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Serum Levels of Fetuin-A in Patients with Acne Vulgaris

Alaa Hassan Maraee [1], Sara Kamal Rizk [2], Ayman Mohamed Abdelwahab *[1]

[1] Dermatology, Andrology and STIs Department, [2] Medical Biochemistry and Molecular Biology, Faculty of Medicine, Menoufia University, Shiben Elkom , Egypt

Corresponding author Ayman Mohamed Abdelwahab,

Email: ayman.abdelwahab200100@gmail.com, **Phone:** +20 10 27171767

ABSTRACT

Background: Acne vulgaris (AV) is a prevalent cutaneous condition that is distinguished by the malfunction of the pilosebaceous unit. It may persist in certain individuals even after adulthood, despite the fact that it is most frequently observed in adolescents. The objective of this study was to evaluate the serum Fetuin-A levels of AV patients and evaluate their correlation with the clinical severity of the disease.

Methods: A case-control study was conducted on 105 participants at the Outpatient Clinic of Dermatology and Andrology at Menoufia University. They were divided according to the global acne classification system, group I (Patients group) (n=70) was divided into three subgroups based on the various presentations of AV: group (A) (mild AV, n=20), group (B) (moderate AV, n=20)), and group C (severe AV, n=20), group D (very severe, n=10) and group II (Controls) involved 35 healthy subjects as control group.

Results: At a cut-off value of ≥14 ng/ml, serum fetuin A demonstrated a statistically significant 65.71% sensitivity and 62.86 % specificity for discriminating between patients and controls (p= 0.001).

Conclusions: Patients with acne Vulgaris exhibit statistically significant higher serum fetuin-A levels compared to controls. Furthermore, serum Fetuin-A levels were found to be significantly correlated with the course of disease, stress, and diet. Additional research is needed to elucidate the causal nature of these relationships.

Keywords: Serum Levels, Fetuin-A, Acne Vulgaris, Severity.

INTRODUCTION

Acne vulgaris (AV) is a well-known cutaneous condition that is characterized by the dysfunction of the pilosebaceous unit. An estimated 80% of Americans will experience AV at some stage in their lives. Although adolescents are likely to experience it the most, it may persist in certain individuals into adulthood [(1)]. There have been four components to the traditional paradigm of acne, which are frequently presented in a sequential order. The follicle is plugged as a result of follicular hyperkeratinization and increased sebaceous production that arises from an excess of androgen. This process enables the Propionibacterium acnes bacterium to expand within the follicle, resulting in a clinically evident disease and an inflammatory cascade [(2)].

Serum Fetuin-A, a multifunctional glycoprotein, primarily derived from the liver and is predominantly responsible for the promotion of insulin resistance [(3)]. A robust inflammatory response and the accumulation of late mediators (e.g., High-Mobility-Group-Protein B1) that stimulate hepatic fetuin-A expression are facilitated by the downregulation of fetuin-A by early proinflammatory mediators (IL-1, IL-6, TNF-α, Interferon-gamma) [(4)]. Fetuin-A functions as an anti-inflammatory mediator, which contributes to the macrophages deactivation [(5)].

Alpha-2-Heremans Schmid glycoprotein (AHSG) is a protein that is measured at 62 kilodaltons and is present in the embryonic tissue. Bone resorption is the function of the cysteine proteinase inhibitor class, to which it belongs. It is a negative acute phase reactant that is produced by the hepatic cells. It serves as an inhibitor of calcium and phosphate precipitation by increasing their solubility in the circulation and inhibiting the formation of calcium crystals [(6)].

In acne, cutaneous inflammation is induced by Corynebacterium acnes via increasing pro-inflammatory chemical signals production, such as I-8, tumor necrosis factor alpha (TNF-α), interleukin-1 alpha (IL-1α), and Leukotriene B4. IL-1α is a critical factor in the formation of comedones [(7)].

Few studies evaluated the role of fetuin A in AV and its relation to the clinical severity and the results were debating and the function of fetuin A remain poorly understood.

This investigation was designed to assess the serum Fetuin-A levels of patients with AV and to determine their correlation with the clinical severity of the condition.

PATIENTS AND METHODS

This case-control study was conducted on 70 patients and 35 apparently healthy controls, at the Outpatient Clinic of Dermatology and Andrology at Menoufia University.

Inclusion criteria

AV patients of both sexes with varying degrees of AV severity who had not received any systemic (for 6 weeks) or topical treatment for AV (2 weeks).

Exclusion criteria

Patients with systemic diseases, hepatic or renal or malignancy, a body mass index (BMI) of ≥30, pregnancy, lactation, polycystic ovary or hirsutism, infectious or autoimmune cutaneous or and inflammatory disease.

Grouping:

All subjects were divided into:

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- Group I (Patients group) (n=70), using the global acne grading system (GAGS) and based on the AV diverse manifestations, was categorized into three subgroups [8]: Group A (n=20): had mild AV, group B (n=20): had moderate AV, group C (n=20): had severe AV and group D (n=10): with very severe AV.
- Group II (Controls (n=35): included apparent healthy subjects of matched age and sex.

All cases underwent history-taking including present history (duration of AV, onset, course, and previous treatment), demographic data, past history (systemic or dermatological diseases, family history of AV and past history of drug intake), comprehensive general examinations, with a particular emphasis on their BMI (BMI was determined by dividing their weight (Kg) by their squared height (m), as follows: BMI = weight (kg)/height^2 (m^2) [9], where BMI ≥30 30 kg/m^2: obesity, >25–29.9 kg/m^2: overweight, >18.5–24.9 kg/m^2: normal weight, ≤18.5 kg/m^2: underweight, ≤ 16.5 kg/m^2: severely underweight [10].

Two expert dermatologists conducted a dermatological examination to ensure the diagnose AV, identify the location of AV, and evaluate the severity of AV using GAGS.

The enzyme-linked immunosorbent assay (ELISA) was implemented for evaluation of embryonic serum levels in a laboratory environment. Five millilitres of venous blood were obtained and placed in a plain tube. The tube was allowed to clot for 30 minutes. Separation and centrifugation of the serum were performed at 2000 rpm for 20 minutes. Following this, the serum was refrigerated at -20°C until the analysis commenced. The AHSG concentration was plotted on the y-axis and the absorbance on the x-axis to construct a standard curve. This was achieved by either sketching a best-fit curve through the nodes on the graph or plotting the mean O.D. and concentration of each standard on log-log graph paper. Additionally, the utilization of plotting software, including curve expert 1.30, was advised. The concentration determined from the standard curve must be multiplied by the dilution factor for samples that have been diluted. In order to generate a standard curve, the absorbance was plotted on the x-axis and the AHSG concentration on the y-axis.

This was accomplished by either plotting the mean O.D. and concentration of each standard on log-log graph paper or sketching a best-fit curve through the nodes on the graph. Furthermore, it was recommended to employ plotting software, such as curve expert 1.30In the case of diluted samples, the concentration as determined from the standard curve must be multiplied by the dilution factor.

Sample size:

The sample size calculation was performed using G.power 3.1.9.2 (Universitat Kiel, Germany). The sample size was calculated according to mean serum fetuin A level which was significantly higher in patient group than control group (164.485±91.432 vs. 109.381±40.127) according to a previous study [11]. Based on the following considerations: 0.05 α error and 95% power of the study. Seven cases were added to overcome dropout. Therefore, 105 participants were allocated.

Ethical approval: The patients provided a written informed consent. The investigation was authorized by the Ethics Committee of the Faculty of Medicine at Menoufia University (5/2023 DERM 12). The study complied with the ethical standards of the Helsinki Declaration of the World Medical Association.

Statistical analysis

Statistical analysis was done by SPSS v26 (IBM Inc., Armonk,NY,USA). Quantitative variables were presented as mean and standard deviation (SD) and compared between the two groups utilizing ANOVA (F) test. Qualitative variables were presented as frequency and percentage (%) and were analysed utilizing the Chi-square test. A two tailed P value < 0.05 was considered statistically significant. The overall diagnostic performance of each test was assessed by characteristic curve (ROC-curve) analysis, a curve that extends from the lower left corner to the upper left corner then to the upper right corner is considered a perfect test. The area under the curve (AUC) evaluates the overall test performance (where the area under the curve >50% denotes acceptable performance and area about 100% is the best performance for the test).

RESULTS

Table 1 shows clinical data of the studied acne cases. The age, sex, smoking and BMI were insignificantly different between patients and control groups. The serum fetuin A was significantly increased in patients than controls (18.6 ±9.22 vs 12.02±6.41, P value =0.001).

Table 1: Comparison between the two studied groups according to demographic data, Serum Fetuin-A and clinical data of the studied acne cases (N=70)

Studied variables			Cases (N = 70)	Control (N = 35)	P value
Demographic data	Age (years)		18.69 ± 4.18	19.40 ± 3.05	0.327
	Sex	Male	30 (42.9%)	13 (37.1%)	0.575
		Female	40 (57.1%)	22 (62.9%)	
	Smoking		12 (17.1%)	9 (25.7%)	0.301
	BMI (Kg/m^2)		20.83 ± 1.85	20.83 ± 1.76	1.000
Serum Fetuin-A			18.62 ± 9.22	12.02 ± 6.41	**0.001***
Clinical data	Family history		52 (74.3%)	--	--
	Relation To Diet		54 (77.1%)	--	--
	Relation To Stress		47 (67.1%)	--	--
	Onset	Sudden	26 (37.1%)	--	--
		Gradual	44 (62.9%)	--	--
	Course	Stationary	19 (27.1%)	--	--
		Progression	39 (55.7%)	--	--
		Regression	12 (17.1%)	--	--
	Duration (years)		5.21 ± 3.08	--	--
	Type of acne	Comedones	19 (27.1%)	--	--
		Papules	42 (60.0%)	--	--
		Pustules	47 (67.1%)	--	--
		Nodules	29 (41.4%)	--	--
	Severity	Mild	20 (28.6%)	--	--
		Moderate	20 (28.6%)	--	--
		Severe	20 (28.6%)	--	--
		Very severe	10 (14.3%)	--	--
	GAGS		25.23 ± 12.03	--	--

GAGS: global acne grading system. No: number, p: p value for comparing between the two studied groups, *: Statistically significant at $p \leq 0.05$, Data are presented as mean ± SD or frequency (%). IQR: interquartile range, SD: standard deviation, BMI: body mass index.

The serum Fetuin-A level was insignificantly different among the different grades of acne severity, however very severe cases had higher levels but still with no significant difference. **Table 2**

Table 2: Comparison between the different studied groups according to severity of acne and serum fetuin-A

	Severity of acne lesions				P
	Mild (n = 20)	Moderate (n = 20)	Severe (n = 20)	Very severe (n = 10)	
Serum Fetuin-A	16.63 ± 9.42	18.08 ± 9.49	19.43 ± 9.19	22.05 ± 8.44	0.410

Data are presented as mean ± SD or frequency (%). SD: standard deviation, no: number: p value for comparing between the studied groups.

The course of the disease, diet, and stress all exhibited a statistically significant correlation with the mean level of serum fetuin-A (p=0.014, 0.001, 0.013, respectively). **Table 3**

Table 3: Relationship between Serum Fetuin-A and different parameters in cases group (n = 70)

		N	Serum Fetuin-A	P
Gender	**Male**	30	20.19 ± 9.53	0.146
	Female	40	17.44 ± 8.91	
Onset	**Sudden**	26	18.92 ± 9.21	0.913
	Gradual	44	18.44 ± 9.32	
Course	**Stationary**	19	13.23 ± 8.63	**0.014***
	Progressive	39	20.36 ± 8.53	
	Regressive	12	21.48 ± 9.46	
Family history	**Negative**	18	17.78 ± 9.67	0.600
	Positive	52	18.91 ± 9.13	
Severity	**Mild**	20	16.63 ± 9.42	0.410
	Moderate	20	18.08 ± 9.49	
	Severe	20	19.43 ± 9.19	
	Very Severe	10	22.05 ± 8.44	
Smoking	**Negative**	58	18.06 ± 9.03	0.115
	Positive	12	21.31 ± 10.04	
Relation to diet	**Negative**	16	11.45 ± 8.20	**0.001***
	Positive	54	20.74 ± 8.45	
Relation to stress	**Negative**	23	14.31 ± 9.04	**0.013***
	Positive	47	20.73 ± 8.63	

Data are presented as mean ± SD. p: p value for comparing between different categories, * Significant as P-value ≤ 0.05.

The correlation between serum Fetuin A and age was a positive non-significant (r=0.027 and p=0.825), duration of disease (r=0.191 and p=0.114) and GAGs (r=0.155 and p=0.200), but with BMI, there was negative non- significant correlation (r=-0.067 and p=0.580). **Table 4**

Table 4: Correlation between Serum Fetuin-A and different parameters in cases group

	Serum Fetuin-A	
	r_s	P
Age	0.027	0.825
Duration	0.191	0.114
GAGS	0.155	0.200
BMI	-0.067	0.580

Data are presented as numbers. GAGS: global acne grading system, BMI body mass index, rs: Spearman coefficient, Significance level at P value <0.05.

In the present study, serum fetuin A exhibited a statistically significant 65.71% sensitivity and 62.86 % specificity for discriminating between patients and controls at a cut-off value of >14 ng/ml (p= 0.001). **Figure 1**

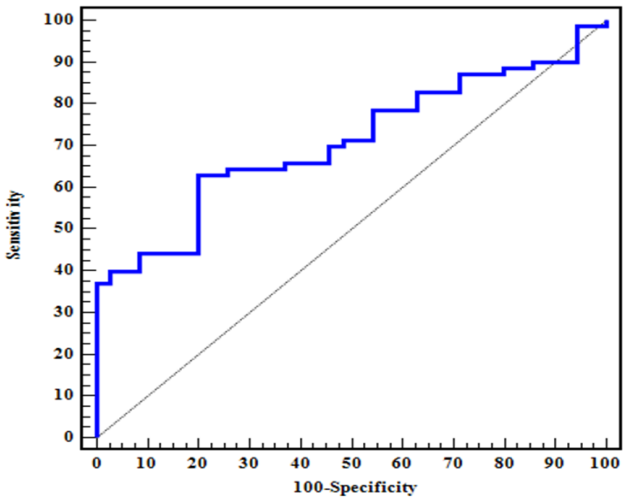

Figure 1: ROC curve for Serum Fetuin-A to discriminate patients (n = 70) from control (n = 35).

DISCUSSION

AV is a persistent chronic skin inflammatory disorder affecting the pilosebaceous follicles and affects populations worldwide [12]. Fetuin-A is a glycoprotein that is mainly produced by hepatocytes and released into the circulation in high concentrations among individuals with fatty liver disease. Insulin resistance is the result of this binding to the insulin receptor, which inhibits hepatic and muscle insulin signals. Conversely, metabolic syndrome and atherogenic lipid profiles are linked to elevated fetuin-A levels, whereas vascular calcification and inflammation are linked to decreased fetuin-A levels [12].

In the present study, the AV patients age ranged from 12 to 25 years. The study down by **Janani *et al.*** [13], has shown that the onset of puberty, which is marked by an increase in sebum secretion, is typically associated with the onset of acne. As a result, the incidence of acne increases with age, with adolescents experiencing the highest incidence and pre-pubertal children experiencing a relatively low incidence. As

individuals age, the prevalence of acne decreases after they reach late adolescence or young adulthood.

The cases had a mean age of 18.69±4.18. This agreed with the research conducted by **Lideikaite *et al.*** [14], who revealed that acne is most frequently observed in Lithuanian adolescents, with an average age of 21 years.

The current study demonstrated that the preponderance of cases were females in both the control and the cases. This agreed with the study conducted by **Carmina *et al.*** [15], who documented that hormonal acne in women is influenced by a variety of factors, including genetic predispositions, hormonal fluctuations, and exogenous factors such as diet, medication use, hygiene products and heightened stress levels.

In the present study; positive family history was found in most of patients. In line with results of the study conducted by **Melnik [16]**, who clarified that the elevated rates of acne among first-degree relatives and twins are likely to account for the genetic component of the predisposition to acne in specific individuals.

The current study showed that, most of cases have relation to diet.

Saturated lipids, refined carbohydrates, dairy products, and chocolate are all elements of a diet that can exacerbate acne by activating metabolic signals that are derived from the food [16]. Modern Western diets frequently contain substantial quantities of refined carbohydrates, However, they frequently lack long-chain omega-3 fatty acids. The development of acne is widely acknowledged to be significantly influenced by a discrepancy between omega-6 and omega-3 fatty acids [17].

The current study showed that there was association between acne and psychological stress.

These results agree with Hammill.,[18] Their findings indicated that the severity of acne is directly correlated with the degree of anxiety and the extent of self-image impairment.

Regarding to clinical data of the patients; the results of the current study indicate that the majority of AV patients have a progressive course with a gradual onset, with a duration that ranges from six months to twelve years. In line with the study of **Bhatt & Kumar.** [19], indicating the chronic nature of AV. Mean GAGS of the patients was 25.23 ± 12.03. This agreed with the study by **Ozuguz *et al.*** [20], whose findings indicated that all of their patients had moderate AV (GAGS 19–30).

In the current investigation, serum fetuin-A levels of patients with AV were significantly elevated in comparison to controls. Similarly **Saleh *et al.*** [11], showed that compared to the control group, patients with AV exhibited a significantly higher mean serum level of fetuin A. However, **Chekol *et al.*** [21], and **Genc *et al.*** [22], demonstrated that all patients with psoriasis exhibited significantly decreased fetuin-A levels than the controls. They suggested that inflammation may be the cause of this decrease, as fetuin A was previously classified as a negative acute phase protein. **Uyar *et al.*** [23], showed that the fetuin-A values of the control group and the psoriasis patients were comparable.

The serum fetuin-A level and the course of AV were found to be significantly correlated in the present study.

Uysal *et al.* [24], reported that the severity index score and psoriasis area are positively correlated with fetuin-A levels.

Fetuin-A levels were found to be significantly correlated with diet in the current study. This agreed with the study by **Akpinar Kara *et al.*** [25], who recognized increasing individuals who consuming diet filled with high-calorie, carbohydrate and high glycemic load. This diet enhances sebaceous secretion, which serves as a perfect condition for Propionibacterium acnes in addition to microorganisms that are involved in the pathogenesis of acne. Regarding positive relation of serum fetuin-A level to stress in cases of study.

This agree with study carried by **Ghodsi *et al.*** [26], whose findings indicated a positive correlation between the severity of acne and mental tension. This disagrees with the study by **Sak *et al.*** [27] who reported that fetuin-A levels were not significantly correlated with stress parameters.

LIMITATIONS

The limitations of the current study include the failure to consider additional factors and biochemical variables, the comparatively patients small number involved in the study, and the reliance on single measurements of fetuin-A for results. Consequently, it is impossible to ascertain whether any unmeasured covariates may have influenced the observations. Additionally, this could have resulted in the introduction of random measurement errors during the determination of biochemical variables.

CONCLUSION

Patients with acne exhibit elevated serum fetuin-A levels. Additionally, it exhibits statistically significant correlations with the progression of disease, stress, and diet.

As a result, it is imperative to conduct additional research to investigate the role of fetuin-A and its potential impact on the production of androgenic hormones in acne patients. These studies should include a greater number of patients and controls, as well as the evaluation of Serum Fetuin A in various types of AV. Furthermore, additional research is necessary to evaluate the efficacy of AV treatment on Serum Fetuin A in AV patients. specifically recommend a larger, multi-center study to overcome the small sample size and a longitudinal design to explore causality.

AHSG	Alpha 2-Heremans Schmid glycoprotein
AV	Acne vulgaris
BMI	Body mass index
GAGS	Global acne grading system
IL	Interleukin
O.D.	Optical density
ROC	Receiver operating characteristic curve
TNF-α	Tumor necrosis factor alpha

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