Research Article / Araştırma Makalesi

Investigation of Vaspin and Visfatin -4689G/T Gene Polymorphisms in Alopecia Areata Patients

Alopesi Areata Hastalarında Vaspin ve Visfatin -4689G/T Gen Polimorfizmlerinin Araştırılması

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Abstract: Alopecia Areata (AA) is a chronic autoimmune condition that causes recurrent hair bereavement. Genetic and immunological factors act a part in the pathogenesis of AA. The aim of this study was to look into relationship between the vaspin and visfatin -4689G/T gene polymorphisms and AA sensibility in the Turkish population. This study included 80 AA patients and 80 healthy controls. Genomic DNA was extracted of blood samples Vaspin and visfatin -4689G/T gene polymorphisms were determined using polymerase chain reaction (PCR) and restriction fragment length polymorphism (RFLP) methods. The observed disparity in vaspin genotypes and allele distribution amid AA patients and healthy controls did not reach statistical significance ($\chi 2$ = 2.51, df = 1, p = 0.11 and $\chi 2$ = 1.75, df = 1, p = 0.18, respectively). Although visfatin GT genotype was higher in AA patients compared to control, it was not statistically significant. People with the visfatin GT genotype were more likely to be AA than people with the GG genotype [OR (95% CI) = 2.11 (1.04-4.27), p = 0.03]. This study shows that there is no affair amid vaspin and visfatin -4689G/T polymorphism and AA in the Turkish population. However, the TT genotype for the vaspin gene and the GT genotype for the visfatin -4689G/T gene are risk factors for people with AA disease.
Keywords: Alopecia areata, adipokine, vaspin, visfatin, single nucleotide polymorphism.

Özet: Alopecia Areata (AA), tekrarlayan saç kaybına neden olan kronik bir otoimmün durumdur. AA patogenezinde genetik ve immünolojik faktörler rol oynamaktadır. Bu çalışmanın amacı Türk toplumunda vaspin ve visfatin -4689G/T gen polimorfizmleri ile AA duyarlılığı arasındaki ilişkiyi araştırmaktır. Bu çalışmaya 80 AA hastası ve 80 sağlıklı kontrol dahil edildi.
Kan örneklerinden genomik DNA elde edildi. Vaspin ve visfatin -4689G/T gen polimorfizmleri, polimeraz zincir reaksiyonu (PCR) ve restriksiyon fragman uzunluğu polimorfizmi (RFLP) yöntemleri kullanılarak belirlendi. AA hastaları ve sağlıklı kontroller arasında vaspin genotipleri ve alel dağılımında gözlenen farklılık istatistiksel anlamlılığa ulaşmadı (sırasıyla, $\chi 2$ = 2.51, df = 1, p = 0.11 and $\chi 2$ = 1.75, df = 1, p = 0.18). Visfatin GT genotipi AA hastalarında kontrole göre daha yüksek olmasına rağmen istatistiksel olarak anlamlı değildi. Visfatin GT genotipine sahip kişilerin AA olma olasılığı, GG genotipine sahip kişilere göre daha yüksekti [OR (95% CI) = 2.11 (1.04-4.27), p = 0.03]. Bu çalışma Türk toplumunda vaspin ve visfatin -4689G/T polimorfizmi ile AA arasında bir ilişki olmadığını göstermektedir. Bununla birlikte, vaspin geni için TT genotipi ve visfatin -4689G/T geni için GT genotipi AA hastalığı olan kişiler için risk faktörleridir.
Anahtar Kelimeler: Alopesi areata, adipokin, vaspin, visfatin, tek nükleotid polimorfizmi.

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1. Introduction

Hair follicles are also recognized as an immunologically specialized component of the skin and are known to possess important characteristics in defense against pathogenic microorganisms (1,2). The hair follicle is composed of concentric layers, each with distinct functions and characteristics. Recent molecular-level studies have revealed that these layers also exhibit different immunological properties (1-3). It has been observed that some of the common conditions associated with hair loss and alopecia are rooted in etiopathogenetic mechanisms involving the disruption of the natural immune privilege of hair follicles (4). Alopecia areata (AA) is an autoimmune skin disorder that targets hair follicles (5). The AA, which is characterized clinically by patchy hair loss resulting from a T-cell response to follicular antigens, has been the subject of numerous studies. These studies have supported the important role of T-cells and autoimmunity in the pathogenesis of the disease (6). The AA is results from T-cell-mediated inflammation in the hair follicle area, which disrupts its function and the hair growth cycle without damaging the follicle. Therefore, the loss of hair follicle immune prerogative, autoimmune destruction of hair follicle intervene by cytotoxic mechanisms, and the upregulation of inflammatory pathways all play an important role in the AA (7).

The multifactorial dermatological disorder AA is defined by non-marking hair loss on the scalp or any other hair-bearing area (8,9). Multiple investigations have demonstrated that AA impacts approximately 1% to 2% of the overall population, with a conjectural lifespan risk of 1.7% (10,11). It is unknown what specifically causes AA. However, it is thought to be a disease that occurs with the effect of autoimmunity, environmental factors and genetic predisposition (12). A family history is present in 20% of AA patients, indicating a hereditary susceptibility (13). Therefore, AA can be viewed as an immune-mediated disease with hereditary predisposition.

Vaspin, derived from visceral adipose tissue, holds prominence as one of the most notable identified adipokines due to its role as a serine protease inhibitor (14). Recent research has shown that vaspin can influence vascular cells, have anti-inflammatory and anti-apoptotic properties, and cause insulin resistance (15). Keratinocytes have been found to be the primary source of vaspin in human skin. The vaspin is linked to keratinocyte development and inhibits inflammatory mediator expression in the skin. It has been suggested that it contributes significantly to the pathophysiology of various inflammatory diseases, including psoriasis (16,17). The vaspin gene has six exons and five introns and is located on chromosome 14q32.13 (14). Vaspin (rs2236242) gene polymorphism has been examined in many ailments like obesity, cardiovascular disease, polycystic ovary syndrome, metabolic syndrome and diabetes mellitus (18).

Visfatin is a 52 kDa protein that is principally generated in mice and human visceral adipose tissue (19). There are 11 exons and 10 introns in the visfatin gene, which is found on chromosome 7q22.2.2 (20). Visfatin and other inflammatory cytokines disrupt insulin pathways and signaling, hence the genes that regulate these cytokines are linked to type 2 diabetes, insulin resistance, obesity and inflammation (21).

The link between visfatin -4689G/T and vaspin and AA illness has not been studied in the literature. Therefore in this study, we objected to examine the influence of the vaspin and visfatin -4689G/T gene polymorphisms in AA patients as well as to link the genotypes found in these individuals with other clinical characteristics of AA. The impact of the visfatin -4689G/T and vaspin gene polymorphisms on AA illness is being examined for the first time in this study.

2. Materials and Methods

Ethics Committee Approval: The study was approved by Kütahya Health Sciences University Noninterventional Clinical Research Ethical Committee (Decision no:

2022/09-28, Date: 14. 09.2022). The study protocol was in adherence with the principles in the Declaration of Helsinki. Informed consent was obtained from all participants.

2.1. Study Cohort

This study was performed with 80 patients aged between 18-70 years who were diagnosed with AA and 80 healthy controls with no past or family history of AA in the dermatology outpatient clinic of Kütahya Health Sciences University Faculty of Medicine. The patient and control groups were randomly selected from the patients who applied to the outpatient clinic. The dermographic characteristics as family history, duration of disease, nail dystrophy, autoimmune disease, and AA severity of the patients were recorded. Exclusion criteria included individuals with autoinflammatory diseases, pregnancy, lactation, malignancies and chronic drug intake.

2.2. DNA Isolation

Classical phenol-chloroform extraction method was used to isolate genomic DNA. The obtained products were visualized in 0.7% gel electrophoresis and the DNA concentration was evaluated by reading the OD260 value at the optical density at 260 nm on the photometer. The obtained DNAs were stored at -20°C until analysis.

2.3. Analysis of Vaspin Gene Polymorphism

The PCR method was used to determine the genotype using appropriate primers for the vaspin gene region. The primary sequences were as follows: F0: 5'-GGA GGC AGA CCA GGC ACT AGA AA-3', R0: 5'-ACC ATC TCT CTG GCT TCA GGC TTC-3', FI: 5'-AAG ACG CCG CTT CTG TGC ACT-3', R1: 5'-CAC AGG GAC CCA GGA TAA CTT GCT-3' (16). 20 µl was the total volume used for the PCR amplification. 100 ng of genomic DNA sample obtained from peripheral blood, 10 µl PCR master mix, 0,8 µl F0 and R0 primer, 1 µl F1 and R1 primer, were added to the reaction mixture. The steps in the PCR amplification technique were as follows: a three-minute initial denaturation at 95°C; thirty seconds of denaturation at 95°C; forty seconds of annealing at 62.2°C; one minute of extension at 72°C; and five minutes of final extension at 72°C. Electrophoresis was performed on a 2% agarose gel containing ethidium bromide for PCR products. The gel was viewed using the ultraviolet imaging system, and the bands that were seen were assessed and genotyped. Allele nomenclature was as follows: 174-378 bp TT, 248-378 bp AA, 174-248-378 bp AT (Figure 1).

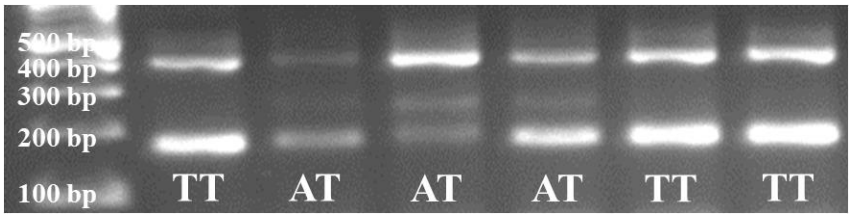

Figure 1. The PCR for the detection of vaspin rs2236242 gene polymorphism. Product sizes were 174 bp - 378 bp for the TT genotype, 174 bp - 248 bp - 378 bp for the AT genotype, and 248 bp - 378 bp for the AA genotype. M: DNA molecular weight marker.

2.4. Analysis of Visfatin -4689G/T Gene Polymorphism

Visfatin -4689G/T gene polymorphism were analyzed by PCR-RFLP. Genomic DNA amplification was conducted via PCR utilizing a Thermal Cycler (Thermo Scientific, Lithuania [European Union]). PCR was performed with a 25 µl reaction mixture containing 100 ng DNA, 12,5 µl of PCR master mix (abm, Canada), 1 µl forward (5'-TGC TGT TTT CAC ATC CTC CA-3') and reverse primers (5'-AGG GCA AAA ATG GTG CTC ATC-3'). The steps in the PCR amplification technique were as follows: 5 minutes of initial denaturation at 95°C; 30 cycles of denaturation at 95°C for 30 seconds; 40 seconds of annealing at 57°C; and 2 minutes of extension at 72°C, with a final 5

minutes of extension at 72°C. PCR products were treated with AluI restriction enzyme at 37°C for 16 hours. These samples were electrophoresed using 5% ethidium bromide on a 2% agarose gel. The gel was viewed using an ultraviolet imaging device, and genotyping was done after analyzing the bands that were visible. The PCR product sizes of visfatin -4689G/T gene polymorphism were 185-215 bp for TT, 185-260 bp for GG, and 185-215-260 bp for GT (Figure 2).

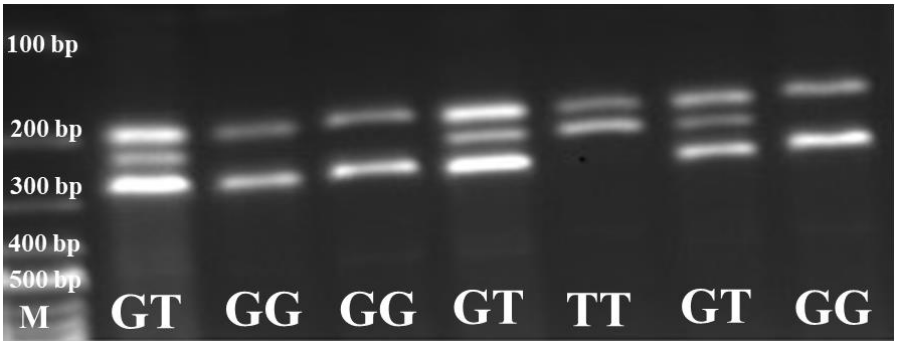

Figure 2. Electrophoresis of the visfatin -4689G/T gene polymorphism by enzyme digestion. Product sizes were 185 bp - 215 bp for the TT genotype, 185 bp - 260 bp for the GG genotype, and 185 bp - 215 bp - 260 bp for the GT genotype. M: DNA molecular weight marker.

2.5. Statistical analysis

Statistical analysis of the study data was performed using the Statistical Package for the Social Sciences (SPSS) program (IBM SPSS Statistics for Windows, Version 20.0: IBM Corp.). The independent Student's t-test was used to compare the clinical values of the two groups generated for any attribute. Chi-square ($\chi 2$) test was used to evaluate the frequency of genotypes and alleles of vaspin and visfatin gene polymorphisms. ANOVA and an independent Student t-test were used to evaluate the clinical parameter values between the AA groups with the vaspin and visfatin -4689G/T gene polymorphisms. The results are shown as mean ± standard deviation. Chi-square ($\chi 2$) test was used to display categorical data, such as gender, as numbers and percentages. The accepted threshold for statistical significance was set at $p < 0.05$.

3. Results

3.1. Demographics and clinical characteristics of the study population

This study included 80 AA disease cases and 80 healthy controls. The study's participant groups' demographic and clinical data were analyzed. Table 1 displays the acquired results. The age and gender distributions of the AA group and the control group do not differ significantly ($p = 0.08$, $p = 0.113$).

Table 1. The clinical characteristics of control and AA groups

Clinical findings	AA (n = 80)	Control (n = 80)	p value
Age (years)	31.6 ± 10.4	34.8± 12.5	0.08
Gender (n%)			
Male	44 (55.0)	33 (41.2)	0.113
Female	36 (45.0)	47 (58.8)	
Disease duration (months)	11.3 ± 22.3	-	
Alopecia severity			
< 25%	64 (80.0)		
25-50%	16 (20.0)	-	
> 50%	-		
Alopecia localization (n%)			
Scalp	55 (68.8)		
Beard/Mustache	15 (18.8)		
Hair/Beard	10 (12.5)	-	
Body	-		
Nail dystrophy (n%)			
Yes	11 (13.8)	-	
No	69 (86.2)		
Family history (n%)			
Yes	14 (17.5)	-	
No	66 (82.5)		
Other skin disease (n%)			
Yes	11 (13.8)	-	
No	69 (86.2)		
Other autoimmune disease (n%)			
Yes	5 (6.2)	-	
No	75 (93.8)		

AA: Alopecia Areata. Age, disease duration were described as mean ± standard deviation (SD) and determined by independent Student t-test. Proportion n % was determined by chi-square.
$p \leq 0.05$ is considered signifcant.

3.2. Hardy–Weinberg equilibrium

Table 2 shows that while the patient group and control groups were not in balance with regard to the vaspin gene polymorphism (p = 0.02 and p = 0.001), the observed and expected frequencies of the visfatin -4689G/T gene polymorphism in the patient and control groups were in Hardy-Weinberg equilibrium (p = 0.349 and p = 0.08).

Table 2. Hardy-Weinberg equilibrium for vaspin rs2236242 and visfatin -4689G/T gene polymorphisms on AA patients and controls

Vaspin rs2236242						
Genotypes	**Observed**	**Expected**	**χ2**	**p**	**Alleles**	**Frequency**
AA						
AA	0	3.2			A	0.80
AT	32	25.6	5	0.02	T	0.20
TT	48	51.2				
Control						
AA	0	5.5			A	0.74
AT	42	31	10.1	0.001	T	0.26
TT	38	43.5				
Visfatin -4689G/T						
AA						
GG	21	24.8			G	0.44
GT	47	39.5	2.889	0.08	T	0.56
TT	12	15.8				
Control						
GG	32	30			G	0.39
GT	34	38	0.876	0.349	T	0.61
TT	14	12				

AA: Adenin-Adenin, AT: Adenin-Thymine, TT: Thymine-Thymine, GG: Guanine-Guanine, GT: Guanine-Thymine
Data were analyzed by Chi-Square (χ2) test.
$p \leq 0.05$ is considered signifcant.

3.3. Genotype and allele frequency distributions

Table 3 displays the genotype and allele frequency distributions for the gene polymorphisms in the visfatin -4689G/T and vaspin genes. The vaspin genotype frequencies for the AA patient group were 43.2% for AT (32), and 55.8% for TT (48); in the control group, the rates were 56.8% for AT (42), and 44.2% for TT (38). There was no discernible difference in the distribution of vaspin genotypes across the groups ($\chi2 = 2.51$ df = 1 p = 0.11). According to the results, it was shown that TT genotype did not increase AA risk [OR (95% CI) = 1.65 (0.88-3.10), p = 0.11], although vaspin TT genotype was higher in AA patients. In controls, the frequencies for A and T alleles were 56.8% and 48%, and in AA patients 43.2% and 52% respectively. The difference in the allele frequency between the AA and controls was found to be non-significant ($\chi2 = 1.75$, df = 1, p = 0.18).

Table 3. Distribution of genotypes and allele frequencies of vaspin and visfatin -4689G/T gene polymorphisms in AA patients and control groups

Polymorphic sites		Control		AA		OR	(95%CI)	p
		n = 80	%	n = 80	%			
Vaspin	**Genotypes**							
	AA	0	0	0	0	-	-	-
	AT	42	56.8	32	43.2	1	-	-
	TT	38	44.2	48	55.8	1.65	0.88-3.10	0.11
	$\chi 2 = 2.51$, df = 1, p = 0.11							
	Allele							
	A	42	56.8	32	43.2	1	-	-
	T	118	48.0	128	52.0	1.42	0.84-2.40	0.18
	$\chi 2 = 1.75$, df = 1, p = 0.18							
Visfatin -4689G/T	Genotype							
	GG	32	60.4	21	39.6	1	-	-
	GT	34	42.0	47	58.0	2.11	1.04-4.27	*0.03
	TT	14	53.8	12	46.2	1.31	0.51-3.37	0.58
	$\chi 2 = 4.52$, df = 2, p = 0.10							
	Allele							
	G	98	52.4	89	47.6	1	-	-
	T	62	46.6	71	53.4	1.26	0.80-1.96	0.3
	$\chi 2 = 1.04$, df = 1, p = 0.30							

AA: Alopecia Areata, OR: Odds ratio, CI: Confidence interval, AA: Adenin-Adenin, AT: Adenin-Thymine, TT: Thymine-Thymine, GG: Guanine-Guanine, GT: Guanine-Thymine. Data were analyzed by Chi-Square ($\chi 2$) test. $p \leq 0.05$ is considered signifcant

The visfatin -4689G/T gene polymorphism genotype frequencies did not differ significantly ($\chi 2 = 4.52$, df = 2, p = 0.10). The frequencies of the GG, GT, and TT genotypes in the AA group were 39.6%, 58.0%, and 46.2%, respectively, whereas they were 60.4%, 42.0%, and 53.8%, respectively, in the control group. The risk of developing AA is 2.11 times higher in people with the GT genotype than in people with the GG genotype [OR (95% CI) = 2.11 (1.04-4.27), p = 0.03]. The visfatin -4689G/T gene polymorphism allele frequencies did not differ statistically significantly between the AA and control groups ($\chi 2 = 1.04$, df = 1, p = 0.30).

3.4. The frequencies of vaspin and visfatin -4689G/T genotypes and clinical characteristics in Alopecia Areata

The frequencies and clinical features of vaspin and visfatin -4689G/T genotypes genotypes in AA patients are shown in Table 4 and Table 5. Age, gender, duration of the disease, alopecia severity and localization, nail dystrophy, and localization, family history, other skin disease and the presence of another autoimmune disease were analyzed. There was no statistically significant relationship between vaspin and visfatin -4689G/T genotypes with clinical and demographic parameters of AA patients and in our investigation.

Table 4. Clinical characteristics of the study population according to genotypes of the vaspin rs2236242 gene

Visfatin -4689G/T genotypes				
	GG	**GT**	**TT**	**p**
Age (years)	30.3 ± 10.6	31.1 ± 9.28	35.5 ± 13.7	0.346[a]
Gender n (%)				
Male	12 (57.1)	24 (51.1)	8 (66.7)	0.609[b]
Female	9 (42.9)	23 (48.9)	4 (33.3)	
Disease duration (months)	5.52 ± 10.4	15.5 ± 27.5	5.0 ± 5.17	0.132[a]
Alopecia severity n (%)				
< 25%	17 (81.0)	37 (78.7)	10 (83.3)	
25-50%	4 (19.0)	10 (21.3)	2 (16.7)	0.931[b]
> 50%	-	-	-	
Alopecia localization (n %)				
Scalp	13 (61.9)	34 (72.3)	8 (66.7)	
Beard/Mustache	6 (28.6)	7 (14.6)	2 (16.7)	0.729[b]
Hair/Beard	2 (9.5)	6 (12.8)	2 (16.7)	
Body	-	-	-	
Nail dystrophy (n%)				
Yes	2 (9.5)	8 (17.0)	1 (8.3)	
No	19 (90.5)	39 (83.0)	11 (91.7)	0.595[b]
Family history (n%)				
Yes	2 (9.5)	9 (19.1)	3 (25.0)	
No	19 (90.5)	38 (80.9)	9 (75.0)	0.477[b]
Other skin disease (n%)				
Yes	2 (9.5)	8 (17.0)	1 (8.3)	
No	19 (90.5)	39 (83.0)	11 (91.7)	0.595[b]
Other autoimmune disease (n%)				
Yes	2 (9.5)	2 (4.3)	1 (8.3)	
No	19 (90.5)	45 (95.7)	11 (91.7)	0.673[b]

[a]*Independent Student t-test,* [b]*Chi-square (χ2) test, p ≤ 0.05 is considered significant*

Table 5. Clinical characteristics of the study population according to genotypes of the visfatin -4689G/T gene

Visfatin -4689G/T genotypes	GG	GT	TT	p
Age (years)	30.3 ± 10.6	31.1 ± 9.28	35.5 ± 13.7	0.346[a]
Gender n (%)				
Male	12 (57.1)	24 (51.1)	8 (66.7)	0.609[b]
Female	9 (42.9)	23 (48.9)	4 (33.3)	
Disease duration (months)	5.52 ± 10.4	15.5 ± 27.5	5.0 ± 5.17	0.132[a]
Alopecia severity n (%)				
< 25%	17 (81.0)	37 (78.7)	10 (83.3)	
25-50%	4 (19.0)	10 (21.3)	2 (16.7)	0.931[b]
> 50%	-	-	-	
Alopecia localization (n %)				
Scalp	13 (61.9)	34 (72.3)	8 (66.7)	
Beard/Mustache	6 (28.6)	7 (14.6)	2 (16.7)	0.729[b]
Hair/Beard	2 (9.5)	6 (12.8)	2 (16.7)	
Body	-	-	-	
Nail dystrophy (n%)				
Yes	2 (9.5)	8 (17.0)	1 (8.3)	
No	19 (90.5)	39 (83.0)	11 (91.7)	0.595[b]
Family history (n%)				
Yes	2 (9.5)	9 (19.1)	3 (25.0)	
No	19 (90.5)	38 (80.9)	9 (75.0)	0.477[b]
Other skin disease (n%)				
Yes	2 (9.5)	8 (17.0)	1 (8.3)	
No	19 (90.5)	39 (83.0)	11 (91.7)	0.595[b]
Other autoimmune disease (n%)				
Yes	2 (9.5)	2 (4.3)	1 (8.3)	0.673[b]
No	19 (90.5)	45 (95.7)	11 (91.7)	

[a]*ANOVA,* [b]*Chi-square (χ2) test, p ≤ 0.05 is considered signifcant*

4. Discussion

In this study, the possible effects of polymorphisms in the vaspin and visfatin -4689G/T genes on AA patients and their relationship with demographic and clinical data were investigated. To the best of our knowledge, this is the first investigation into the connection between AA and the vaspin and visfatin -4689G/T gene polymorphisms. Therefore, it is impossible to compare the findings of this study to those of others.

The genotypes of vaspin do not differ statistically significantly amidst AA patients and controls, according to our data. In spite of this, we found that AA patients had a higher frequency of the vaspin TT genotype than the control group, and that this genotype was linked to a 1.65-fold increased risk of AA disease in comparison to the AT genotype. But it was not statistically significant. To the best of our knowledge, there is no information about this polymorphism in AA patients. Therefore, we cannot compare our results.

Although several studies have established a link between the vaspin gene polymorphism

and metabolic syndrome, coronary artery disease, diabetes, and obesity, the exact mechanism is still unknown. A Polish study examined the relationship between the polymorphism of the vaspin gene and the risk of developing metabolic syndrome. The vaspin polymorphism was not significantly linked to metabolic syndrome, according to the study's findings; however, a meta-analysis of genotype dispersion in Polish, Iranian, and Egyptian populations showed that patients with metabolic syndrome were more likely than controls to have the TT genotype (22). In another investigation, when diabetic and non-diabetic obese people were compared for vaspin gene polymorphism, the TT genotype was found to be statistically significant (23). Similarly, in our study, although the vaspin TT genotype was high in the patient group, it was not statistically significant.

In contrast to our investigation, a study conducted within the Turkish population demonstrated that the vaspin AT genotype exhibited an elevated risk of psoriasis when compared to the TT genotype (16). We believe that the disparities in study outcomes are related to differences in ethnic origin and sample size.

Visfatin is a protein secreted by adipose tissue that may mediate pro-inflammatory properties. It has been associated with some diseases such as type 2 diabetes, non-alcoholic fatty liver disease cardiovascular disease and obesity (24). However, there are a limited number of studies on the visfatin -4689G/T gene polymorphism in the literature, and none on the link amidst this gene polymorphism and AA. Our findings reveal that there is no statistically significant difference between AA patients and controls in visfatin -4689G/T genotypes. Although we obtained a significant difference when comparing the GT genotype to other genotypes, it was not statistically significant when viewed as a whole. However, we observed that the risk of developing AA in people with the GT genotype was 2.11 times higher than in people with the GG genotype. In obese adults, the effect of visfatin genotypes and dietary fat intake on bone mineral density (BMD) was investigated. The results indicated that the TT genotype was associated with considerably greater T score and lumbar BMD, whereas the GT genotype was associated with higher hip BMD. The frequency of TT, GT, and GG genotypes was 17.54%, 48.51%, and 33.92%, respectively, in the same study (25). Similarly, in our investigation, we discovered that the GT genotype frequency was greater than in controls, therefore our findings overlap with those of the current study. Another study examined the genetic associations of visfatin polymorphisms with epidermal growth factor receptor (EGFR) status and clinicopathological features in lung adenocarcinoma. Similar to our investigation, it was concluded that the visfatin gene polymorphism and clinical features of the disease were not statistically significant (26). A different study looked at the relationship between a cohort of Taiwanese men's risk of developing oral squamous cell carcinoma and four distinct variants of the visfatin gene polymorphism and carcinogenic lifestyle variables. The visfatin -4689G/T gene polymorphism was not found to be associated with oral squamous cell cancer (27). A study on the influence of visfatin gene polymorphism on glucose homeostasis discovered that blood sugar control status was not connected with visfatin gene polymorphism. The frequency of genotypes with polymorphisms investigated in the study for TT, GG, and GT were 18.3%, 50.5%, and 31.2%, respectively. According to genotypic evaluations, fasting insulin levels were high in patients with GT genotype. It was concluded that patients with the GT genotype needed a lower insulin dose to control blood sugar (28).

More than just storing energy, adipose tissue is a dynamic endocrine tissue that handles various tasks. It contributes to the synthesis of several bioactive substances known as adipocytokines, which control metabolic processes. These adipocytokines have been linked to pro-inflammatory and autoimmune processes, as well as metabolic diseases such obesity and insulin resistance (29). Visfatin, a novel adipocytokine, possesses pro-inflammatory properties (30). According to recent research, inflammatory diseases such as cutaneous T-cell lymphoma, atopic dermatitis, psoriasis and Behcet's illness are associated

with elevated serum visfatin levels (31). In a study, serum visfatin levels and their relationship with disease severity were examined in psoriasis patients, and it was reported that psoriasis patients showed significantly higher visfatin levels than controls (32). On the contrary, in a different investigation of psoriasis patients, no significant change in plasma visfatin levels was identified between patients and controls (33). In a case-control study investigating the evaluation of ischemia-modified albumin level and metabolic profile in AA patients, visfatin levels did not significantly change across the groups (31). Although visfatin has a possible role in the pathogenesis of metabolic diseases, a study conducted on male patients with androgenetic alopecia did not report any difference between visfatin serum concentrations in patients and healthy controls (34).

5. Conclusion

This study shows that there is no relationship between vaspin and visfatin -4689G/T polymorphism and AA in the Turkish population. However, the TT genotype for the vaspin gene and the GT genotype for the visfatin -4689G/T gene are risk factors for people with AA disease. Since there is no research on the relationship between AA and vaspin and visfatin -4689G/T gene polymorphisms, we cannot compare our findings with any published studies. Therefore, it is expected that our research will serve as a source for future research. This increases the originality of our study and the additional studies we plan to do on this subject.

Inconsistencies across research suggest that genetic diversity in the population may need to be investigated. The very small sample size of our single-center study is its primary drawback. Considering the fact that the genetic polymorphism of the visfatin gene has not been evaluated in the Turkish population, future studies should include genotyping the vaspin and visfatin genes to understand the relationship between visfatin and AA.

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Ethics

Ethics Committee Approval: The study was approved by Kütahya Health Sciences University Noninterventional Clinical Research Ethical Committee (Decision no: 2022/09-28, Date: 14. 09.2022).
Informed Consent: The authors declared they get consent from the patients.
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