Assessment of Tumor necrosis factor alpha gene polymorphisms (-857and -863) In Acne Vulgaris Patients and their Correlation with Disease Severity

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ABSTRACT

Background: Numerous cellular processes are linked to the tumor necrosis factor alpha (TNF-α) superfamily and its corresponding receptors. Furthermore, a great deal of research has been done on their function in many pathogenic diseases, particularly in skin illnesses. The formation of acne lesions may be influenced by genetic variables that alter TNF-α expression.

Objective: To assess TNF-α polymorphisms (-857 and -863) in Egyptian patients with acne vulgaris and their correlation to disease severity.

Patients and methods: This study included 50 patients suffering from acne vulgaris of different severity (Patients group). In addition, 50 apparently healthy individuals of matched age and sex were chosen as a control group. They were recruited at random for one year duration from the outpatient clinic of Dermatology, Andrology and STD Department in Mansoura University Hospital.

Results: Regarding TNF-α −857 polymorphism, CC was considered as the reference genotype and C was considered the reference allele. Our study revealed that AV cases were significantly associated with higher proportion of CT, CT+TT genotypes ($p<0.05$ for each), with risk to develop AV (OR>1 for each). While, TT genotype in general and recessive models, as well as T allele were not associated with AV susceptibility. Regarding TNF-α −863 polymorphism, CC was considered as the reference genotype and C was considered the reference allele. Our study revealed that AV cases were significantly associated with higher proportion of AA, genotype, and A allele ($p<0.05$ for each), with risk to develop AV (OR>1 for each). CA, CA+AA were not associated with AV susceptibility.

Conclusion: We have demonstrated a statistically significant association between the TNF-α −857 and -683 single nucleotide polymorphism and the susceptibility to acne in Egyptian patients with acne vulgaris.

Keywords: Acne Vulgaris, TNF-α, Polymorphisms (-857and -863).

INTRODUCTION

Acne vulgaris (AV) is a worldwide inflammatory skin disorder affecting the sebaceous follicles that is persistent and chronic [(1)]. 9.4% of people worldwide are expected to suffer from acne, making it the ninth most common skin condition. Over 85% of teens suffer with acne, which can last until adulthood. Females are more likely to develop acne than boys, and two thirds of dermatologist visits for acne are related to acne [(2)]. The distinct lesions can be classified as inflammatory (papules, pustules, nodules, and cysts) or non-inflammatory (open/black and closed/white comedones), which can result in scar formation and skin pigmentation and require long-term, consistent treatment [(3)]. The etiology of acne is complex and involves several factors, including the complex interactions between androgen-mediated sebum generation, follicular keratinization, inflammatory responses, and Cutibacterium acnes colonization of pilosebaceous follicles [(4)].

Genetics is estimated to be the cause of 80% of cases [(5)]. TNF is a multipurpose cytokine that affects many various aspects of biological processes, including differentiation, proliferation, survival, and death of cells. TNF is a pro-inflammatory cytokine released by inflammatory cells that may have a role in the carcinogenesis of inflammation. The TNF gene promoter has a number of single nucleotide polymorphisms (SNPs), some of which may control TNF expression [(6)].

Cytokines during an inflammatory reaction, start and control the cytokine cascade. It was discovered that acne lesions had significantly higher levels of TNF-αgene transcripts [(7)].

Given the high prevalence of AV infection, which primarily affected adolescents and young adults, with 85% of them suffering from the disease, previous study sought to identify S. epidermidis in acne patients as well as the genetic polymorphisms of TNF-α and AV disease [(8)]. **Younis *et al.*** [(9)] discovered a substantial correlation between acne in the general population and the TNF-α -863 polymorphism.

The aim of the present study was to assess TNF-α polymorphisms (-857 and -863) in Egyptian patients with acne vulgaris and their correlation to disease severity.

PATIENTS AND METHODS

One hundred persons were included in this study. They were chosen from the out-patient clinic of Dermatology, Andrology and STDs Department, Mansoura University Hospitals within a duration of one year.

They were separated into two groups:

- **Patients group**: 50 patients with acne vulgaris.
- **Control group:** 50 healthy controls of matched age and sex.

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Inclusion Criteria:

All participants included in the study had typical acne vulgaris lesions of different severity.

Exclusion criteria:

- Patients with history of topical or systemic therapy for acne vulgaris in the last 2 months.
- Subjects with a history or clinical evidence of:
  - Acute or chronic infection.
  - Diabetes mellitus, cardiovascular disease, chronic renal or liver disease or endocrinopathies.
  - Pregnancy.
  - Malignancy.
  - Autoimmune diseases.
  - Using systemic drugs as non-steroid anti-inflammatory drugs or immune modulators as steroids.

All patients underwent the following:

1. **Complete history taking including** age, sex, duration of disease, presence of family history of AV, smoking and previous medical history.
2. **Clinical examination:**
   - Complete general examination.
   - Complete cutaneous examination including determination of type of acne vulgaris lesions and areas involved.

3. Assessment of disease severity using Global Acne Grading System (GAGS): according to **Doshi *et al.*[(10)].**

The severity was graded as: Mild with score 1 – 18, Moderate with score 19 – 30, Severe with score 31 – 38, and Very severe if the score was more than 38.

4. Genetic study:

Assessment of TNF-α polymorphisms (-857 and -863) by polymerase chain reaction (PCR) conducted at the Medical Biochemistry Department.

Sampling:

All individuals underwent venipuncture to acquire a sample of two milliliters (2 ml) of peripheral blood, which was then transferred to EDTA tubes, appropriately labeled, and kept at -20°C for further molecular analysis.

Molecular analysis:

1) **DNA extraction:** Spin column approach (GeneJET Whole Blood Genomic DNA Purification Mini Kit - Thermo Scientific, USA, Cat. no. K0781) was used to extract genomic DNA from peripheral blood leucocytes. DNA concentration and purity were evaluated using a (Thermo Scientific, USA), NanoDropTM 2000 Spectrophotometer.
2) **Polymerase Chain Reactions (PCR)** Detection of TNFα (-857and -863) gene polymorphisms by allele-specific polymerase chain reaction (AS-PCR).

   PCR was done in a 25 µL reaction volume for DNA samples that contained 5 µL DNA template (20 ng), 1 µL of each primer, and 12.5 µL 2X PCR Master Mix (Cosmo PCR Red M.Mix, willowfort, England), and 5.5 µL nuclease free water.

   PCR was done in a programmed thermal cycler (Applied Biosystems, model 2720) as follows: initial denaturation at 95°C for 60 s, followed by 35 cycles of 30 s at 94°C, 30 s at 55°C, 40 s at 72°C, and then a final extension for 5 minutes at 72°C.
3) **Agarose gel electrophoresis:** After the amplification, electrophoresis was performed on 1.5% agarose gel stained with ethidium bromide (0.5 µg/µL) for separation of the PCR products with the help of 50 bp DNA ladder (Thermo Scientific, USA, Cat. no #SM0373). The amplified PCR products were seen under ultraviolet light.

Ethical approval:

Mansoura Faculty of Medicine's Institutional Review Board approved this report (MS.21.10.1706). Participants provided informed permission before being included in the trial. Every precaution was made to ensure the data's privacy. All data was utilized solely for research reasons. The Helsinki Declaration was observed at all stages of the study.

Statistical Analysis

With SPSS Version 25.0, the gathered data were coded, tabulated, and statistically managed. To verify if the data distribution was normal, the Shapiro-Wilk test was used. The mean± SD, median, and range were used to express the numerical data. The proportion and frequency were used to express the non-numerical data. The statistical significance of the difference in the means of the two study groups was evaluated using the Student T-Test and for more than two study group, one-way ANOVA test was employed. The association between two qualitative variables was investigated using the X^2-test. When the predicted count is less than five in more than 20% of cells, the Monte Carlo test was utilized to investigate the association between two qualitative variables. When utilizing generalized linear models to predict risk variables for a categorical dependent variable, logistic and ordinal regression analyses were employed. An indicator of the degree of correlation between an exposure and an outcome is the odds ratio, or OR. The accuracy of the OR is estimated using the 95% CI. In contrast, a narrow confidence interval (CI) denotes a better degree of OR accuracy than a wide CI. A p-value is deemed significant when it was less than 0.05 at the 95% confidence interval.

RESULTS

The current study included 50 patients with AV (patient group). Mean age was 18.2 years, ranging from 13 to 29. Additionally, there were 50 healthy control volunteers (control group). No statistically significant differences were identified between the two groups for age, sex, and BMI (Table 1).

Table (1): Comparison of demographic and anthropometric data among group of patients with acne vulgaris and control group.

	Acne vulgaris patients group n = 50	Control group n = 50	P
Age (years)			
Mean ± SD.	18.22 ± 3.86	18.98 ± 3.80	0.324
Sex			
Male	13 (26%)	20 (40%)	0.137
Female	37 (74%)	30 (60%)	
BMI (kg/m^2)			
Mean ± SD.	23.90 ± 3.14	23.10 ± 2.39	0.154

SD: standard deviation,

TNF genotypes and alleles among cases and controls

Regarding TNF-α −857 polymorphism, CC was considered as the reference genotype and C was considered the reference allele. AV cases were statistically significantly associated with higher proportion of CT, CT+TT genotypes, with risk to develop AV (OR>1 for each). While, TT genotype in general and recessive models, as well as T allele were not statistically associated with AV susceptibility.

Regarding TNF-α −863 polymorphism, CC was considered as the reference genotype and C was considered the reference allele. AV cases were statistically significantly associated with higher proportion of AA, genotype, A allele, with risk to develop AV (OR>1 for each). CA and CA+AA were not associated statistically with AV susceptibility (Table 2).

Table (2): Comparison of TNF-α −857 and TNF-α −863 polymorphism among acne vulgaris patients and control group.

		Acne Vulgaris n = 50		Control n = 50		P value	OR (95 % CI)
		N.	%	N.	%		
TNF-α −857 polymorphism							
Genotypes	**CC**	21	42	31	62	-	**Reference**
	CT	26	52	14	28	0.021*	2.741(1.167-6.439)
	TT	3	6	5	10	0.877	0.886(0.191-4.110)
Dominant model	**CC**	21	42	31	62	-	**Reference**
	CT + TT	29	58	19	38	0.047*	2.253(1.011-5.019)
Recessive model	**CC + CT**	47	94	45	90	-	**Reference**
	TT	3	6	5	10	0.465	0.574(0.130-2.545)
Alleles	**C**	68	86.0	76	76.0	-	**Reference**
	T	32	32.0	24	24.0	0.209	1.490(0.800-2.776)
TNF-α −863 polymorphism							
Genotypes	**CC**	8	16	13	26	-	**Reference**
	CA	18	36	25	50	0.773	1.170(0.402-3.408)
	AA	24	48	12	24	**0.039***	3.250(1.060-9.967)
Dominant model	**CC**	8	16	13	26	-	**Reference**
	CA + AA	42	84	37	74	0.223	1.845(0.689-4.941)
Recessive model	**CC + CA**	26	52	38	76	-	**Reference**
	AA	24	48	12	24	**0.014***	2.923(1.245-6.865)
Alleles	**C**	34	34.0	51	51.0	-	**Reference**
	A	66	66.0	49	49.0	**0.016***	2.020(1.143-3.573)

*: Significant; N; number, OR; odds ratio, CI; confidence interval, Reference, according to NCBI database; C, Cysteine; T, thymine; P<0.05 is considered significant; OR<1 is considered protective; OR>1 is considered risky.

TNF-α haplotypes among cases and controls

Table (3) shows that TNF-α −857 -683 haplotypes were evaluated. CA haplotype showed the highest frequency among cases and controls. Considering CC as the reference haplotype, TA showed statistically higher frequency associated with AV cases, with risk to develop AV (OR>1) as compared to control.

Table (3): Comparison of TNF-α −857 -683 haplotypes' frequency among acne vulgaris patients and control group.

		Acne Vulgaris	Control	P-value	OR (95 % CI)
TNF-α −857 -683 haplotypes	CC	0.224	0.366	-	Reference
	CA	0.456	0.394	0.195	1.484(0.817-2.695)
	TA	0.204	0.096	**0.031***	2.092(1.936-4.672)
	TC	0.116	0.144	0.617	0.735(0.541-2.818)

*: Significant

Association of TNF-α −857 polymorphism with studied parameters

Table (4) shows that no statistically significant association was found between TNF-α −857 polymorphism with sex and BMI among control subjects and acne vulgaris cases.

Table (4): Association between TNF-α −857 polymorphism with sex and BMI among control subjects and acne vulgaris patients.

	TNF-α −857 control subjects			Test (p1)	Post hoc test		
	CC n = 31	CT n =14	TT n = 5		P2	P3	P4
Sex; N (%)							
Male	15 (48.4%)	4 (28.6%)	1 (20.0%)	X2=2.504 P=0.272	0.213	0.355	0.709
Female	16 (51.6%)	10 (71.4%)	4 (80.0%)				
BMI							
Mean ± SD.	23.53 ± 2.54	22.26 ± 2.02	22.76 ± 2.11	ANOVA= 1.444 p=0.246	0.229	0.780	0.913
	TNF-α −857 acne vulgaris patients			**Test (p1)**	**Post hoc test**		
	CC n = 21	**CT n = 26**	**TT n = 3**		P2	P3	P4
Sex; N (%)							
Male	8 (38.1%)	4 (15.4%)	1 (33.3%)	X^2=3.203 P=0.197	0.076	0.873	0.446
Female	13 (61.9%)	22 (84.6%)	2 (66.7%)				
BMI							
Mean ± SD.	24.00 ± 3.55	23.91 ± 3.02	23.08 ± 0.38	ANOVA= 0.108 p=0.898	0.995	0.889	0.906

P1: comparison between CC, CT and TT; P2: comparison between CC and CT; P3: comparison between CC and TT; P4: comparison between CT and TT

Table (5) shows that no statistically significant association was found between TNF-α −857 polymorphism with GAGS score and acne severity among AV cases.

Table (5): Association between TNF-α −857 polymorphism with score and severity among acne vulgaris patients according to GAGS score [(10)].

	TNF-α −857			Test (p1)	Post hoc test		
	CC n = 21	CT n = 26	TT n = 3		P2	P3	P4
GAGS score							
Mean ± SD.	30.2±7.7	26.6±6.2	31.3±3.6	F=1.127 P=0.333	0.360	0.976	0.386
Acne Severity							
Mild	2 (9.5%)	6 (23.1%)	1 (33.3%)	X2=9.014 MCp=0.110	MC 0.187	MC 0.143	MC 0.172
Moderate	9 (42.9%)	14 (53.8%)	0 (0.0%)				
Severe	8 (38.1%)	3 (11.5%)	1 (33.3%)				
Very severe	2 (9.5%)	3 (11.5%)	1 (33.3%)				

P1: comparison between CC, CT and TT; P2: comparison between CC and CT; P3: comparison between CC and TT; P4: comparison between CT and TT

Association of TNF-α −863 polymorphism with studied parameters

Table (6) shows that no statistically significant association was found between TNF-α −863 polymorphism with sex and BMI among control subjects and acne vulgaris cases.

Table (6): Association between TNF-α −863 polymorphism with sex and BMI among control subjects and acne vulgaris patients.

	TNF-α −863 control subjects			Test (p1)	Post hoc test		
	CC N=13	CA N=25	AA N=12		P2	P3	P4
Sex; N(%)							
Male	5 (38.5%)	9 (36.0%)	6 (50.0%)	X2=0.679 P=0.715	0.881	0.561	0.488
Female	8 (61.5%)	16 (64.0%)	6 (50.0%)				
BMI							
Mean ± SD.	22.60 ± 2.94	23.14 ± 2.26	23.55 ± 2.11	ANOVA= 0.489 p=0.616	0.794	0.592	0.877
	TNF-α −863 acne vulgaris patients			**Test (p1)**	**Post hoc test**		
	CC n = 8	**CA n = 18**	**AA n = 24**		P2	P3	P4
Sex; N(%)							
Male	1 (12.5%)	6 (33.3%)	6 (25.0%)	X2=1.273 P=0.504	0.375	0.646	0.732
Female	7 (87.5%)	12 (66.7%)	18 (75.0%)				
BMI							
Mean ± SD.	23.46 ± 3.55	23.04 ± 2.51	24.68 ± 3.35	ANOVA= 1.532 p=0.227	0.946	0.603	0.217

P1: comparison between CC, CT and TT; P2: comparison between CC and CT; P3: comparison between CC and TT; P4: comparison between CT and TT

Table (7) shows that no statistically significant association was found between TNF-α −863 polymorphism with GAGS score and acne severity among acne vulgaris patients.

Table (7): Association between TNF-α −863 polymorphism with GAGS score and acne severity among acne vulgaris patients.

	TNF-α −863			Test (p1)	Post hoc test		
	CC n = 8	CA n = 18	AA n = 24		P2	P3	P4
GAGS score							
Mean ± SD.	28.9±5.7	28.7±5.0	28.0±5.9	F=0.046 P=0.955	0.999	0.969	0.799
Acne Severity							
Mild	2 (25.0%)	2 (11.1%)	5 (20.8%)	X2=2.189 MCp=0.953	MC 0.921	MC 1.0	MC 0.811
Moderate	3 (37.5%)	9 (50.0%)	11 (45.8%)				
Severe	2 (25.0%)	4 (22.2%)	6 (25.0%)				
Very severe	1 (12.5%)	3 (16.7%)	2 (8.3%)				

P1: comparison between CC, CT and TT; P2: comparison between CC and CT; P3: comparison between CC and TT; P4: comparison between CT and TT.

Prediction of AV susceptibility and severity

Logistic regression analysis was conducted for prediction of acne vulgaris susceptibility, using BMI and TNF polymorphisms. TNF 857 CT and TNF 863 TT were considered statistically unfavourable risk predictors for AV susceptibility in uni- and multivariate analyses (Table 8).

Table (8): Logistic regression analysis for prediction of acne vulgaris susceptibility.

	Logistic regression	Univariate		Multivariate	
		p	**OR (95% CI)**	**p**	**OR (95% CI)**
BMI		0.157	1.112(0.960-1.289)		
TNF 857	CC	-	**Reference**		
	CT	0.021*	2.741(1.167-6.439)	0.019*	2.903(1.194-7.055)
	TT	0.877	0.886(0.191-4.110)	0.982	0.918(0.207-5.005)
TNF 863	CC	-	**Reference**		
	CT	0.773	1.170(0.402-3.408)	0.899	1.075(0.354-3.260)
	TT	0.039*	3.250(1.060-9.967)	0.049*	3.203(1.002-10.237)

OR; odds ratio, CI; confidence interval, Reference, according to NCBI database; C, cysteine; A, adenine; P<0.05 is considered significant; OR<1 is considered protective; OR>1 is considered risky.

Ordinal regression analysis was conducted for prediction of acne vulgaris severity using age, sex, BMI, TNF polymorphisms. None was considered risk predictor for AV severity (Table 9).

Table (9): Ordinal regression analysis for prediction of acne vulgaris susceptibility.

Ordinal regression	p	OR (95% CI)
Age	0.117	0.901(0.827-1.182)
Sex	0.068	1.899(0.954-3.781)
BMI	0.993	1.002(0.907-1.107)
Duration	0.803	1.048(0.726-1.511)
TNF 857		
CC	-	**Reference**
CT	0.822	0.860(0.232-3.191)
TT	0.364	0.547(0.149-2.011)
TNF 863		
CC	-	**Reference**
CT	0.940	1.034(0.437-2.446)
TT	0.389	1.335(0.692-2.577)

DISCUSSION

AV is an inflammatory skin condition affecting the pilosebaceous unit, mostly affecting the face and trunk. It affects around 9% of people globally, with 85% of patients aged 12-24 and 50% of patients aged 20-29 being affected [(11)]. Since all acne lesions were discovered to be inflammatory, an inflammation in the pilosebaceous unit region can be thought of as a distinctive feature of acne [(12)].

Propionibacterium acnes (P. acnes) stimulates proinflammatory cytokines, such as TNF-α and interleukin 1 alpha (IL-1α), which in turn cause inflammation in the development of acne [(13)]. TNF α is mostly linked to inflammatory cascades, while IL1 α is primarily linked to comedogenesis [(14)]. Based on research conducted in vitro and in vivo, P. acnes is thought to have a role in the pathophysiology of acne by generating IL-1β oversecretion and activating inflammasomes [(15)].

Given that some patients with severe acne have a family history of severe acne vulgaris, genetic factors are thought to play a significant role in the pathophysiology of acne [(16)]. Researchers are interested in polymorphisms of genes encoding significant cytokines that are crucial in the etiology of acne (such as TNF gene variations as -308, -863, -857, -238, -1031) [(17)]. A preliminary investigation on the Pakistani population revealed a correlation between TNF-α −238 and −308 polymorphisms and acne [(7)]. Thus, the formation of acne lesions may be influenced by genetic variables that alter TNF-α expression [(18)].

Al-Hilali *et al.* [(7)] suggested that, the TNF-α -308 G>A and -238 G>A SNPs are linked to the pathophysiology of acne. There was a correlation found between the AA genotypes of -308 and -238 with a higher likelihood of acquiring AV. When comparing the sick group to the HC group, there was a noticeably greater frequency of the mutant A allele at positions -308 and -238. Additionally, a strong and statistically significant correlation between the severity of acne and the variation -308 and -238 genotypes was seen in these data. According to the research's findings, the pathophysiology of acne in the study population may be influenced by the TNF promoter SNPs at positions -308 and -238, in the research population.

A TNF-α promoter polymorphisms association research in a community of Central Europeans revealed that the TNF-α −857C/T polymorphism protects against the development of acne, but the TNF-α −863C/A and −1031 T/C polymorphisms do not [(8)].

In the present study, the mean age of AV patients was 18.2 ± 3.86, ranged from 13 to 29 years. They were 74% females and 26% males. The mean age of our patients was in concordance with **Gurung *et al.*** [(19)] who found that the average age of his research participants was 21.26 (SD=5.94), with the youngest person being 13 years old and the oldest participant being 56 years. Moreover, the mean age was found to be 14.7 in the study of **Uslu *et al.*** [(20)] and 16.7 in the study of **Yahya** [(21)]. Tthese dissimilar results in AV mean age may be explained by the finding that the way acne presents is impacted by demographics. Usually, acne appears at the same time as puberty, when sebum production rises. As a result, acne is more common as people age, with teens having the greatest incidence and prepubescent children having a comparatively low frequency. Acne prevalence rates tend to decrease with age as a person reaches late adolescence or early adulthood [(22)].

Our result revealed that females with AV were predominant. Similarly, **Kassem *et al.*** [(23)] found of their patients 36 (34%) were males, and 71 (66%) were females. Likewise, **Gurung *et al.*** [(19)] reported that there were 73 (66.4%) more female participants than male ones (37, 33.6%). However, it was discovered by **Kaushik *et al.*** [(24)] that acne impacted men twice as often as it did women. These findings can be the consequence of variations in the characteristics of the nation under study or the sampled population [(25)].

The range of the illness duration in our study was 0.3 to 4 years, with a median of 1.5 years. In line with this, **Shetty *et al.*** [(26)] reported that the duration of onset of the illness was more than 3 months which implies that the disease was chronic and recurrent.

In the current study, median GAGS score was 28 (ranged from 14 to 49). Among all studied cases, 18% had mild, 46% had moderate, 24% had severe and 12% had very severe grades according to GAGS score. In consistent with our findings, **Naveed *et al.*** [(27)] found that the average GAGS score was 19.8 (±7.7, range 10-40); 64 (39.3%), 82 (50.3%), and 17 (10.4%) of the patients had mild, moderate, or severe acne respectively. Furthermore, the results contradicted

those of **Kaushik *et al.*** [24] who found that 82% of research participants had mild acne, 16% had moderate acne, and just 2.5% had severe acne based on the GAGS score.

Regarding TNF-α −857 polymorphism, CC was considered as the reference genotype and C was considered the reference allele. Our study revealed that AV cases were statistically significantly associated with higher proportion of CT, CT+TT genotypes ($p<0.05$ for each), with risk to develop AV (OR>1 for each). While, TT genotype in general and recessive models, as well as T allele were not associated with AV susceptibility.

Regarding TNF-α −863 polymorphism, CC was considered as the reference genotype and C was considered the reference allele. Our study revealed that AV cases were statistically significantly associated with higher proportion of AA, genotype, and A allele ($p<0.05$ for each), with risk to develop AV (OR>1 for each). CA and CA+AA were not associated with AV susceptibility.

In the current study, TNF-α −857 -863 haplotypes were evaluated. CA haplotype showed the highest frequency among cases and controls. Considering CC as the reference haplotype, TA showed statistically higher frequency associated with AV cases ($p<0.05$), with risk to develop AV (OR>1).

In Younis *et al.* [9] study, to explore the role of three polymorphisms in the inflammatory events of acne development, genotyping was done on the TNF-α gene promoter region at −857C/T, −863C/A, and −1031 T/C. 468 patients and 297 controls had their TNF-α −857C/T genotypes determined. The general model of association penetrance ($p = 0.072$) showed that there was no statistically significant difference in the genotype frequency between the control and patient groups.

Similar to our results, **Younis *et al.*** [9] reported that according to the general genotyping model, patients had higher major (C/C) and heterozygous (C/T) genotype frequencies, whereas minor (T/T) genotype rates were the same in both groups. In line with our results, the TNF-α −857 T allele may provide protection against acne. Additionally, the additive model indicates that the main allele frequency was higher in the sick group compared to the control group ($p = 0.140$).

Younis *et al.* [9] examined the TNF-α −863C/A polymorphism in 441 patients and 303 controls. The genotype distribution of the C/A polymorphism at −863 position showed a significant difference between the patient and control groups ($p = 0.009$), which is similar to our results. It's interesting to note that the control group had higher rates of major (C/C) and minor (A/A) genotypes, whereas the sick group had higher frequencies of the heterozygous genotype (C/A) and this is not similar to our results. Although the p-value was not significant, the comparison of allele frequency in the additive model showed that minor allele (A) was more common in patients than in controls. The analysis of TNF-α −1031 T/C polymorphism in 505 patients and 310 controls revealed that AV is not linked to this polymorphism. TNF-α −863C/A polymorphism has been linked to AV, according to research on TNF-α promoter polymorphisms [9]. In line with our results, in Pakistani populations, TNF-α polymorphism at −1031 T/C is not linked to the etiology of acne, and the TNF-α −857 T allele may have a protective effect against acne illness.

Similarly, Szabó *et al.* [28] found the −857 T allele in the population of Central Europe plays a protective effect. The LD Matrix analysis revealed a correlation between the AV and −863 A and −863 C alleles.

In our study, TNF-α −857 and −863 polymorphisms did not have a significant connection with age, gender, or BMI in AV patients. There was no significant connection between TNF-α −857 and −863 polymorphisms and AV score or severity. **Younis *et al.*** [9] have also discovered that there was no correlation between the severity of acne and TNF-α polymorphisms and serum levels. Nonetheless, there wasn't many distinctions between the Central European and Pakistani populations when it comes to the relationship between TNF-α polymorphisms and acne and acne severity.

Consistent with these findings, **Szabó *et al.*** [28] did not discover a correlation between −863 and acne in groups based on severity or gender.

The current investigation might serve as a first evaluation of the function of genetically modified TNF α variants in the development of AV.

CONCLUSIONS

We have demonstrated a statistical significant association between the TNF-α −857 and -683 single nucleotide polymorphism and the susceptibility to acne in Egyptian patients with acne vulgaris.

Current study proved that no statistically significant association was found between TNF-α −857 and −863 polymorphisms with GAGS score and acne severity among AV cases.

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