Journal of
Babol University of Medical Sciences
JBUMS
e-ISSN: 2251-7170 p-ISSN: 1561-4107

MicroRNA-1246 Gene Expression as a Biomarker in Patients with Systemic Lupus Erythematosus in Iraq

F. A. Shihab (PhD)[*1], R. M. Abed (PhD)[1]

1.Department of Biotechnology, College of Science, University of Baghdad, Baghdad, Iraq.

•Corresponding Author: F. A. Shihab (PhD)
Address: Department of Biotechnology, College of Science, University of Baghdad, Baghdad, Iraq.
Tel:-. E-mail: fatma.aamer1706b@sc.uobaghdad.edu.iq

Article Type

Research Paper

Received:
Feb 4th 2025
Revised:
Apr 15th 2025
Accepted:
Jul 28th 2025

ABSTRACT

Background and Objective: MicroRNAs are non-coding gene regulators that may serve as biomarkers for the diagnosis or prognosis of systemic lupus erythematosus (SLE). This study aimed to evaluate microRNA-1246 gene expression using quantitative real-time PCR (qRT-PCR).

Methods: This cross-sectional study was conducted on 100 women with systemic lupus erythematosus (SLE) and 100 healthy women serving as controls. Based on medication status, SLE patients were divided into two groups: the first group comprised 24 newly diagnosed patients who were not receiving any medication, while the second group included 76 patients who had been diagnosed with the disease for 6.23±0.58 years and were on active treatment. All patients were compared using laboratory tests including antinuclear antibodies (ANA), anti-double-stranded DNA antibodies (anti-dsDNA), complement 3 (C3), complement 4 (C4), C-reactive protein (CRP), erythrocyte sedimentation rate (ESR), hemoglobin, white blood cell count, and platelet count.

Findings: The mean age of women with SLE was 30.93±9.85 years, while that of the control group was 32.97±9.13 years. The mean body mass index (BMI) in SLE patients was 28.62±6.50 kg/m², which did not differ significantly from the control group (27.11±5.60 kg/m²; p=0.07). A positive family history was reported in 74% of SLE patients. The study results demonstrated a significant increase in MicroRNA-1246 expression, with a mean fold change of 2.31 compared to the control group (1.00-fold). No correlation was found between MicroRNA-1246 expression changes and any demographic or laboratory variables in the present study.

Conclusion: Based on the results of this research, microRNA-1246 may be considered a biomarker for susceptibility to SLE.

Keywords: *Autoimmune Disease, Gene Expression, Microrna, Systemic Lupus Erythematosus, Biomarker.*

Cite this article: Shihab FA, Abed RM. MicroRNA-1246 Gene Expression as a Biomarker in Patients with Systemic Lupus Erythematosus in Iraq. *Journal of Babol University of Medical Sciences*. 2026; 28: e58.

Copyright © 2026 Babol University of Medical Sciences. Published by Babol University of Medical Sciences. This work is licensed under a Creative Commons Attribution-NonCommercial 4.0 International license (https://creativecommons.org/licenses/by-nc/4.0/). Noncommercial uses of the work are permitted, provided the original work is properly cited.

Introduction

Systemic lupus erythematosus (SLE) is an autoimmune inflammatory disease with a wide spectrum of clinical manifestations, characterized by dysregulated cytokine activity, autoantibody production, dysregulated autoreactive T cell and B cell activation, and loss of tolerance to self-antigens (1). The principal feature of the disease is the production of various autoantibodies resulting from diminished immunological tolerance. These autoantibodies subsequently target self-antigens, including cellular components, immune complexes, DNA, and host nuclear antigens (2, 3). The clinical presentation of SLE varies considerably among individuals, and the disease tends to affect every organ and tissue in the body (4). Autoimmune diseases such as lupus and rheumatoid arthritis share similar symptoms, as both may attack the joints and kidneys, causing swelling, fever, and anemia, along with elevations in acute-phase reactants such as erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP) (5). Systemic lupus erythematosus may cause damage to several end organs, including the lungs, kidneys, neuropsychiatric systems, and cardiovascular system. The most common cause of death is cardiovascular complications. Multiple environmental, hormonal, and genetic factors seem to have a role in the development of SLE, but the specific cause of the illness is still unclear. Minority women of childbearing age account for the vast majority of SLE cases (65%) (6). The fundamental hallmark of SLE is sex bias, which affects one out of every nine males (7). Numerous variables, including epigenomics, inhibitory factors, ribosomes, and transposons, impact DNA transcription and translation, making it difficult to determine the precise mechanisms by which these processes occur (8). MicroRNAs, which are noncoding RNAs that are just 22 nucleotides long, regulate genes after transcription has occurred. They are involved in several biological processes, for example, differentiation, cell proliferation, metabolism, and death, by preventing mRNA translation. Recent studies have shown a connection between abnormal microRNA expression and the onset of many disorders, including tumors (9), inflammatory diseases (10, 11), and autoimmune diseases (10, 12). Human embryonic stem cells were the first target of a high-throughput sequencing effort that led to the discovery of microRNA-1246 (13). Subsequently, researchers located the miRNA-1246 gene on chromosome 2q31.1 and demonstrated that its expression was regulated by p53 (14). A study by Ren et al. (15) showed that miRNA-1246 expression was elevated in patients with multiple myeloma, independent of long noncoding RNA (lncRNA) high expression of HOX transcript antisense intergenic RNA (HOTAIR), miRNA-1246, chromosome 13 deletion, bone marrow plasma cell population, hemoglobin, myeloma protein, creatinine, albumin, calcium, serum levels of β2-microglobulin (β2-MG), age, sex, and disease stage, which may be linked to a poor prognosis in different tumor types. Gene expression patterns have been linked to a number of illnesses. Furthermore, the binding of target genes to miRNA-1246 and HOTAIR expression is essential for metabolic reprogramming in breast cancer (BC). This suggests that miRNA-1246 and HOTAIR hold unique potential for enhancing BC detection, although their expression patterns vary among BC patients (16). Evidence indicates that miRNA-1246 signaling affects the motility, invasion, metastasis, and proliferation of cancer cells (17). The miRNA-1246 gene has been shown to function as a proto-oncogene in several types of cancer, including those of the liver, lungs, and colon (2, 18). To date, no studies have been conducted in Iraq regarding miRNA-1246 and any autoimmune diseases; therefore, this research is the first to assess the role of miRNA-1246 in susceptibility to SLE in Iraqi patients. This study aims to evaluate the expression of miRNA-1246 in women with systemic lupus erythematosus.

Methods

The present cross-sectional study was carried out after obtaining approval from the Ethics Commission (approval No. CSEC/0924/0060). Written informed consent was obtained from each participant prior to enrollment in the study.

The present study included 100 women diagnosed with SLE who were recruited from Baghdad Medical City and Baghdad Hospital in Baghdad, Iraq. The diagnosis was established by rheumatologists at the unit clinic using the classification criteria for systemic lupus erythematosus (SLE) developed by the European League Against Rheumatism and the American College of Rheumatology (EULAR/ACR) (19). Analytical and clinical assessments were also conducted as part of the study protocol. In addition, the attending physician utilized blood samples to determine each patient's SLEDAI-2K score. Demographic and clinical data, including age, body mass index (BMI), and other relevant variables, were collected thereafter. All participants were required to sign an informed consent form before being enrolled in the study. Under the guidance of medical professionals at Baghdad Medical City and Baghdad Hospital, a research group from the University of Baghdad conducted this investigation in Baghdad, Iraq. One hundred samples from the healthy control (HC) group were obtained from the National Blood Transfusion Centre. For the purpose of comparing gene expression levels between patients and healthy controls, blood samples were used as specimens for measuring the expression levels of immune cell-specific genes (20).

Blood Sample Collection: Five milliliters of venous blood was collected from each subject. Each sample was divided into three parts:

1.The first part (2 mL) of whole blood was transferred into an EDTA tube for laboratory tests.

2.The second part (0.25 mL) of blood was added to 0.75 mL of TRIzol™ Reagent for RNA extraction.

3.The third part (2.5 mL) was transferred into a gel tube, placed in a serum-separating tube, and then centrifuged for 10 minutes at 3000 rpm to separate the serum for other laboratory tests.

Laboratory investigations: Demographic information and a number of clinical signs were gathered from each patient's medical record. Laboratory investigations were conducted to perform standard and other laboratory tests. C-reactive protein, complete blood count, and erythrocyte sedimentation rate were all measured as samples were collected. Measurements of complement 4 (C4) and complement 3 (C3) were performed using radial immunodiffusion (RID). For the analytical assessment of anti-dsDNA and anti-nuclear autoantibodies, the enzyme-linked immunosorbent assay (ELISA) method was used.

Total RNA extraction: Total RNA was isolated from the whole blood of patients and healthy controls (HC). In this study, RNA extraction from blood samples was performed using TRIzol for RNA isolation, and pure RNA was prepared for analysis (TransZol Up Plus RNA Kit; TransGen Biotech; Cat. No. ER501). The isolated RNA was stored at -80 °C until processing for downstream applications. The purity and concentration of the extracted RNA were assessed using a NanoDrop spectrophotometer (Thermo Fisher), which showed that the purity (A260/A280 ratio) was 2.0 and the concentration of extracted RNA ranged from 43 to 77 ng/µL. The results indicated that the purity and concentration of the extracted RNA from blood samples were sufficient for subsequent analysis.

Primer design for microRNA-1246 gene expression: With microRNA-U6 serving as the control (reference) gene, the cDNA sequences corresponding to the microRNA-1246 gene were retrieved from the National Center for Biotechnology Information (NCBI) GenBank database. The specific primer sequences employed in the present study are presented in Table 1.

Table 1. The microRNA-1246 forward and reverse primers utilized in this work

Primer of miRNA-1246	Sequences 5' to 3'	References
miRNA1246 forward primer	TACGAAATGGATTTTTGGAGCAGG	In this study
MiRNA1246 reverse primer	GTCCTTGGTGCCCGAGTG	In this study
U6 forward primer	ATTGGAACGATACAGAGAAGATT	In this study
miRNA-universe R.P.	GCGAGCACAGAATTAATACGAC	In this study

Synthesis of cDNA from microRNA: DNA synthesis supermix and EasyScript® One-Step gDNA Removal were used. The conditions for cDNA reverse transcription consisted of a thermal cycler incubation step at 25 °C for 10 minutes, incubation of the 20 µL cDNA synthesis reaction at 42 °C for 15 minutes, and an enzyme inactivation step at 85 °C for 5 seconds. For downstream qRT-PCR amplification, the cDNA product must be stored at -20 °C.

Quantitative Real-Time PCR (qRT-PCR) for microRNA: Quantitative real-time PCR (qRT-PCR) was utilized for estimation of microRNA expression levels. PerfectStart® Green qPCR SuperMix (TransGen Biotech; Cat. No. AQ601) was used. The SYBR® channel on the qRT-PCR instrument was used to detect and quantify target microRNA using qPCR MasterMix, an optimized 2× reaction mix (Qiagen Rotor-Gene Q). The total reaction mix (20 µL) contained 6 µL of nuclease-free water, 2 µL of template cDNA, 1 µL of each forward and reverse primer (10 µM), and 10 µL of Green qPCR SuperMix. The cycling protocol was programmed according to the following optimized thermal profile: one cycle at 94 °C for 1 minute for initial denaturation, followed by 35 cycles of denaturation at 94 °C for 10 seconds and extension at 58 °C for 15 seconds.

Calculation of mi-RNA 1264 expression: The relative cycle threshold method (2^−ΔΔCt), first described by Livak and Schmittgen, was utilized to quantify fold changes in the quantitative expression of the miRNA. The relative gene expression ratio compares the two groups (control and test) and determines the mean value.

Statistical analysis: Study results were subjected to statistical evaluation using IBM SPSS Statistics 27.0. Graphs were plotted with GraphPad Prism 9.4.1. Significant variations were assessed using the Pearson chi-square test, with a significance level of 0.05. The correlation coefficient between variables was estimated in this study. Normally distributed (parametric) variables were given as mean±standard deviation (SD) for each sample (patient and control) variable, and $p<0.05$ was considered significant.

Results

This case-control investigation included 100 women diagnosed with SLE and 100 women without SLE. The mean age of the SLE group was 30.93±9.85 years, whereas the mean age of the HC group was 32.97±9.13 years (p=0.13), indicating no significant difference between the two groups (Figure 1). Female SLE patients and HCs ranged in age from 15 to 50 years. In the present study, the age distribution of SLE patients was categorized into four groups: 15-23, 24-32, 33-42, and 42-50 years (Figure 2). The percentage of SLE patients in the 24-32-year age group was 32.00%, which was higher than that observed in the other age groups: 15-23 years (25.00%), 33-42 years (29.00%), and 42-50 years (14.00%). The mean BMI of SLE patients was 28.62±6.50 kg/m², which was not significantly different from that of the HC group (27.11±5.60 kg/m²; p=0.07) (Figure 3).

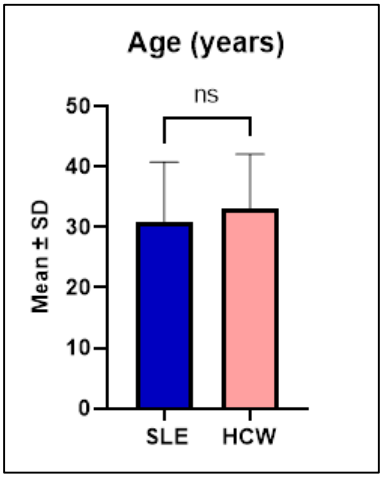

Figure 1. Mean±SD of age in systemic lupus erythematosus (SLE) patients and healthy controls (HC). Columns represent the mean; bars represent standard deviation (SD). ns: not significant (p>0.05)

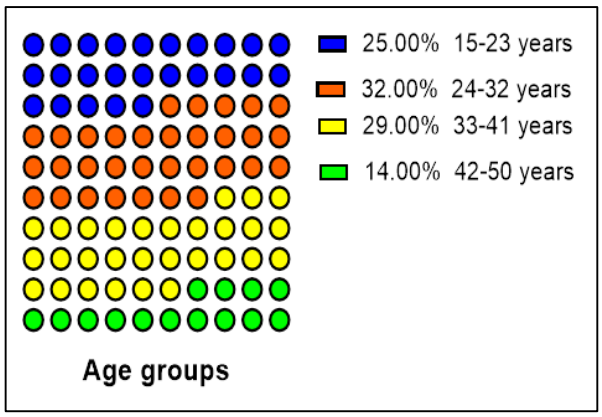

Figure 2. Distribution of SLE patients according to the age groups

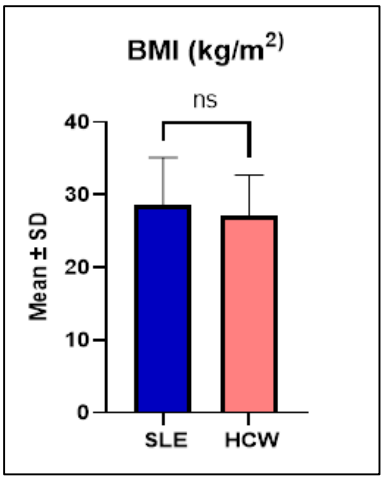

Figure 3. Mean±SD of BMI in systemic lupus erythematosus patients and healthy controls (HC). Columns represent the mean; bars represent standard deviation (SD). ns: not significant (p>0.05)

Disease duration and family history: With respect to medication status, the SLE patients were divided into two groups: group one (24%) comprised patients who were newly diagnosed and had not taken any medications, while group two (76%) comprised patients with a disease duration of 6.23±0.58 years who were taking medication. Among the SLE patients, 74% had a positive family history, whereas 26% had no family history, as illustrated in Figure 4. Patients who took their treatment regularly experienced fewer symptoms compared with those who were newly diagnosed (i.e., not taking any treatment), who suffered from more severe symptoms of the disease. No statistically significant differences were observed with respect to SLE duration, use of immunosuppressants and antimalarials, prednisone dosage, or age among the female participants in the study.

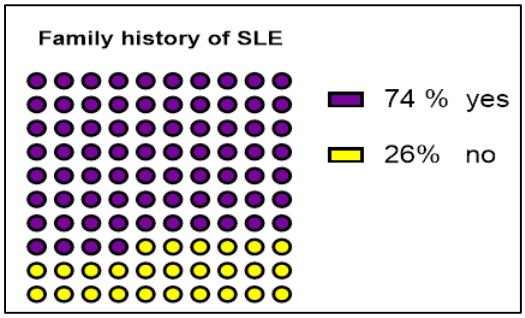

Figure 4. SLE patients distributed according to family history

The mean Hb level was decreased (8.44±0.12 g/dL) in SLE patients. Table 2 shows an elevation in the WBC count in patients with SLE (2.65±8.39 × 10⁹/L). The erythrocyte sedimentation rate (ESR) results are listed in Table 2; the results showed an elevation (42±2.91 mm/h) in ESR among SLE patients. The results for platelet levels showed normal values in patients suffering from SLE, with a mean of 361±0.11 × 10⁹/L.
Expression levels of miRNA-1246: To quantify miRNA-1246 expression, quantitative real-time polymerase chain reaction (qRT-PCR) was used. Gene expression was quantified using the Livak method, with the miRNA-U6 gene serving as a housekeeping gene for normalization. Amplification of miRNA-1246 by qRT-PCR is shown in Figure 5, and the fold change in miRNA-1246 expression according to the 2^−ΔΔCt method is shown in Table 3.

Table 2. Laboratory tests of SLE patients

Parameters	Patients (n=100) Mean±SD	Normal values
Hb g/dl	8.44±0.12	11.6-15
WBC x10^9/L	2.65±0.39	4.5-11.0
ESR (mm/h)	42±2.91	0-20
CRP mg/dl	17±1.86	<10
C3 mg/dl	15±2.35	88-201
C4 mg/dl	8.04±1.30	16-48
Anti-dsDNA (AU/mL)	41±4.03	<10
ANA (AU/mL)	4.86±0.59	<40.0
PLTx10^9/L	361±0.11	150-450

Hb: hemoglobin; C4: fourth component of complement; CRP: C-reactive protein; PLT: platelet; ANA: anti-nuclear antibody; ESR: erythrocyte sedimentation rate; WBC: white blood cell; C3: third component of complement; anti-dsDNA: anti-double-stranded DNA antibody

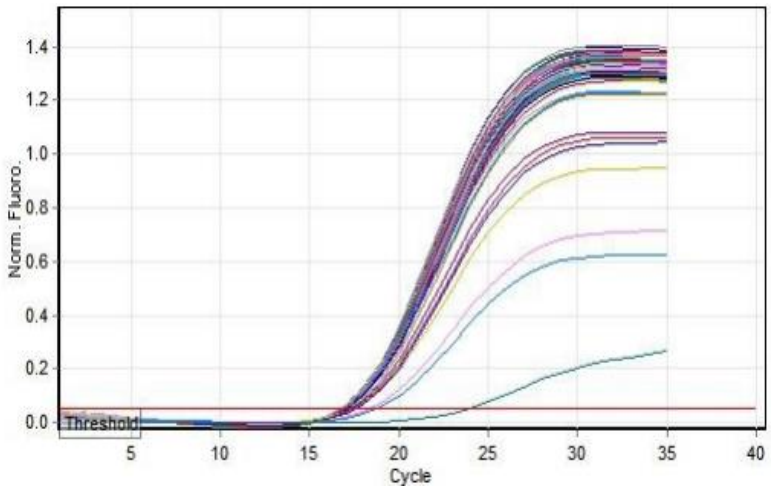

Figure 5. Amplification curve obtained by qRT-PCR for microRNA-1246

Table 3. Fold change in microRNA-1246 expression determined by the 2^−ΔΔCt method

Groups	Means Ct of miRNA-1246	Means Ct of miRNA-U6	Mean ΔCt Target (ct miRNA 1246-ct miRNAU6)	ΔΔCt	A fold of gene expression 2^−ΔΔCt	p-value
Patients	16.91	18.83	-1.91	-1.21	2.31	<0.0001
HC	18.13	18.84	-0.70	0	1.00	<0.0001

Patients with systemic lupus erythematosus had a higher mean fold change (2.31-fold) of the miRNA-1246 gene relative to the mean fold change among HC women, which was 1.00-fold, with statistically significant variation (p<0.0001) (Figure 6). There was a substantial increase in miRNA-1246 and HOTAIR expression in all BC subjects. Furthermore, there was an impressive increase in the expression of miRNA-1246 and HOTAIR across all stages of BC patients. Notably, in stage III, the expression of both genes was much greater than in stages I and II. Also, among breast cancer patients, there was a clear positive correlation between the differential expression fold of miRNA-1246 and HOTAIR. In addition, correlation analysis was conducted to evaluate the relationship between miRNA-1246 expression and all parameters under study in patients with SLE.

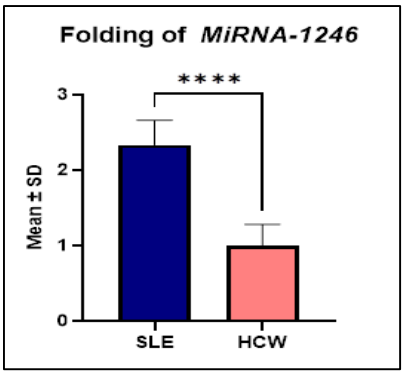

Figure 6. Column-bar plot of miRNA-1246 fold change in patients with systemic lupus erythematosus (SLE) and healthy controls (HC). Columns represent the mean; error bars represent standard deviation (SD). The Mann–Whitney test was used to examine significant differences (** p<0.0001)**

In individuals with systemic lupus erythematosus, miRNA-1246 was shown to have a correlation coefficient and p-value with all of the study parameters (Table 4). There was no statistically significant relationship between miRNA-1246 and any of the variables included in the correlation analysis. From each patient's medical record, demographic information and a number of clinical signs were recorded. The standard and additional laboratory tests were evaluated by laboratory examinations.

Distribution of SLE Patients According to Medication: With regard to their medication status, patients with SLE were distributed into two groups: one group comprised newly diagnosed patients, and the other group comprised patients who were receiving medication. Comparisons between the two groups across all parameters under study are presented in Table 5. The results showed significant differences in some parameters, including BMI, disease duration, ANA, anti-dsDNA, ESR, and CRP, while other parameters appeared non-significant between the two groups, as shown in Table 5.

Table 4. Correlation analysis of microRNA-1246 with baseline data parameters in patients with systemic lupus erythematosus

Parameters	Folding of microRNA-1246 in SLE patients	
	rs_p	p-value
Ages(years)	-0.026	0.791[ns]
BMI	-0.051	0.610[ns]
Disease duration	0.061	0.543[ns]
Hb g/dl	-0.031	0.759[ns]
WBC x10^9/L	-0.043	0.669[ns]
C4 mg/dl	-0.047	0.635[ns]
CRP mg/dl	-0.087	0.388[ns]
ANA (AU/mL)	-0.013	0.891[ns]
ESR mm/h	0.035	0.729[ns]
Anti-dsDNA (AU/mL)	0.061	0.872[ns]
C3 mg/dl	0.056	0.575[ns]
PLTx10^9/L	-0.028	0.779[ns]

p: probability (ns: not significant); PLT: platelet; ANA: anti-nuclear antibody; anti-dsDNA: anti-double-stranded DNA antibody; CRP: C-reactive protein; correlation coefficient (r); BMI: body mass index; C4: fourth component of complement; Hb: hemoglobin; WBC: white blood cell; C3: third component of complement; ESR: erythrocyte sedimentation rate

Table 5. Comparison between newly diagnosed and medicated patients with SLE in all parameters under study

Parameters	Newly diagnosis SLE (n=24) Mean±SD	Medicated SLE patients (n=76) Mean±SD	p-value
Ages(years)	31.79±9.39	28.42±9.44	0.134[ns]
BMI	29.33±6.60	26.38±5.73	0.040*
Disease duration	0.0±0.0	6.23±0.58	0.0001****
ANA (AU/mL)	2.98±2.13	10.82±1.89	0.0004****
MiRNA-1246	2.34±0.35	2.28±0.20	0.266[ns]
WBC x10^9/L	2.66±0.10	2.59±0.15	0.693[ns]
C4 mg/dl	6.10±1.11	14.18±3.39	0.058[ns]

Hb g/dl	8.46±1.19	8.39±1.43	0.883[ns]
CRP mg/dl	12.58±1.60	31.85±4.91	0.001***
ESR mm/h	37.47±3.24	59.17±5.30	0.001***
PLTx10^9/L	363±1.09	353±1.21	0.723[ns]
C3 mg/dl	17.04±2.99	10.74±2.41	0.104[ns]
Anti-dsDNA (AU/mL)	34.28±4.23	63.83±8.83	0.004***

(p<0.01; ns: not significant); p: probability (*p<0.05); complement 3 (C3); correlation coefficient (r); anti-double-stranded DNA (anti-dsDNA); erythrocyte sedimentation rate (ESR); platelet (PLT); complement 4 (C4); hemoglobin (Hb); C-reactive protein (CRP); BMI: body mass index; anti-nuclear antibody (ANA); white blood cell (WBC)**

Discussion

The present study demonstrated that 32% of SLE patients belonged to the 24-32-year age group, a proportion higher than that observed in the other age groups. Román Ivorra et al. (21) reported that the illness predominantly affects young individuals, particularly young women, and the age distribution observed in our study corroborated this finding. In Europe, systemic lupus erythematosus affects 0.09% of the population and is most commonly diagnosed among working-age women, with a median age at diagnosis of 33 years (21). According to another study conducted in Japan, females between the ages of 15 and 44 are more susceptible to SLE (22). This age distribution was congruent with the findings of the Iraqi study, which indicated that, compared with late-onset disease (>45 years), the majority of patients were young adults (≤45 years) (23). Furthermore, female patients from Iraq and other Arab populations have also demonstrated a similar pattern with respect to age distribution (24). Nevertheless, the purpose of our research was not to examine age groups among Iraqi women with SLE; rather, we utilized patients' age classifications to collect healthy controls (HC) that were age-matched.

The present study concluded that the presence of a family history of SLE increases the likelihood of developing the disease at a higher rate than in individuals who do not have a family history. At the same time, the presence of a family history does not preclude the possibility of non-occurrence. Our study does not agree with the findings of Ulff-Møller et al. (25), who reported that patients with a family history represented 18 (18%) compared with patients without any family history, who represented 82 (82%).

SLE is an autoimmune disorder characterized by the production of autoantibodies directed against self-antigens, accompanied by variable clinical manifestations and an unpredictable course of flares (26).

Anemia is a prevalent hematological condition, and it may be observed in up to 50% of patients with SLE, suggesting that autoimmune antibodies contribute to a reduced red blood cell count (27). The present study was in agreement with the findings of Bharath et al. (28), who reported that the mean hemoglobin level was lower in patients with SLE.

Despite the low WBC counts observed in SLE patients in previous studies (29, 30), contrary results were obtained in the present investigation. The reasons for this discrepancy may be attributed to a number of factors, including patient heterogeneity, small sample size, heterogeneity of disease presentation, and different treatment approaches. According to the study by Li et al. (31), individuals with SLE exhibit an elevated white blood cell percentage owing to bacterial or viral infection. Conversely, the incidence of infection is higher because of the treatment method and the number of medications used to suppress the immune system. Neutropenia and leukopenia may have been less common as a result of the therapy, according to another study, which also found that concomitant disorders could influence leukocyte counts. In order to obtain a more accurate estimate of the prevalence of leukopenia, they excluded individuals who had infections, liver cirrhosis, cancer, other connective tissue diseases, or comparable conditions (32).

The ESR is a common hematological test that may indicate elevated inflammatory activity within the human body, triggered by one or more factors, such as autoimmune disorders, infectious diseases, or tumors. With respect to specific disease diagnosis, ESR lacks high specificity. However, when combined with other laboratory tests, it may serve as an indicator of elevated inflammatory activity (33). Several investigations have confirmed the favorable association between ESR and SLE, and all reported that ESR levels in patients with SLE were considerably higher than normal (34-37). An elevation in ESR is observed in autoimmune disorders, infections, and cancer (38).

As a consequence of the persistent inflammatory response of the immune system and the overproduction of immunoglobulins, individuals with SLE often exhibit elevated ESR levels (39, 40). Consistent with the study by Mok et al. (41), our findings support the concept that SLE patients may exhibit an inflammatory response via increased CRP levels. Reduced levels of complement proteins (C3 and C4) were observed in SLE patients in our investigation. This finding is in line with the results of an earlier study conducted in Iraq (42). Overproduction of autoantibodies and cytotoxicity mediate a reduction in complement proteins in SLE (43). Additionally, our research revealed that SLE patients had an elevated mean serum anti-dsDNA level (41±4.03). The presence of the anti-dsDNA antibody serves as a standard diagnostic and classification criterion for systemic lupus erythematosus (SLE) and as a marker for tracking the course of the illness. This criterion is further strengthened when patients test positive for ANA (44).

In SLE, anti-dsDNA contributes to damage in various organs, particularly the kidneys, skin, and central nervous system. The complement cascade—which involves the infiltration of immune cells and the production of cytokines—may be activated when anti-dsDNA binds to DNA or cross-reactive antigens in kidney cells. Inducing autoantibody synthesis and breaking tolerance to autoantigens requires the presence of double-stranded DNA at immunologically privileged sites. Due to elevated anti-dsDNA levels, a poor prognosis is anticipated (44). The current investigation found that SLE patients had a very high level of anti-dsDNA. Regarding anti-dsDNA positivity, previous research conducted in Iraq reported results comparable to those of the current study (42, 44). Consistent with other research, this finding confirms that SLE patients have elevated anti-dsDNA levels (45).

Patients with SLE had an elevated mean serum ANA level (4.86±0.59). The percentage of ANA-positive individuals in this research was greater than that reported by others (46), who found an increased incidence of elevated ANA, with a positive rate of 90% to 99% in SLE patients. Compared with males, females had higher and more age-related anti-nuclear antibody levels (47). The pathophysiology of autoimmune illnesses is significantly impacted by sex hormones, especially estrogen, which also makes women more likely to experience a recurrence of these conditions (48). Moreover, the results for platelet levels showed normal values in patients suffering from SLE, with a mean of $361\pm0.11 \times 10^9$/L, and this result agreed with the study by Abdullah et al. (49), who found non-significant differences between SLE patients and healthy individuals.

Small noncoding RNA molecules known as microRNAs (miRNAs) target messenger RNA (mRNA) for translational repression or degradation, thereby enabling post-transcriptional quantitative gene regulation (50). According to this review of recent studies, the majority of the immunological processes that cause SLE involve a number of miRNAs that are both ubiquitous and functional. With regard to miRNA-1246, very little is known about its function in the etiology of autoimmune disease. Tumor cell invasion, migration, proliferation, and metastasis have all been shown to be affected by miRNA-1246 signaling (14, 18). The miRNA-1246 gene has been shown to act as a proto-oncogene in several types of cancer, including colon, lung, and liver cancers (51). Using quantitative real-time PCR (qRT-PCR), the present work validated miRNA-1246 expression patterns and explored their role in SLE etiology. In our study, there is a contradiction with the study by Luo et al. (52), who found that the expression of miRNA-1246 in B cells

between 20 inactive SLE patients and 20 HCs showed no significant difference. Additionally, another study found that circulating levels of miRNA-1246 decrease in discoid lupus erythematosus (DLE) and subacute cutaneous lupus erythematosus (SCLE) patients (53). Our study is consistent with the study by Khaliefa et al. (16), which assessed the miRNA-1246/HOTAIR differential expression levels in healthy subjects compared with those in female patients with BC.

The study of Luo et al. and Abed et al. found a substantial drop in miRNA-1246 expression in B cells from SLE patients (52, 54), which is at odds with our findings. A small number of studies have examined the correlation between the selected miRNA-1246 expression and the illness parameters under investigation. Consistent with the study by Luo et al. (52), our research did not find any evidence linking changes in miRNA-1246 fold expression to SLE demographics or laboratory investigations. Findings point to miRNA-1246 as a potential biomarker in SLE etiology. These results are in line with the study by Abed et al. (55), which demonstrated that environmental and genetic variables may play a role not only in the development of heart failure but also in its progression. These results suggest that miRNA-1246 could be a novel biomarker or target for SLE. It was found that the parameters that gave significant differences were increased in medicated patients compared with newly diagnosed patients, except for BMI, which showed a decreased value in medicated patients. The explanation for these results may be due to patients' failure to take medications or their use of another medication that conflicts with the treatment, leading to worsening of the disease and increased concentrations of some parameters. In the autoimmune disorder known as drug-induced lupus (DIL), symptoms similar to those of systemic lupus erythematosus (SLE) manifest after exposure to certain drugs (56). This research concluded that the level of miRNA-1246 is regarded as a disease incidence risk factor and that it is higher in the blood of SLE patients. More research is needed to determine the precise role of miRNA-1246 in the immunopathogenesis of SLE.

Conflict of Interest: The authors have declared that no conflict of interest exists.

Acknowledgment

None.

References

1.Dörner T, Furie R. Novel paradigms in systemic lupus erythematosus. Lancet. 2019;393(10188):2344-58.
2.Horst AK, Kumashie KG, Neumann K, Diehl L, Tiegs G. Antigen presentation, autoantibody production, and therapeutic targets in autoimmune liver disease. Cell Mol Immunol. 2021;18(1):92-111.
3.Taheri M, Eghtedarian R, Dinger ME, Ghafouri-Fard S. Exploring the Role of Non-Coding RNAs in the Pathophysiology of Systemic Lupus Erythematosus. Biomolecules. 2020;10(6):937.
4.Fava A, Petri M. Systemic lupus erythematosus: Diagnosis and clinical management. J Autoimmun. 2019;96:1-13.
5.Tabarek AA, Reema MA. Genetic polymorphisms of HLA-G gene in rheumatoid arthritis. Iraqi J Agric Sci. 2023;54(2):378-87.
6.Rees F, Doherty M, Grainge MJ, Lanyon P, Zhang W. The worldwide incidence and prevalence of systemic lupus erythematosus: a systematic review of epidemiological studies. Rheumatology (Oxford). 2017;56(11):1945-61.
7.Tanaka Y. State-of-the-art treatment of systemic lupus erythematosus. Int J Rheum Dis. 2020;23(4):465-71.
8.Elsahookie MM, Cheyed SH, Dawood AA. An overview on mechanism of gene expression regulation. Iraqi J Agric Sci. 2021;52(2):454-60.
9.Luo JW, Wang X, Yang Y, Mao Q. Role of micro-RNA (miRNA) in pathogenesis of glioblastoma. Eur Rev Med Pharmacol Sci. 2015;19(9):1630-9.
10.Hussein AM, Al Sahlanee R. Serological and molecular evaluation of MRP14 in thyroiditis and its role in pro-inflammatory chemokines activation. Iraqi J Agric Sci. 2022;53(6):1368-76.
11.Jiang X, Kanda T, Wu S, Nakamura M, Miyamura T, Nakamoto S, et al. Regulation of microRNA by hepatitis B virus infection and their possible association with control of innate immunity. World J Gastroenterol. 2014;20(23):7197-206.
12.Li X. MiR-375, a microRNA related to diabetes. Gene. 2014;533(1):1-4.
13.Morin RD, O'Connor MD, Griffith M, Kuchenbauer F, Delaney A, Prabhu AL, et al. Application of massively parallel sequencing to microRNA profiling and discovery in human embryonic stem cells. Genome Res. 2008;18(4):610-21.
14.Zhang Y, Liao JM, Zeng SX, Lu H. p53 downregulates Down syndrome-associated DYRK1A through miR-1246. EMBO Rep. 2011;12(8):811-7.
15.Ren Y, Li X, Wang W, He W, Wang J, Wang Y. Expression of Peripheral Blood miRNA-720 and miRNA-1246 Can Be Used as a Predictor for Outcome in Multiple Myeloma Patients. Clin Lymphoma Myeloma Leuk. 2017;17(7):415-23.
16.Khaliefa AK, Desouky EM, Hozayen WG, Shaaban SM, Hasona NA. miRNA-1246, HOTAIR, and IL-39 signature as potential diagnostic biomarkers in breast cancer. Noncoding RNA Res. 2023;8(2):205-10.
17.Dai YC, Pan Y, Quan MM, Chen Q, Pan Y, Ruan YY, et al. MicroRNA-1246 Mediates Drug Resistance and Metastasis in Breast Cancer by Targeting NFE2L3. Front Oncol. 2021;11:677168.
18.Li XJ, Ren ZJ, Tang JH, Yu Q. Exosomal MicroRNA MiR-1246 Promotes Cell Proliferation, Invasion and Drug Resistance by Targeting CCNG2 in Breast Cancer. Cell Physiol Biochem. 2017;44(5):1741-8.
19.Aringer M, Leuchten N, Johnson SR. New Criteria for Lupus. Curr Rheumatol Rep. 2020;22(6):18.
20.Abdullah D, Aloubaidy RM. Genetic polymorhism of caspase 8 and 9 in iraq. Iraqi J Agric Sci. 2022;53(3):505-14.
21.Román Ivorra JA, Fernández-Llanio-Comella N, San-Martín-Álvarez A, Vela-Casasempere P, Saurí-Ferrer I, González-de-Julián S, et al. Health-related quality of life in patients with systemic lupus erythematosus: a Spanish study based on patient reports. Clin Rheumatol. 2019;38(7):1857-64.

22.Ohta A, Nagai M, Nishina M, Tomimitsu H, Kohsaka H. Age at onset and gender distribution of systemic lupus erythematosus, polymyositis/dermatomyositis, and systemic sclerosis in Japan. Mod Rheumatol. 2013;23(4):759-64.
23.Hameed Abdulridha R. Assessment of Immune Biomarkers and Polymorphisms of miR-146a and its Target geneTRAF6 in Female Patients with Systemic Lupus Erythematosus [PhD Thesis]. College of Science, University of Baghdad; 2022.
24.Adwan M. Clinical and Serologic Characteristics of Systemic Lupus Erythematosus in the Arab World: A Pooled Analysis of 3,273 Patients. Arch Rheumatol. 2018;33(4):455-63.
25.Ulff-Møller CJ, Simonsen J, Kyvik KO, Jacobsen S, Frisch M. Family history of systemic lupus erythematosus and risk of autoimmune disease: Nationwide Cohort Study in Denmark 1977-2013. Rheumatology (Oxford). 2017;56(6):957-64.
26.Morawski PA, Bolland S. Expanding the B Cell-Centric View of Systemic Lupus Erythematosus. Trends Immunol. 2017;38(5):373-82.
27.Santacruz JC, Mantilla MJ, Rueda I, Pulido S, Rodriguez-Salas G, Londono J. A Practical Perspective of the Hematologic Manifestations of Systemic Lupus Erythematosus. Cureus. 2022;14(3):e22938.
28.Bharath G, Kumar P, Makkar N, Singla P, Soneja M, Biswas A, et al. Mortality in systemic lupus erythematosus at a teaching hospital in India: A 5-year retrospective study. J Family Med Prim Care. 2019;8(7):2511-5.
29.Carli L, Tani C, Vagnani S, Signorini V, Mosca M. Leukopenia, lymphopenia, and neutropenia in systemic lupus erythematosus: Prevalence and clinical impact--A systematic literature review. Semin Arthritis Rheum. 2015;45(2):190-4.
30.Lertchaisataporn K, Kasitanon N, Wangkaew S, Pantana S, Sukitawut W, Louthrenoo W. An evaluation of the association of leukopenia and severe infection in patients with systemic lupus erythematosus. J Clin Rheumatol. 2013;19(3):115-20.
31.Li Z, Xiao Y, Zhang L. Application of procalcitonin, white blood cell count and neutrophil-to-lymphocyte ratio in the diagnosis of systemic lupus erythematosus with a bacterial infection. Ann Palliat Med. 2020;9(6):3870-6.
32.Lu W, Zhong Y, Zhang Y, Liu Z, Xue L. The Clinical Characteristics of Leukopenia in Patients with Systemic Lupus Erythematosus of Han Ethnicity in China: A Cross-Sectional Study. Rheumatol Ther. 2021;8(3):1177-88.
33.Dima A, Opris D, Jurcut C, Baicus C. Is there still a place for erythrocyte sedimentation rate and C-reactive protein in systemic lupus erythematosus?. Lupus. 2016;25(11):1173-9.
34.Abed RM, Yaaqoob LA. Novel single nucleotide polymorphism (rs1600485907) of IL-41 gene associated with systemic lupus erythematous. Asia-Pacific J Mol Biol Biotechnol. 2023;31(4):1-8.
35.Abira M, Akhter QS. Relationship between CBC and ESR and duration of disease in patients with systemic lupus erythematosus (SLE). J Bangladesh Soc Physiol. 2018;13(2):41-6.
36.Macejova Z, Madarasova Geckova A, Husarova D, Zarikova M, Kotradyova Z. Living with Systematic Lupus Erythematosus: A Profile of Young Female Patients. Int J Environ Res Public Health. 2020;17(4):1315.
37.Mohamed OS, Azmy GJ, Elfadl EM. Clinical significance of red blood cell distribution width in systemic lupus erythematosus patients. Egypt Rheumatol Rehabil. 2020;47(1):38.
38.Keenan RT, Swearingen CJ, Yazici Y. Erythrocyte sedimentation rate and C-reactive protein levels are poorly correlated with clinical measures of disease activity in rheumatoid arthritis, systemic lupus erythematosus and osteoarthritis patients. Clin Exp Rheumatol. 2008;26(5):814-9.
39.Hamza KM, Bashir ZM. Hematological profile of Sudanese patients with systemic lupus erythematosus. Clin Med J. 2016;2(4):40-5.

40.Stojan G, Petri M. Epidemiology of systemic lupus erythematosus: an update. Curr Opin Rheumatol. 2018;30(2):144-50.
41.Mok CC, Birmingham DJ, Ho LY, Hebert LA, Rovin BH. High-sensitivity C-reactive protein, disease activity, and cardiovascular risk factors in systemic lupus erythematosus. Arthritis Care Res (Hoboken). 2013;65(3):441-7.
42.Alaanzy MT, Alsaffar JM, Bari AA. Evaluate the correlation between antioxidant capacity and interferon Γ level with the disease activity of Sle patients in Iraqi Woman. Indian J Public Health Res Dev. 2020;11(1):1278-82.
43.Liu H, Yang LH, Yin G, Xie QB. [Correlation of Thyroid Autoantibodies,System Lupus Erythematosus Immunologic Indicators and Disease Activity in SLE with HT]. Sichuan Da Xue Xue Bao Yi Xue Ban. 2018;49(2):179-82. [Chinese]
44.Grygiel-Górniak B, Rogacka N, Puszczewicz M. Antinuclear antibodies in healthy people and non-rheumatic diseases - diagnostic and clinical implications. Reumatologia. 2018;56(4):243-8.
45.Gheita TA, Abaza NM, Hammam N, Mohamed AA, El-Gazzar II, Eissa AH. Anti-dsDNA titre in female systemic lupus erythematosus patients: relation to disease manifestations, damage and antiphospholipid antibodies. Lupus. 2018;27(7):1081-7.
46.Choi1 M, Fritzler M, Costenbader K, Urowitz M, Hanly J, Gordon C, et al. Dynamics of Anti-Nuclear Antibodies in a Longitudinal Study of a Large Systemic Lupus Erythematosus Cohort. Arthritis Rheumatol. 2020;72(Suppl 10). Available from: https://acrabstracts.org/abstract/dynamics-of-anti-nuclear-antibodies-in-a-longitudinal-study-of-a-large-systemic-lupus-erythematosus-cohort/
47.Wiik AS, Bizzaro N. Missing links in high quality diagnostics of inflammatory systemic rheumatic diseases: It is all about the patient! Auto Immun Highlights. 2012;3(2):35-49.
48.Moulton VR. Sex Hormones in Acquired Immunity and Autoimmune Disease. Front Immunol. 2018;9:2279.
49.Abdullah WL, Abed RM. Gene expression and single nucleotide polymorphism (rs1140713) of microRNA-126. Iraqi J Agric Sci. 2024;55(Special):1-11.
50.Saliminejad K, Khorram Khorshid HR, Soleymani Fard S, Ghaffari SH. An overview of microRNAs: Biology, functions, therapeutics, and analysis methods. J Cell Physiol. 2019;234(5):5451-65.
51.Ghafouri-Fard S, Khoshbakht T, Hussen BM, Taheri M, Samadian M. A Review on the Role of miR-1246 in the Pathoetiology of Different Cancers. Front Mol Biosci. 2022;8:771835.
52.Luo S, Liu Y, Liang G, Zhao M, Wu H, Liang Y, et al. The role of microRNA-1246 in the regulation of B cell activation and the pathogenesis of systemic lupus erythematosus. Clin Epigenetics. 2015;7(1):24.
53.Méndez-Flores S, Furuzawa-Carballeda J, Hernández-Molina G, Ramírez-Martinez G, Regino-Zamarripa NE, Ortiz-Quintero B, et al. MicroRNA Expression in Cutaneous Lupus: A New Window to Understand Its Pathogenesis. Mediators Inflamm. 2019;2019:5049245.
54.Abed RM, Abdulmalek HW, Yaaqoob LA. The role of miRNA20a and miRNA320 in Iraqi patients with COVID-19: a case–control study. Egypt J Med Hum Genet. 2023;24(1):68.
55.Abed RM, Abdulmalek HW, Yaaqoob LA, Altaee MF, Kamona ZK. Genetic polymorphism of TLR5 and TLR6 in Iraqi patients with heart failure disease. Iraqi J Sci. 2023;64(4):1662-74.
56.Xiao X, Chang C. Diagnosis and classification of drug-induced autoimmunity (DIA). J Autoimmun. 2014;48-49:66-72.