to allowance of these organism to make colony and grow, and reproduction in pustules, and this case called (G-ve folliculitis)- inflammation of the hair follicle by negative microbiology with gram stain form which inflame the pustules and creates inflammatory papules more than comedone. Testing microbes sensitivity of plant extracts by using (Moller-Hinton Agar)

The results showed that *Quercus Robur L.* has fatal effect (100%) to all bacterial isolates under consideration. The results were agreed with results of international studies (15), it may be due to the efficiency of this plant extract as containing (Tannins) which belong to phenol category as anti-bacteria factors. accordingly this extracts has great efficiency more than other extracts

The variance in the effect of plant extracts under study could be interpreted that these extracts contain natural compounds existed in plant extracts, in addition to variance of bacteria types under consideration especially regarding cellular formation, genetic factors and the nature of the target which will be affected by plant extracts.

Also the effective materials existed in the plant and counter-efficiency will affected by many factors such as soil environments, climate, way of collection, means of keeping plants parts, storage conditions of used plants parts in extraction, besides the size of bacterial vaccination which is used in the experiment. All these factors give effects upon anti-effectiveness of plant extract of microbiology under test (16).

4. Conclusions

We have noticed in the conclusions of the study that infections with the females were more than male regarding clinical study. Also we noticed that (*P. acnes*) bacteria was dominated in the samples of acnes (comedones), while (*S. epidermidis*) bacteria has dominated in (pustules samples) regarding bacteriology study. Regarding the study of the effect of medical herb noticed the extract of oakcauliflower is the most effective extract and effects all kinds of aerobic and anaerobic bacteria.

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