Medical Science Journal for Advance Research

Vol. 6 , No. 2 , June 2025
e-ISSN: 2774-4892

Diagnostic Significance of MIG (Monokine Induced by Gamma Interferon) and IP-10 (Interferon gamma-induced protein 10) Chemokines in Alopecia Areata

Zinah Abbass Ali[1], Khawla A. Shemran[2], and Hiba Resheed Behayaa[3]
[1,2,3]*Department of Biochemistry, College of Medicine, University of Babylon, Babylon, Iraq.*
Email: Zenaabbass1980@gmail.com, shemrankhawla@uobabylon.edu.iq, hiba_resheed80@yahoo.com

ABSTRACT

Background: *The condition of Alopecia Areata (AA) is a type of autoimmune disease that results in the loss of hair in patches. Some immunologically active chemokines, such as MIG (CXCL9) and IP-10 (CXCL10), which are stimulated by interferon-gamma, are believed to be involved in the inflammatory processes of AA, and may contribute to its development.* ***Objectives:*** *To measure the serum levels of MIG and IP-10 in patients with Alopecia Areata and evaluate those levels for possible differential diagnostic significance.* ***Methods:*** *The study consisted of 60 patients with AA, alongside 60 age-matched healthy controls. The participants were sourced from dermatology clinics where they were evaluated clinically. Patients with complex systemic autoimmune diseases, active infections, or those who had recently undergone immunosuppressive treatments were not eligible for the study. Serum levels of MIG and IP-10 were quantified using the ELISA technique. Correlation studies as well as ROC curve analysis were performed for the statistical evaluation.* ***Results:*** *In comparison to controls, AA patients had a mean serum MIG level that was significantly higher at (864.13 ± 201.08 pg/ml) while controls had 299.04 ± 130.4 pg/ml ($p<0.01$). Moreover, AA patients also had elevated IP-10 levels 504.61 ± 122.2 pg/ml compared to controls 297.9 ± 77.3 pg/ml ($p<0.01$). Within patients, Pearson correlation showed moderate-to-strong correlation of MIG with IP-10, BMI, and age. In ROC curve analysis, MIG had an AUC of 0.972 (sensitivity 80%, specificity 87%) whereas IP-10 had an AUC of 0.89 (sensitivity 62%, specificity 76%).* ***Conclusion:*** *AA patients showed significantly elevated levels of MIG and IP-10 indicating possible involvement in pathogenesis. Particularly, MIG showed strong diagnostic capability. These findings suggest that these chemokines may be potential biomarkers for the early diagnosis and follow-up of AA.*

Keywords: Alopecia Areata, MIG, CXCL9, IP-10, CXCL10, Biomarkers, Chemokines, ELISA.

Article Information
Received: Received: April 29, 2025; Revised: May 30, 2025; Online: June, 2025

INTRODUCTION

Alopecia Areata (AA) is a long-lasting disorder in which the body's immune system mistakenly attacks hair roots, leaving patchy, non-scarring bald spots on scalp, beard or limbs. About 2 percent of the global population falls ill, most often children or young adults, and neither sex nor ethnic background seems to matter [1]. Although finer details remain under debate, growing evidence suggests that self-destructive cytotoxic T cells cluster around follicles in the antigen stage, interrupting growth, cycling, and pigmenting [2]. An important hallmark of each AA plaque is its unusual set of chemokines-small, structurally related messengers that lure white blood cells to trouble spots. Interferon-? (IFN-?) stands out as a prime mover, sparking and extending the immune attack on scalp and skin [3]. Two IFN- driven chemokines-Monokine induced by gamma interferon (MIG or CXCL9) and interferon-?-induced protein 10 (IP-10 or CXCL10)-also appear in type 1 diabetes, systemic lupus erythematosus, and vitiligo [4-6]. Both MIG and IP-10 signal mainly through the CXCR3 receptor, directing activated

https://doi.org/10.46966/msjar.v6i2.278

Th1 lymphocytes to inflamed tissues like hair follicles in alopecia Areata [7]. Although these chemokines rise sharply in other autoimmune skin disorders, their specific role in the pathophysiology of alopecia Areata remains inadequately studied [8]. The current study therefore quantifies serum MIG and IP-10 in alopecia Areata patients compared with healthy volunteers, evaluating their potential utility as diagnostic biomarkers for the disease.

MATERIALS AND METHODS

Study Design and Participants

This case-control study aimed to quantify serum levels of Monokine Induced by Gamma Interferon (MIG/CXCL9) and Interferon-gamma-induced protein 10 (IP-10/CXCL10) in patients with alopecia Areata and then compare these findings against concentrations measured in healthy volunteers. The analysis recruited 120 individuals, consisting of 60 dermatologically confirmed alopecia sufferers and 60 age- and sex-matched controls free of scalp or immunological disorders [9].

Inclusion and Exclusion Criteria

Eligible participants, aged 27-37 years, were drawn from dermatology outpatient clinics at neighboring hospitals. Each case was diagnosed as alopecia Areata following detailed clinical assessment by senior dermatologists. Patients with systemic autoimmune disease, active infection, endocrine or renal dysfunction, or who had used corticosteroids or other immunosuppressant's in the preceding six months were excluded [10]. Control volunteers presented with neither autoimmune illness nor inflammatory scalp changes.

Ethical Approval and Informed Consent

The research protocol received approval from the Institutional Ethics Committee of the College of Medicine, University of Babylon. Informed written consent, obtained in accordance with the Declaration of Helsinki, was collected from each volunteer before any study procedure was undertaken [11].

Sample Collection and Processing

Under strict sterile techniques, 5 mL of blood was collected from each volunteer, processed without delay in the lab, and frozen at -80C for later analysis. Samples were left to clot at room temperature, and clear serum was then separated by centrifugation at 3000 rpm for 10 minutes [12]. The resulting serum was portioned into small tubes and stored at -20°C to preserve cytokine integrity [13].

Biochemical Assay

Serum concentrations of MIG (CXCL9) and IP-10 (CXCL10) were measured using commercial enzyme-linked immunosorbent assay (ELISA) kits. MIG was analyzed with the Human CXCL9/MIG ELISA kit (Catalog No. E-EL-H6004, Elabscience®, Houston, TX, USA), whereas IP-10 was quantified with the Human CXCL10/IP-10 ELISA kit (Catalog No. E-EL-H0054, Elabscience®. All procedures were followed exactly as outlined by the supplier [14]. Absorbance was read at 450 nm with a BioTek microplate reader (USA), and cytokine levels were calculated from standard curves [15].

Statistical Analysis

Data were analyzed using GraphPad prism version 8. Continuous variables are expressed as mean ± standard deviation (SD). Differences between groups were assessed by the independent-samples t-test.

Pearson correlation coefficients were calculated to examine the linear relationships among the continuous variables. The diagnostic utility of each measure was investigated through receiver-operating-characteristic (ROC) curve analysis that computes the area under the curve (AUC) alongside sensitivity, specificity, and the optimal cut-off point. Statistical significance was defined as a two-tailed p-value less than 0.05 [16, 17].

RESULTS

Participant Demographics

Sixty people living with Alopecia Areata and sixty healthy volunteers matched for age were enrolled. Age and body-mass index did not differ meaningfully between the two groups. Patients averaged 31.72 3.09 years, while controls were 32.7 2.93 years (p 0.064). Mean BMI was 25.49 2.17 kg/m2 in the AA cohort and 25.48 2.45 kg/m2 in controls (p 0.098), confirming similar baseline characteristics.

Chemokine Levels: MIG and IP-10

Serum concentrations of MIG (CXCL9) and IP-10 (CXCL10) were markedly higher in patients than in healthy volunteers. The average MIG level in the alopecia group was 864.13 201.08 pg/mL versus 299.04 130.4 pg/mL in controls ($p < 0.01$). Likewise, mean IP-10 was 504.61 122.2 pg/mL in patients compared to 297.9 77.3 pg/mL in controls ($p < 0.01$).

Correlation Analysis

In the clinical group, Pearson correlation revealed a strong, positive link between MIG and IP-10 ($r = 0.80$, $p = 0.03$). MIG also showed moderate relationships with age ($r = 0.40$, $p = 0.07$) and body mass index (BMI; $r = 0.50$, $p = 0.04$). Likewise, IP-10 correlated with BMI ($r = 0.80$, $p = 0.09$) and age ($r = 0.50$, $p = 0.06$), suggesting that these chemokines track important demographic variables.

Diagnostic Value of MIG and IP-10

Receiver-operating-characteristic (ROC) analysis indicated that MIG distinguished alopecia areata (AA) patients from controls with an area under the curve (AUC) of 0.972 (95% confidence interval [CI] 0.94-0.99), achieving 80% sensitivity and 87% specificity at a cut-off of 535.7 pg/mL. IP-10 also performed well, producing an AUC of 0.89 (95% CI 0.83-0.95) with 62% sensitivity and 76% specificity at 396.9 pg/mL.

Table 1: Mean ages of Alopecia Areata patients and healthy controls. Age distributions between groups were similar ($p = 0.064$).

Study variable	Groups	N	Mean ± SD	p-value
Age	Patient	60	31.72 ± 3.09 (27-37)	0.064
	Control	60	32.7±2.93 (28-37)	

Table 2: Body mass indices of Alopecia Areata patients and healthy controls. BMIs did not differ between cohorts ($p = 0.098$).

Study variable	Groups	N	Mean ± SD	p-value
BMI	Patient	60	25.49 ± 2.17 (22.09-29.7)	0.098
	Control	60	25.48± 2.45 (21.04-29.7)	

Table 3: Serum MIG and IP-10 levels in patients and in healthy controls. Both chemokines were markedly elevated in patients compared with controls ($p < 0.01$ for each).

Variable	N	Patients (Mean ± SD)	Control (Mean ± SD)	P-value
MIG (Pg/ml)	60	864.13 ± 201.08 (640.1-1390.1)	299.04 ± 130.4 (175.4-570.09)	<0.01
IP-10 (Pg/ml)	60	504.61±122.2 (234.3-690.6)	297.9 ± 77.3 (210.3-420.9)	<0.01

Description of Figures

ROC curve: (Figure 1,2) shows a Receiver Operating Characteristic (ROC) curve that determines how well serum MIG (CXCL9) and IP-10 separate people with alopecia areata (AA) from healthy subjects. Sensitivity appears on the vertical axis, given as a percentage, while specificity is plotted along the horizontal axis in the same unit. The black dots trace MIGs and IP-10 diagnostic performance at several analytical thresholds. The red dashed line marks no discrimination guessing, a baseline that corresponds to an area under the curve (AUC) of 0.5. Because the curve climbs sharply and hugs the upper left corner, it proves that serum MIG and IP-10 provide super group separation. Taken together, these observations imply that MIG and IP-10 could serve as a valuable biomarker, with the AUC estimated at nearly 0.97 and 0.89, respectively.

Q-Q plot: (Figure 1,2) shows a normal quantile-quantile (Q-Q) plot that compares measured serum MIG and IP-10 levels with those expected from a Gaussian distribution. Observed concentrations appear along the horizontal x-axis, while the vertical y-axis shows the theoretical normal values for the same quantiles. Control participants are displayed as blue dots and AA patients as red dots. Points lying close to the black diagonal conform to normality, whereas systematic deviation from that line suggests potential distribution differences between groups. Most blue dots sit snug against the baseline, but the red dots from the patient group float noticeably above it, hinting at higher and perhaps non-standard MIG levels in people with the disease. Taken together, these trends confirm that MIG amounts in patients stray outside the healthy band, mirroring spikes usually tied to inflammation. The Q-Q plot suggests that IP-10 levels are significantly elevated and non-normal distribution in the patient group, due to outliers or many red dots deviate upward in the patient group.

Box plot: The box plot presents all these results by laying the serum MIG levels measured in controls and those found in alopecia areata volunteers. Picograms per milliliter make up the vertical scale of the graph. An opaque box chips away at the bottom and top quarters of the data, while the line that cuts through it marks the median. Thin vertical whiskers then stretch outward to the smallest and largest values, consciously leaving eccentric dots off the end. Control samples settle into a lower median and a tighter cloud, roughly between 200 and 400 pg/mL. Patients, by contrast, cluster around a median close to 900 pg/mL and spread wider still, reaching roughly 1400 pg/mL. Taken together, these pairs of results tell us that people with alopecia areata carry significantly higher MIG, and they show the level bouncing much more between individuals.

Bar Plot: This bar plot displays shows, for both healthy controls and alopecia areata (AA) patients, average serum IP-10 levels with standard deviations. Given in picograms per milliliter (pg/mL), values show up on the

vertical axis. The brown bar for the AA group reaches about 505 pg/mL; the green bar for controls rests close to 298 pg/mL.

From each bar, error bars show the standard deviation's data capture spread. Above the bars asterisks (****) indicate that the difference reaches statistical significance at p 0.0001. These results taken together show significantly greater IP-10 in AA patients, therefore highlighting the molecules likely involved in disease processes and their use as a diagnostic marker.

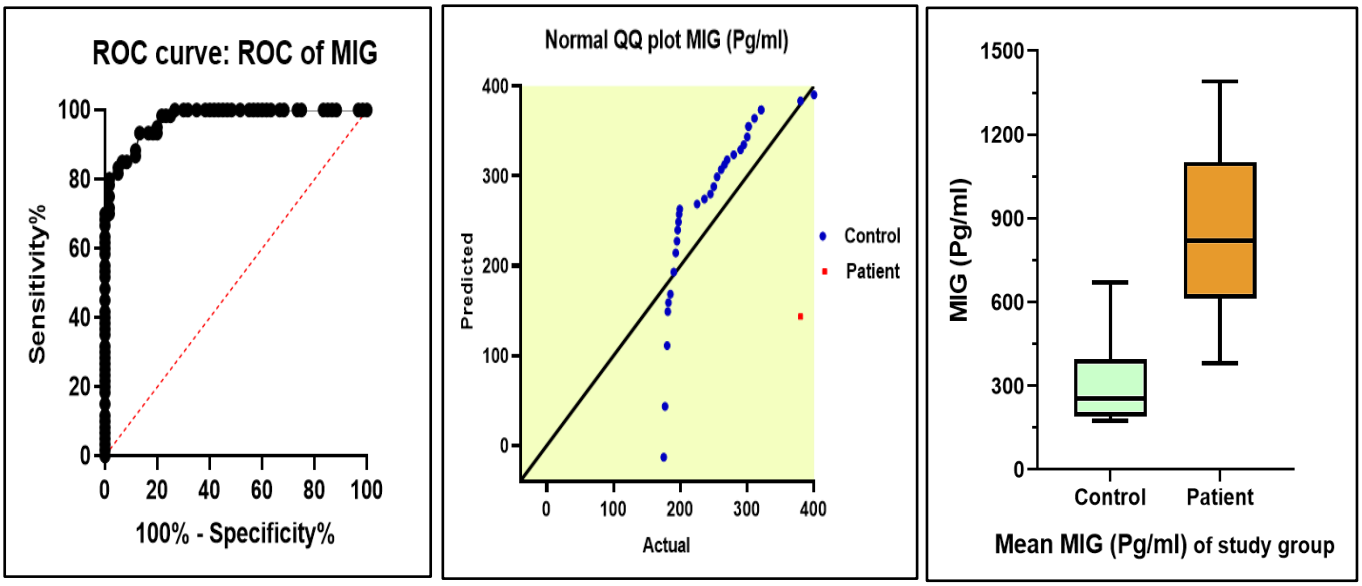

Figure 1 Explain the diagnostic value of MIG (CXCL9) levels for both the alopecia areata and the control cohort. The companion receiver-operating-characteristic curve confirms outstanding diagnostic accuracy, with an area under the curve approaching 0.97. Quantile-quantile plots also show that patient MIG values differ significantly from normality, supporting the idea of heightened production. Box-plot tests reveal that serum concentrations in alopecia areata subjects exceed those in controls by a wide range ($p < 0.01$).

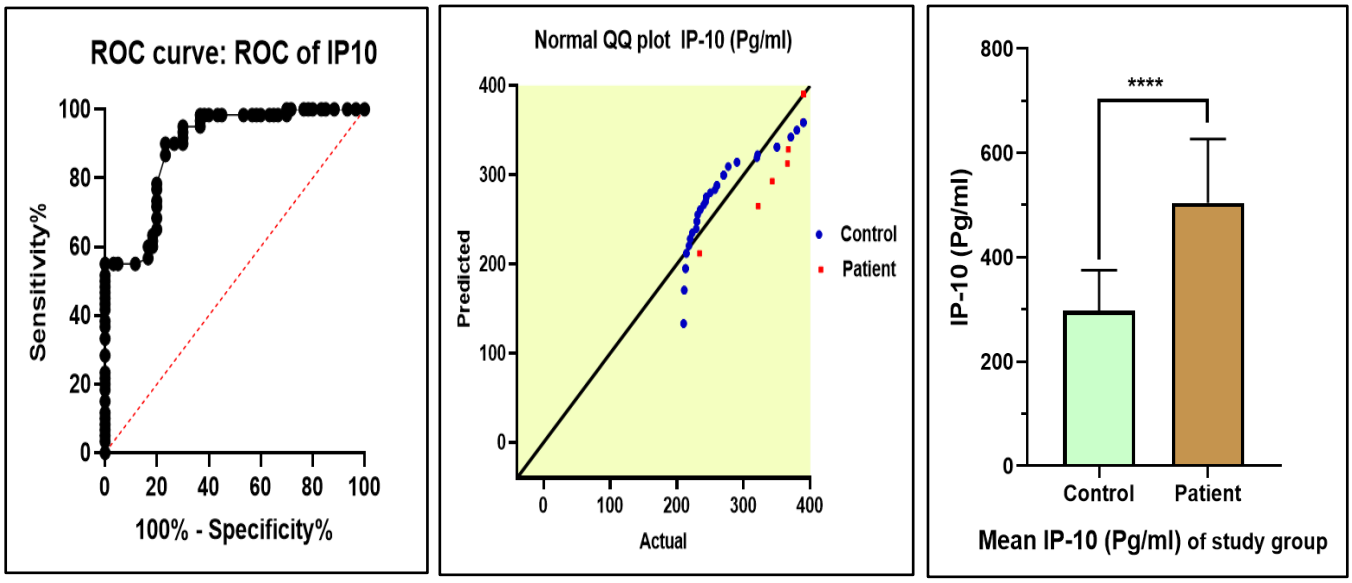

Figure 2. Explain the diagnostic value of Serum IP-10 levels for both the alopecia areata and the control cohort. The companion ROC curve confirms outstanding diagnostic accuracy, with an area under the curve approaching 0.89. Q-Q plots also show that patient IP-10 values differ significantly from control. Bar-plot tests reveal that serum IP-10 was significantly higher in patients than in controls ($p < 0.01$).

Table 4: Pearson correlations among MIG, IP-10, age, and BMI in the patient cohort. Significant associations were observed, particularly between MIG and IP-10 and between MIG and BMI.

Study variables		MIG	IP-10	Age	BMI
MIG	R	----	0.8	0.4	0.5
	p-value	----	0.03	0.07	0.04
IP-10	R	0.6	----	0.5	0.8
	p-value	0.06	----	0.06	0.09
Age	R	0.7	0.89	----	0.9
	p-value	0.05	0.07	----	0.07
BMI	R	0.8	0.56	0.4	----
	p-value	0.06	0.08	0.045	----

Table 5: Receiver-operator-characteristic results for MIG and IP-10 in Alopecia Areata diagnosis. MIG outperformed IP-10 as a diagnostic marker (area under the curve 0.972 versus 0.890).

Parameters	AUC	p-value	Sensitivity	Specificity	Cut off value	Asymptotic 95% Confidence Interval	
						Lower Bound	Upper Bound
MIG pg/ml	0.972	0.00	80%	87%	535.7	0.94	0.99
IP-10 pg/ml	0.89	0.00	62%	76%	396.9	0.83	0.947

DISCUSSION

In this study we measured serum levels of MIG (CXCL9) and IP-10 (CXCL10), two chemokines produced after interferon-gamma exposure, and found both markers to be significantly elevated in people with alopecia areata compared to age-matched healthy controls. These findings support the view that these molecules are involved in the immune-mediated hair loss of the disease and point to their potential use as blood-based biomarkers for diagnosing alopecia areata and monitoring its activity. Researchers propose that alopecia areata occurs when the immune system wrongly targets hair follicles, particularly those in the antigen growth phase, the only stage in which follicles maintain a degree of immune privilege that is lost during disease onset [18,19]. Once that privilege disappears, effector T cells and other inflammatory mediators begin to infiltrate, a process heavily driven by interferon-gamma signaling [20]. MIG and IP-10, principal downstream products of interferon-gamma, attract CXCR3-expressing Th1 cells and other lymphocytes to sites of inflammation, including the follicular microenvironment [21,22]. The increased serum concentrations of both chemokines in our alopecia areata patients thus reinforce this inflammatory model and hint at a pathway for future targeted treatments. Similar trends arise in other autoimmune diseases where MIG and IP-10 seem to fuel damage, such as vitiligo, systemic lupus erythematosus, and type 1 diabetes [23-25]. A transcriptomic survey led by Suarez-Farinas and colleagues likewise singled out these chemokines as central to a

robust interferon signature in lesion scalp skin from alopecia areata patients, lending further weight to their putative pathogenic role [26]. The current study went on to test MIG and IP-10 as potential clinical biomarkers. Receiver operating characteristic plots showed that MIG surpassed IP-10, yielding an area under the curve of 0.972 versus 0.89 for the latter. The greater sensitivity and specificity tied to MIG consequently hint that it might provide a more reliable signal for early diagnosis or for monitoring shifts in disease activity. These findings echo earlier studies that nominated CXCL9the MIG ligands a marker of severity in several autoimmune and inflammatory conditions [27, 28].

Pearson correlation analysis showed a moderate to strong positive link between MIG and IP-10 levels in alopecia areata patients, mirroring a coordinated chemokine response likely driven by interferon-gamma. Even though the linked patterns with age and body mass index are modest, they suggest systemic factors could govern chemokine expression; this idea should be tested in future longitudinal work [29]. Both Q-Q and box plots clearly departed from normality and showed markedly higher levels of the two chemokines in patients, proving their ability to set sick people apart from healthy controls. MIG had the widest dynamic range and best separation yet IP-10 was still significantly raised, showing it is an important partner marker. These reassuring results must be viewed alongside several caveats. First, although the present sample is large enough for a pilot project, its modest size still limits generalization. Second, because the study is cross-sectional, we cannot see whether MIG or IP-10 rises or falls with treatment or during spontaneous remission. Larger longitudinal trials are therefore needed to test whether these molecules can become prognostic or therapy-response tests in Alopecia Areata (AA). In brief, both MIG and IP-10 are elevated in AA, linking them to the disorder's immune imbalance. MIG alone appears strongest as a possible diagnostic test. Taken together, the two chemokines may also open fresh paths for treating interferon-gamma-driven autoimmune hair loss.

CONCLUSION

The present investigation reveals that serum levels of MIG (CXCL9) and IP-10 (CXCL10) are markedly increased in individuals with alopecia areata, lending credence to their participation in the interferon-gamma-mediated immune network believed to underlie disease onset. Of the two molecules, MIG exhibits superior diagnostic performance and correlates more consistently with clinical criteria, indicating its potential role as a sensitive biomarker for early detection. IP-10 is also elevated but appears to function in a supportive capacity. Collectively, these chemokines offer a non-invasive means of diagnosis and may become pharmacological targets for future treatment. Additional research is required to track their serum fluctuations over the course of illness and in response to therapy.

ACKNOWLEDGEMENTS

All patients included in this study are gratefully appreciated for their participation. We also acknowledge the help from the technical staff of the Biochemistry department of University of Babylon.

FINANCIAL SUPPORT

Support Received from Individual Funded Sources

ETHICAL CONSIDERATIONS

The study protocol received ethical approval from the Institutional Review Board of the University of Babylon. All procedures were in accordance with the ethical principles of the Declaration of Helsinki. All participants were provided with detailed participant information sheets and gave their written informed consent prior to participation.

CONFLICTS OF INTEREST

No competing interests have been declared by the authors for this manuscript from other sources, financial or otherwise.

REFERENCES

1. Wasserman D, Guzman-Sanchez DA, Scott K, McMichael A. Alopecia areata. *J Am Acad Dermatol.* 2007;57(4):S1–S14.
2. Gilhar A, Etzioni A, Paus R. Alopecia areata. *N Engl J Med.* 2012;366(16):1515–25.
3. Ito T, Hashizume H. Interferon-gamma: The key cytokine in the pathogenesis of alopecia areata. *Autoimmun Rev.* 2011;10(8):495–8.
4. Liu Y, Mei C, Chen L, Chen J. CXCL9 and CXCL10 in autoimmune diseases. *Autoimmun Rev.* 2011;10(7):721–8.
5. Lo BK, Shi VY, et al. CXCL10 and CXCL9 expression in lesional skin of vitiligo patients. *Arch Dermatol Res.* 2013;305(9):785–92.
6. Groom JR, Luster AD. CXCR3 ligands: redundant, collaborative and antagonistic functions. *Immunol Cell Biol.* 2011;89(2):207–15.
7. Sallusto F, Mackay CR, Lanzavecchia A. The role of chemokine receptors in primary, effector, and memory immune responses. *Annu Rev Immunol.* 2000;18:593–620.
8. Suarez-Farinas M, Tintle SJ, Shemer A, Chiricozzi A, Nograles K, Cardinale I, et al. Alopecia areata transcriptome profiling shows IFN signature. *J Clin Invest.* 2013;123(11):4609–19.
9. Fletcher RH, Fletcher SW, Fletcher GS. *Clinical Epidemiology: The Essentials.* 5th ed. Philadelphia: Lippincott Williams & Wilkins; 2012.
10. Gilhar A, Kalish RS. Alopecia areata: a tissue-specific autoimmune disease of the hair follicle. *Autoimmun Rev.* 2006;5(1):64–9.
11. World Medical Association. World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects. *JAMA.* 2013;310(20):2191–4.
12. Tietz NW. *Fundamentals of Clinical Chemistry.* 6th ed. Philadelphia: Saunders Elsevier; 2008.
13. Boyum A. Isolation of mononuclear cells and granulocytes from human blood. *Scand J Clin Lab Invest Suppl.* 1968;97:77–89.
14. Elabscience Biotechnology Inc. [Product Datasheet: Human CXCL9 and CXCL10 ELISA Kits]. Houston, TX: Elabscience; 2023.
15. Crowther JR. *The ELISA Guidebook.* 2nd ed. New York: Humana Press; 2009.
16. Field A. *Discovering Statistics Using IBM SPSS Statistics.* 5th ed. London: Sage Publications; 2018.
17. Hajian-Tilaki K. Receiver Operating Characteristic (ROC) Curve Analysis for Medical Diagnostic Test Evaluation. *Caspian J Intern Med.* 2013;4(2):627–35.
18. Zlotogorski A, Molho-Pessach V, Horev L. The genetics of alopecia areata. *Clin Dermatol.* 2013;31(2):180–6.
19. Bertolini M, et al. Hair follicle immune privilege and its collapse in alopecia areata. *Exp Dermatol.* 2020;29(7):703–12.
20. Yu X, et al. IFN-γ–dependent recruitment of CXCR3+ T cells mediates dermal inflammation in autoimmune disease. *J Invest Dermatol.* 2015;135(3):726–35.
21. Antonelli A, Ferrari SM, et al. Role of CXCL10 and its receptor CXCR3 in autoimmune diseases. *Autoimmun Rev.* 2014;13(3):272–80.

22. Yang D, et al. Chemokines in autoimmune diseases: Pathogenic and therapeutic implications. *Cytokine Growth Factor Rev.* 2022;64:33–45.

23. Lo BK, et al. Expression of MIG and IP-10 in vitiligo skin: Implications for Th1-mediated recruitment. *Arch Dermatol Res.* 2007;299(6):301–6.

24. Afraz S, et al. Diagnostic value of serum IP-10 and MIG in children with autoimmune hepatitis. *BMC Gastroenterol.* 2020;20(1):113.

25. Bieganowska K, et al. Chemokine abnormalities in childhood type 1 diabetes: associations with autoimmunity and metabolic control. *Clin Immunol.* 2006;120(3):272–9.

26. Suarez-Farinas M, et al. Alopecia areata transcriptome reveals IFN-driven inflammatory profile. *J Clin Invest.* 2013;123(11):4609–19.

27. Lee AY. Role of CXCL9 and CXCL10 as markers of inflammation and disease activity in autoimmune disease. *Clin Chim Acta.* 2019;495:417–25.

28. Dufour JH, et al. CXCL9 induces Th1 cell recruitment and is a marker of disease activity. *J Immunol.* 2002;168(9):4508–15.

29. Kalra M, et al. Correlation of cytokines with obesity and metabolic profiles. *J Clin Endocrinol Metab.* 2016;101(10):3909–16.

