

Evaluation of Hematological Parameters And DHEA-S Hormone Association with Acne In The Province Of Thi-Qar

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Abstract

This study was conducted in the labs of the College of Education for Pure Sciences, Al-Hussein Educational Hospital and Al-Nahrain Specialized Laboratory of the Health Department of Thi-Qar province, during the period from October 2018 to March 2019. The study included of 100 blood sample of patients with acne (39 males) and (61 females) and their age between 10-35 years. The aim of the study was to assess the level of DHEA-S hormone in acne vulgaris patients in the serum using a technique enzyme- linked immunosorbent Assay (ELISA).The study included the test of the susceptibility of phagocytic cells on phagocytosis (coefficient of phagocytosis) and measurement of hematological parameters (Red blood cell count, hemoglobin ratio, total and differential white blood cells count).

The results showed the presence elevate in the level of DHEA-S for 21 sample out of 70 patients with acne. Results showed that there was no association between disease incidence and DHEA-S when compared with the healthy control in the population of Thi- Qar province. Increase coefficient phagocytosis was significant ($P \leq 0.05$) in all patients with acne compared with a group of control. As for the blood parameters, the results of the present study showed no significant differences ($P \leq 0.05$) in the rates of red blood cells and hemoglobin in all acne patients compared to the control group. It also showed increased rate of counting the total of WBCs and differential cell neutrophils and lymphocytes ($P \leq 0.004$), ($P \leq 0.004$), ($P \leq 0.025$), respectively in acne patients compared to healthy controls group.

Keywords: *Acne vulgaris, Hematological parameters, DHEA-S.*

Introduction

Acne vulgaris is the most common a chronic inflammatory disease of the pilosebaceous unit and characterized by non- inflammatory lesion (comedones with black and white heads), inflammatory lesions like (papules, pustules and nodules) ^(1,2). Resulting from increased sebum production, inflammatory process, androgen excess states, abnormal follicular epithelial differentiation, insulin resistance, obesity and the proliferation of *Propionibacterium acnes* ³.

Acne can be classified according to its severity into: mild, moderate and severe using the Global acne assessment scale ⁴. It occurs primarily in the oily (seborrheic) areas of the skin that involved face, neck, back and chest ^(5,6).

Androgen enhances sebum production and follicular keratinization plays important role in pathogenesis

of acne. Therefore, testosterone, androstenedione, dehydroepiandrosterone sulphate are involved in the evolution of acne ⁷.

The circulating androgen dehydroepiandrosterone sulphate (DHEA-S) is mostly produced in the adrenal glands. DHEA-S is the highest androgen concentration in the serum in both sexes and is considered the most important regulator of sebum secretion ^(8,9). Elevated DHEA-S levels in male and female with acne have been repeatedly demonstrated in many studies ⁶. Acne is a common feature in the path of endocrine diseases distinguished by elevated levels of androgens ¹⁰.

The aim of the current study was to assessment of some blood parameters (WBCs, RBCs, HB), phagocytosis and knowledge of their relationship with acne and study of dehydroepiandrosterone sulphate (DHEA-S) and its relationship with acne.

Materials and Method

Subjects :- The study groups have the following included:

This study was conducted in the labs of the College of Education for Pure Sciences, Al Hussein Educational Hospital and Al-Nahrain Specialized Laboratory of the Health Department of Thi-Qar province, during the period from October 2018 to March. The patients in this study were divided into three groups According to age :

1-First group : comprised 10 acne patients less than 20 years old.

2-Second group : comprised 10 acne patients whose ages between 21-30 years old.

3-Third group : comprised 6 acne patients whose were 31 years old and above.

In addition, the study included 30 people as a control group, with no history or clinical evidence of common acne or any other chronic disease, and have no obvious abnormalities, divided into three groups according to their age:

1-First group : comprised 10 acne patients less than 20 years old.

2-Second group : comprised 10 acne patients whose ages between 21-30 years old.

3-Third group : comprised 6 acne patients whose were 31 years old and above.

Blood sample collection: -

Blood samples were collected by venipuncture from 100 patients (39 males, 61 females) and 30 controls (five milliliters of venous blood) were drawn by disposable syringe under aseptic technique. each blood sample was divided into two parts:

a- Two milliliters were placed in a sterile tube containing EDTA for WBC deferential count and phagocytosis processes (during 2 hour).

b- Three milliliters were put directly in a sterile Gel tube and allowed to clot , then serum was separated by centrifugation at 4000 rpm for 15 minutes. The serum was stored at -20 C° freezing. These sera 70 acne patients (30 male, 40 female) and 20 controls (10 male,10 female) were used for estimating the concentration of

DHEA-S hormone.

Phagocytosis Procedure :

The procedure was carried out according ¹¹ as follow: 0.025 ml of the collected blood was put in plane tube , then added for it 0.05 ml from Killed yeast suspension which was prepared by soluble 10 grams of *Saccharomyces cerversiae* yeast made in Turkish pakamaya company in 150 milliliters of normal saline and put suspension in water bathe for 60 minutes , then this suspension was filtered after cooling.

0.025 ml of HBSS were added to the mixture and incubated at 37 C° for 30 minutes . One drop of the mixture was placed on a slid and smeared, then left to dry , fixed by methyl alcohol (99%) for min and stained for 20 min with Wright stain. then, examined under oil immersion.

No. of phagocytic cells

Phagocytosis index = $\frac{\text{No. of phagocytic cells}}{\text{Total number of cells}} \times 100\%$

Total number of cells

Hematological assay:

The hematological tests including (total and differential WBCs counts ,RBC and Hb) were done by using Genux Auto Hematology Analyzer in which the results read and printed automatically.

Serological assay: -

A number 1 kit was used for DHEA-S hormone, where DHEA-S hormone concentration were measured for control group (non-infected) and infected group by using the Enzyme-linked Immunosorbent Assay (ELISA) method and using the ELISA device and a measurement kit designed for this device for hormone.

Statistical Analysis

The analyses of data were expressed as mean \pm SD. The comparisons between each Acne patients group with age matched healthy control were performed with T-test using computerized Minitab program. The comparisons among the three age group of Acne patients were performed with analysis of variance (Chi – square, Odds Ratio) by using computerized Minitab 14 program. P< 0.01 was considered to be the least limit of significance . All the statistical analysis were done by

using Pentium-4 computer through the (SPSS program) Statistical Package For Social Sciences (version -23).

Results

Hematological parameters

RBCs , Hb count :

This doesn't show the results of the current study in Table (1), no significant differences ($p \leq 0.05$) in blood picture of patients and healthy control group. The red blood cells (RBCs) count, hemoglobin concentration (Hb) in Acne compared with the healthy control group, where the RBCs mean are $(4.89 \pm 0.58) \times 10^6$ cell/ml for

patients and $(4.98 \pm 0.55) \times 10^6$ cell/ml for healthy control group respectively, while the hemoglobin concentration are (13.40 ± 2.54) g/dl for patients and (13.52 ± 1.64) g/dl for healthy control group respectively, as the table (1).

Phagocytic index :

The results of the current study show significant differences ($p \leq 0.05$) in the rate of phagocytosis, as it increased the rate of phagocytosis in Acne patients to (45.89 ± 1.07) compared with the healthy control groups, and that the rate of phagocytosis (29.58 ± 1.34) , as in the Table (1) and figure (1).

Table (1): some hematological parameters of Acne patients and healthy control

Parameter	Patient (N=100), Mean \pm SD	Healthy (N=30), Mean \pm SD	P.Value
RBC	4.89 ± 0.58	4.98 ± 0.55	0.423
Hb(g/dl)	13.40 ± 2.54	13.52 ± 1.64	0.808
Phagocytosis	45.89 ± 1.07	29.58 ± 1.34	0.001*

Degree freedom (df) = 128 *P.value \leq 0.05

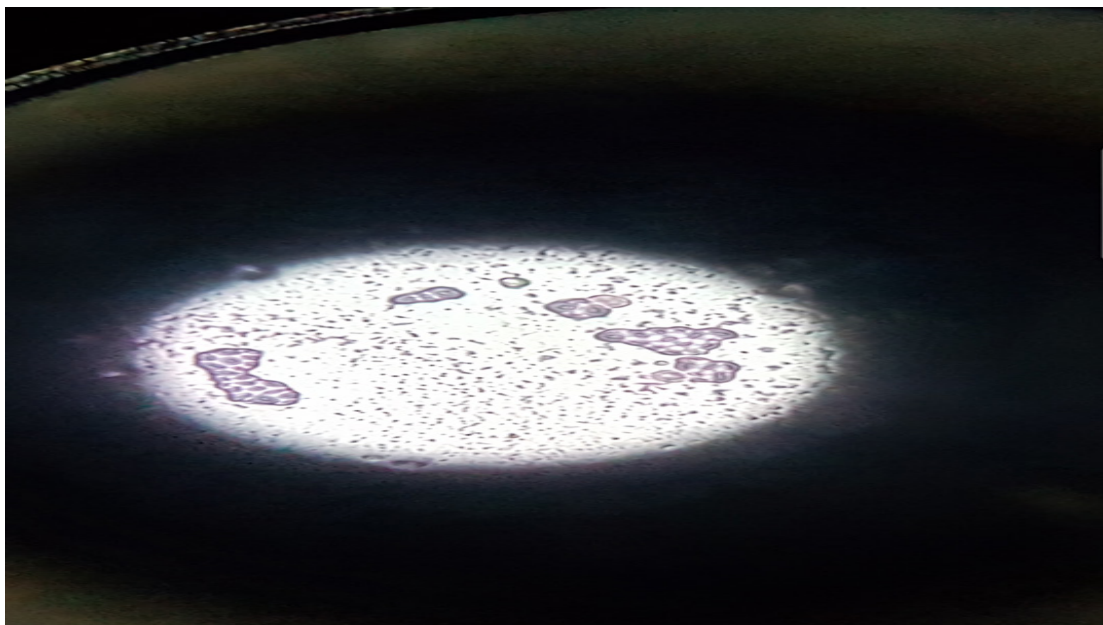


Figure (1): Phagocytosis of yeast cell by Phagocytic cell 1000x

WBCs, Neutrophil, Lymphocytes, Monocytes, Eosinophil and Basophil count :

The results of the current study show significant differences between the two groups of patients compared with healthy controls that has been shown in Table (2). The WBCs count, Neutrophil, Lymphocytes, Monocytes, Eosinophil and Basophil in Acne vulgaris compared with the healthy control group, WBCs count that means are (9.98 ± 3.30) for patients and (8.14 ± 1.73) for healthy controls group respectively, neutrophils means are (6.04

± 2.57) for patients and (4.61 ± 1.32) for healthy control group respectively, lymphocytes means are (2.95 ± 1.08) for patients and (2.60 ± 0.60) for healthy control group respectively, while monocytes means are (3.80 ± 1.80) for patients and (3.85 ± 1.05) for healthy control group respectively, Eosinophil means are (2.05 ± 1.04) for patients and (4.65 ± 1.57) for healthy control group respectively, eventually the basophils there would have no significant difference, too and the means are (0.94 ± 0.80) for patients, and (0.78 ± 0.74) for healthy control group, as in Table (2).

Table (2): Hematological parameters in Acne patients and healthy control group

Parameter	Patient (N=100), Mean ± SD	Healthy(N=30), Mean ± SD	P.Value
WBC	9.98 ± 3.30	8.14 ± 1.73	0.004*
NEU	6.04 ± 2.57	4.61 ± 1.32	0.004*
LYM	2.95 ± 1.08	2.60 ± 0.60	0.025*
MONO	3.80 ± 1.80	3.85 ± 1.05	0.931
ESO	2.05 ± 1.04	4.65 ± 1.57	0.104
BASO	0.94 ± 0.80	0.78 ± 0.74	0.319

Degree freedom (df) = 128 *P.value ≤ 0.05

Serological parameters

Serum DHEA-S hormone concentration:

The results of the current study show no significant difference (p>0.05) concentration of DHEA-S hormone

in a group of patients compared with a group of control as concentration as follow: DHEA-S concentration (3.71 ± 1.51) for male patients compared with a group of control (1.96 ± 0.48), while DHEA-S concentration (3.31 ± 1.16) for female patients compared with a group of control (3.28 ± 1.21) with a significant difference (0.05) as seen in Table (3).

Table(3) : Comparison of serum DHEA-S hormone concentration (µg/ml) of the patient groups with healthy control group

Parameter	Subject	No. of cases	Mean ± SD	P.Value
DHEA-S	Male patient	30	3.71 ± 1.51	0.88
	Male control	10	1.96 ± 0.48	
	Female patient	40	3.31 ± 1.16	
	Female control	10	3.28 ± 1.21	

χ²= 0.66 degree freedom (df)=3 P.value > 0.05

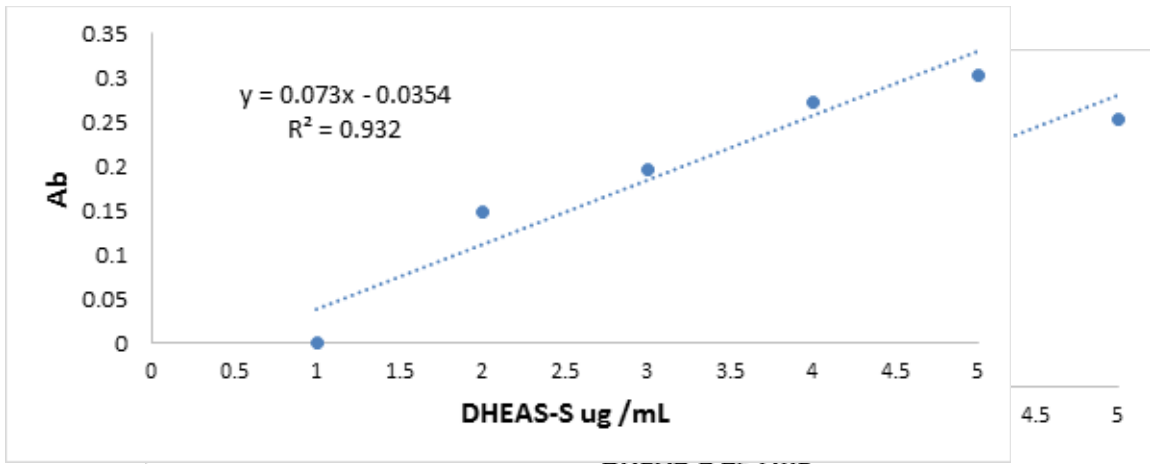


Figure (2): Standard curve of DHEA-S

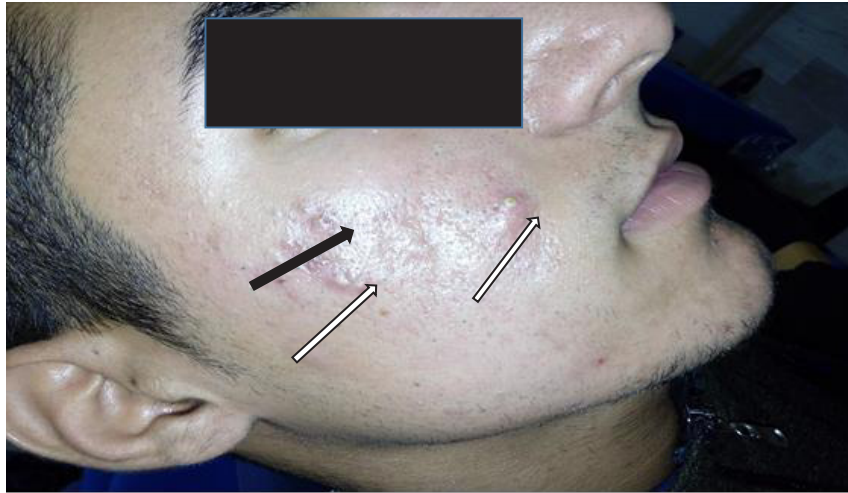


Figure (3): Acne lesions, including comedones (white arrows) and pustule (black arrow) on the facial skin.

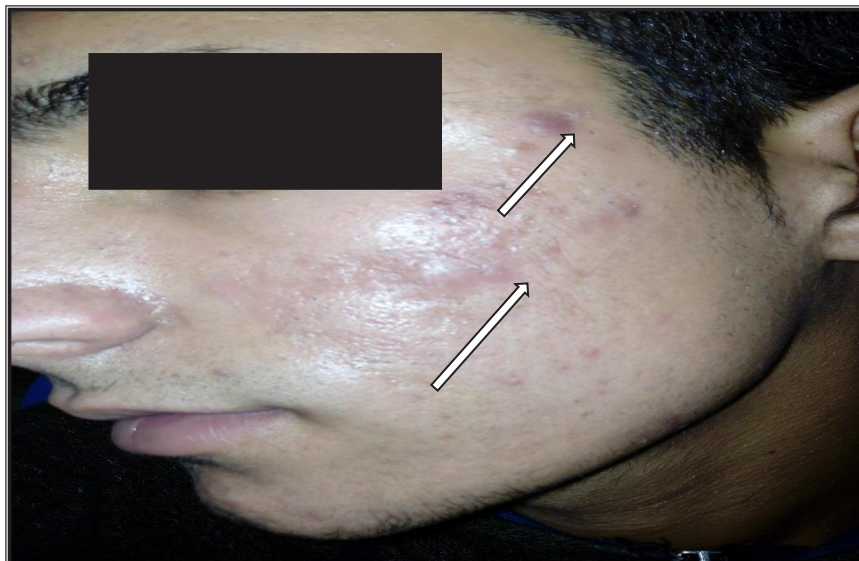


Figure (4): Acne lesions, including papule on the facial skin.



Figure (5) : Acne lesions, including comedones (black heads) on the facial skin.

Discussion

Acne vulgaris belong to a complex and multifactorial disease of pilosebaceous unit. Acne pathogenesis cannot be viewed as impartial since its etiologies influence each other. Pathogenesis of acne vulgaris recently emphasizes inflammatory process. The latest finding indicates the role of *P. acnes* as a triggering factor of inflammatory responses which exerts an influence on the severity of acne vulgaris. Pathways underlying the formation of papules, pustules, and nodulocystic acne which involve cytokines were detected¹². The results of the current study showed a lack of significant difference ($p > 0.05$) in each of the concentrations of RBC between the two groups of patients (4.89 ± 0.58) and healthy control group (4.98 ± 0.55), and the percentage of hemoglobin in patients (13.40 ± 2.54) and healthy control (13.52 ± 1.64) and attribute the lack of significant differences to the inflammation of acne doesn't affect the effectiveness of red blood cells and its ability, as well as RBC membrane not effective and nor old and this is reflected in the level of HB, there has been no change in rate.

Phagocytosis is considered the first line of defense against injury and carried out by Neutrophil and Macrophage and Dendritic cells¹³, which should migrate to the places of injuries or infections in order to do its job and be the migration process in response to

some of the attraction of chemical¹¹ and phagocytosis one of Nonspecific immune mechanism and motivated by the entry of foreign bodies¹⁴

The current results showed significant difference in percentage of phagocytosis ($P < 0.05$) patients with Acne (45.89 ± 1.07) and healthy control (29.58 ± 1.34). The reason of this result due to increasing the number of Neutrophil and Macrophage during the inflammation, and due to high levels of cytokines such IL- 1α and IL-10 which belong to cellular compound inflammation that leads to the increase of phagocytosis especially in patients with acne vulgaris. This study agrees with¹⁵.

On the other hand there was a significant variation ($p < 0.05$) in the WBCs count between Acne patients and the control group, the increase in WBCs may be due to the response of the immune system to face the inflammatory effects in the tissues and increase is the rise in the number of neutrophils in this study are agree with¹⁵.

The results of our study showed that the proportion of hormone in males with acne (3.71 ± 1.51) compared with healthy males (1.96 ± 0.48) is higher than women where the proportion of women infected (3.31 ± 1.16) compared to healthy women (3.28 ± 1.21) where the rise in males significantly while in females is not

significant. The reason is probably due to the amount of DHEA-S hormone produced in the body is related to age and sex, in addition to its concentration in the blood may be undergo changes during the day. As well as, the physiological role of androgens in women during adulthood is unclear. Women who suffer from hyperandrogenic disorders may develop acne, hirsutism, deepening of the voice, androgenic alopecia. This study agree with (16,17,18,19).

Financial Disclosure: There is no financial disclosure.

Conflict of Interest: None to declare.

Ethical Clearance: All experimental protocols were approved under University of Thi-Qar- College of Education for Pure Sciences, Biology Department and all experiments were carried out in accordance with approved guidelines.

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