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Original article

ASSOCIATION OF HOMOCYSTEINE, VITAMIN B12, AND FOLIC ACID WITH CUTANEOUS AND SYSTEMIC DISEASE ACTIVITY IN LUPUS ERYTHEMATOSUS

Pollozhani Nora[1], Arifi Mevlane[1], Damjanovska-Krstikj Ljubinka[2], Bojadzioska Maja[2] Osmani Bashkim[2], Damevska Katerina[1]

[1]University Clinic for Dermatology, Faculty of Medicine, Ss. Cyril and Methodius University in Skopje, Republic of North Macedonia
[2]University Clinic for Rheumatology, Faculty of Medicine, Ss. Cyril and Methodius University in Skopje, Republic of North Macedonia
e-mail: pollozhani.nora@gmail.com

Abstract

Introduction: Cutaneous lupus erythematosus (CLE) is associated with elevated cardiometabolic risk. Homocysteine, a sulfur-containing amino acid, is implicated in immune activation and vascular injury. However, data specifically addressing homocysteine in CLE remain limited.

Objectives: To evaluate the associations between serum homocysteine, vitamin B12, and folic acid and cutaneous disease severity (CLASI) and systemic activity (SLEDAI-2K) in patients with lupus erythematosus.

Materials and methods: A cross-sectional observational study was conducted involving 51 patients with lupus erythematosus (CLE with or without systemic involvement) at the University Clinic **for** Dermatology, Skopje, North Macedonia (May 2024 – May 2025). Serum homocysteine, vitamin B12, and folic acid were measured. Disease activity was assessed using CLASI and SLEDAI-2K. Correlations were evaluated using Pearson and Spearman coefficients.

Results: The cohort was predominantly female (92.2%), with a mean age of 53.18 ± 12.5 years. CLE subtypes included ACLE (25.5%), SCLE (43.1%), and CCLE (31.4%). Mean homocysteine was 14.48 ± 4.2 μmol/L, and 52.9% of patients exhibited hyperhomocysteinemia (>13 μmol/L). Homocysteine correlated positively with CLASI (r=0.3226, p=0.021) and SLEDAI-2K (r=0.3195, p=0.022). Vitamin B12 demonstrated a moderate inverse correlation with SLEDAI-2K (R=−0.4744, p=0.0004). Folic acid was not significantly associated with either disease measure.

Conclusions: Homocysteine is positively associated with both cutaneous and systemic lupus disease activity, while vitamin B12 is inversely associated with systemic activity. These preliminary findings from an ongoing cohort study, which aims to enroll 100 patients and 100 matched healthy controls, support homocysteine as a candidate biomarker spanning both disease domains and warrant confirmation in the complete dataset.

Keywords: cutaneous lupus erythematosus; homocysteine; CLASI; SLEDAI-2K; vitamin B12; folic acid; disease activity

Introduction

The skin is the second most frequently affected organ system in lupus erythematosus, with cutaneous manifestations occurring in approximately 70–85% of patients during the course of systemic lupus erythematosus (SLE) and serving as the initial presentation in up to 25% of cases[1]. Cutaneous lupus erythematosus (CLE) may present as an isolated, skin-limited disease or in association with systemic involvement[2]. CLE encompasses a broad spectrum of dermatologic manifestations and is commonly classified, based on clinical and histopathologic features and supportive laboratory findings, into acute cutaneous lupus erythematosus (ACLE), subacute cutaneous lupus erythematosus (SCLE), and chronic cutaneous lupus erythematosus (CCLE)[3]. Because cutaneous activity and damage may fluctuate and do not necessarily parallel systemic disease activity, validated instruments are essential for standardized assessment. The Cutaneous Lupus Erythematosus Disease Area and Severity Index (CLASI) provides a skin-specific measure of activity and damage[4,5], whereas the SLE Disease Activity Index 2000 (SLEDAI-2K) captures multisystem activity and may not reflect the severity of cutaneous involvement[6]. Beyond their diagnostic value, cutaneous manifestations impose a substantial psychosocial burden; patients with CLE report emotional and functional impairment comparable to or exceeding that observed in major chronic conditions such as congestive heart failure and type 2 diabetes[7-10].

Accumulating epidemiologic evidence indicates that CLE is associated with a substantially elevated cardiometabolic risk. Population-based cohorts and large-scale analyses have reported higher rates of major cardiovascular events and cardiovascular mortality among patients with CLE compared to the general population, with particularly strong signals observed in discoid lupus and for both thromboembolic and atherosclerotic outcomes[11-13]. In parallel, patients with CLE demonstrate increased prevalence of hypertension, diabetes, obesity, chronic kidney disease, and dyslipidemia[11-14]. These observations underscore the need to identify biomarkers that may bridge immune-mediated skin inflammation and systemic vascular injury. Homocysteine, a sulfur-containing aminothiol regulated by folate and vitamin B12 and implicated in both endothelial dysfunction and immune activation, is one such candidate.

Homocysteine is an intermediate of methionine metabolism and is metabolized through folate- and vitamin B12-dependent remethylation or vitamin B6-dependent transsulfuration. [15,16] Homocysteine is an established vascular risk marker with well-characterized mechanistic links to endothelial dysfunction, oxidative stress, impaired nitric oxide bioavailability, and prothrombotic signaling[17,18]. Beyond its vascular role, homocysteine may amplify immune activation and promote autoimmune pathways, suggesting that hyperhomocysteinemia reflects or contributes to disease activity and tissue injury in autoimmune conditions[19,20]. Consistent with this concept, meta-analyses demonstrate significantly higher serum homocysteine levels in SLE patients compared to healthy controls, with variable associations with systemic disease activity and vascular outcomes[21,22]. In CLE specifically, however, evidence remains very limited; to date, only one study has evaluated homocysteine using a validated cutaneous disease severity measure, reporting a positive correlation between homocysteine levels and CLASI activity[23].

Because homocysteine levels are strongly influenced by folate and vitamin B12 status, as well as by lifestyle and anthropometric factors, examining these parameters together may clarify the metabolic-immune axis in lupus erythematosus. Using the upper reference limit applied in the local laboratory (>13 μmol/L) to define hyperhomocysteinemia, serum homocysteine, folate, and vitamin B12 were assessed in relation to cutaneous disease severity (CLASI) and systemic disease activity (SLEDAI-2K) in a cohort of patients with lupus erythematosus (CLE with or without systemic involvement). Clinical correlates of homocysteine, including smoking status, age, and body mass index, were also explored.

Aim

The aim of this study was to evaluate the associations between serum homocysteine, vitamin B12, and folic acid and disease activity in patients with lupus erythematosus, using both a validated cutaneous severity measure (CLASI) and a systemic disease activity index (SLEDAI-2K). Additionally, the study sought to explore clinical determinants of homocysteine, including age, body mass index, smoking status, and disease duration.

Material and methods

Study design and setting

This was a cross-sectional observational study conducted at the University Clinic for Dermatology, Faculty of Medicine, Ss. Cyril and Methodius University, Skopje, North Macedonia, including patients evaluated between May 2024 and May 2025. The University Clinic for Dermatology is the leading tertiary care dermatology center in the country and receives referrals from Skopje and surrounding regions. The following clinical data were recorded at the study visit: demographic characteristics, disease subtype and duration, smoking status, body mass index, and disease activity scores.

Patient selection and assessments

A total of 51 patients with lupus erythematosus (LE) were included in the study. Eligible participants were adults (≥18 years) of both sexes with a confirmed diagnosis of cutaneous lupus erythematosus (CLE), with or without systemic involvement. The diagnosis of CLE was based on medical history, clinical examination, laboratory and serologic findings, and, when available, histopathology and direct immunofluorescence (DIF). CLE subtype (ACLE, SCLE, or CCLE) was defined according to the Gilliam classification[24]. SLE status was determined according to the 2019 EULAR/ACR classification criteria [25]. SLEDAI-2K scores were calculated for all 51 patients[26].

Inclusion criteria were: adults (≥ 18 years) of both sexes with a confirmed diagnosis of CLE, with or without systemic involvement. Patients with systemic features fulfilling the 2019 ACR/EULAR classification criteria for SLE were eligible for inclusion. Exclusion criteria were: pregnancy, severe renal impairment, current folate or vitamin B12 supplementation, and concomitant conditions known to strongly affect homocysteine levels.

Cutaneous disease severity was assessed using the Cutaneous Lupus Erythematosus Disease Area and Severity Index (CLASI) [4,5]. Cutaneous severity was categorized as mild (CLASI 0-9), moderate (CLASI 10-20), and severe (CLASI 21-70). Systemic disease activity was assessed using the Systemic Lupus Erythematosus Disease Activity Index 2000 (SLEDAI-2K)[26]. Demographic and clinical information collected at the study visit included sex, age, disease duration since symptom onset, body mass index (BMI, calculated as weight (kg)/height (m^2)), and smoking status, which was self-reported and categorized as non-smoker, former smoker, or current smoker.

Laboratory measurements

Venous blood samples were obtained during the study visit, after fasting. Serum homocysteine, vitamin B12, and folic acid concentrations were determined by electrochemiluminescence immunoassay (ECLIA) in the Clinical Biochemistry Laboratory, Skopje, and reported in μmol/L, pmol/L, and nmol/L, respectively, according to the manufacturer's instructions. Laboratory procedures were performed in accordance with routine laboratory quality standards. Hyperhomocysteinemia was defined as exceeding the upper reference limit in our laboratory (> 13 μmol/L).

Ethical issues

The study was conducted in accordance with the ethical guidelines of the Declaration of Helsinki and was approved by the Ethics Committee of the Faculty of Medicine, Ss. Cyril and Methodius University, Skopje, North Macedonia (Reference number: 03-2031/13; Date of approval: 17.04.2024). Written informed consent was obtained from all participants prior to enrolment.

Statistical analysis

Statistical analyses were performed using the Statistical Package for the Social Sciences (SPSS Inc., Chicago, IL, USA), version 25.0. Data distribution was assessed using the Shapiro–Wilk test. Categorical variables are presented as absolute and relative frequencies (n, %). Continuous variables are presented as mean ± standard deviation (SD), median, and interquartile range (IQR), as appropriate. To compare serum homocysteine, vitamin B12, and folic acid levels between age groups, the independent-samples t-test or the Mann–Whitney U test was used, depending on data distribution. Comparisons across smoking status groups were performed using one-way analysis of variance (ANOVA) or the Kruskal–Wallis test, as appropriate. Correlations between homocysteine, vitamin B12, and folic acid and variables of interest were evaluated using Pearson's correlation coefficient and Spearman's rank correlation coefficient, as appropriate. Statistical significance was defined as a two-sided p-value < 0.05.

Results

Demographic characteristics

A total of 51 patients with lupus erythematosus were included in the analysis. The cohort was predominantly female (47/51; 92.2%), with 4 men (7.8%). Age ranged from 27 to 84 years, with a mean age of 53.18±12.5 years; 27 patients (52.9%) were older than 50 years. With respect to smoking status, 23 patients (45.1%) were current smokers, 22(43.1%) were non-smokers, and 6(11.8%) were former smokers. Mean body mass index was 22.61±1.80 kg/m². Demographic characteristics are summarised in Table 1.

Table 1. Demographic characteristics of patients (n=51)

Characteristic		n (%)
Sex		
Total patients		51
Female		47(92.2%)
Male		4(7.8%)
Age (years)		
Mean ± SD	53.18±12.5	Range: 27-84
≤50 years		24(47.1%)
>50 years		27(52.9%)
BMI (kg/m²)		
Mean ± SD	22.61±1.80	
Smoking status		
Non-smoker		22(43.1%)
Former smoker		6(11.8%)
Current smoker		23(45.1%)

CLE subtypes

Regarding CLE phenotypes, patients were classified as SCLE (n=22; 43.1%), CCLE (n=16; 31.4%), or ACLE (n=13; 25.5%). All 13 patients with ACLE fulfilled the 2019 EULAR/ACR classification criteria for SLE, whereas the remaining 38 patients had isolated CLE. Mean disease duration was 9.3 ± 11.1 years (median 5.0 years; range 0.2–43). CLE subtype distribution is presented in Table 2.

Table 2. CLE subtypes and disease duration (n=51)

CLE subtype		n (%)
ACLE		13(25.5%)
SCLE		22(43.1%)
CCLE		16(31.4%)
Disease duration (years)		
Mean ± SD	9.3±11.1	Range: 0.2-43
Median	5.0	

Disease activity

Cutaneous severity measured by CLASI ranged from 1 to 48 (mean 14.35±13.1; median 9, IQR 3-24). According to predefined CLASI severity categories, 27 patients (52.9%) had mild disease, 7(13.7%) moderate disease, and 17(33.3%) severe cutaneous involvement. Systemic disease activity measured by SLEDAI-2K ranged from 2 to 10 (mean 4.76±2.2; median 4, IQR 3-6); all patients fell within the mild to moderate activity category (SLEDAI-2K 1-10). Disease activity parameters are presented in Table 3.

Table 3. Disease activity parameters (n=51)

Parameter		n (%) / value
CLASI activity score		
Mean ± SD	14.35±13.1	Range: 1-48
Median (IQR)	9(3-24)	
Mild (0-9)		27(52.9%)
Moderate (10-19)		7(13.7%)
Severe (20-70)		17(33.3%)
SLEDAI-2K score		
Mean ± SD	4.76±2.2	Range: 2-10
Median (IQR)	4(3-6)	
Mild to Moderate activity (1-10)		51(100%)

Immunological findings

Antinuclear antibodies (ANA) were positive in 25 patients (49.0%). Anti-dsDNA antibodies were detected in 19(37.3%) patients. Among extractable nuclear antigen (ENA) antibodies, anti-Ro (SSA) was the most frequently detected, present in 22 patients (43.1%), followed by anti-La (SSB) in 8(15.7%), anti-U1RNP in 5(9.8%), and anti-Sm in 2(3.9%). Anti-cardiolipin antibodies were positive in 5 patients (9.8%) and lupus anticoagulant in 2 (3.9).

Table 4. Immunological findings (n=51)

Parameter	n (%)
Autoantibodies	
Antinuclear antibodies (ANA)	25 (49.0%)
Anti-double stranded DNA (anti-dsDNA)	19 (37.3%)
Anti-Ro (SSA)	22 (43.1%)
Anti-La (SSB)	8 (15.7%)
Anti-Sm	2 (3.9%)
Anti-U1RNP	5 (9.8%)
Anti-cardiolipin antibodies	5 (9.8%)
Lupus anticoagulant	2 (3.9%)
Complement	
C3 (g/L), mean ± SD	1.07 ± 0.33
C4 (g/L), mean ± SD	0.26 ± 0.31
Low C3 (<0.9 g/L), n (%)	10 (19.6%)
Low C4 (<0.16 g/L), n (%)	15 (29.4%)

Mean complement levels were C3 1.07±0.33 g/L and C4 0.26±0.31 g/L; low C3 was found in 10 patients (19.6%) and low C4 in 15 patients (29.4%). Immunological findings are summarised in Table 4.

Biomarker levels

Serum biomarker levels are summarised in Table 5. Mean serum homocysteine (Hcy) was 14.48 ± 4.2 μmol/L (range 7-25 μmol/L), with a median of 13.32 μmol/L (IQR 12.0-18.0). Using the local laboratory upper reference limit (>13 μmol/L), hyperhomocysteinemia was present in 27 patients (52.9%). Vitamin B12 showed a skewed distribution (mean 363.85 ± 270.8 pmol/L; median 284.6, IQR 215-431). Folic acid had a mean of 8.46±6.3 nmol/L and a median of 6.05 nmol/L (IQR 4.92-9.68).

Table 5. Serum biomarker levels (n=51)

Variable	Mean ± SD	Median (IQR)	Range	n
Homocysteine (μmol/L)	14.48±4.2	13.32(12.0-18.0)	7-25	51
Vitamin B12 (pmol/L)	363.85±270.8	284.6(215-431)	-	51
Folic acid (nmol/L)	8.46±6.3	6.05(4.92-9.68)	-	51
Hyperhomocysteinemia (>13 μmol/L) — n (%): 27 (52.9%)				

Homocysteine by CLE subtype

Homocysteine levels did not differ significantly across CLE subtypes (Kruskal–Wallis p=0.579). Mean (±SD) Hcy was 14.89±3.47 μmol/L in ACLE, 14.42±4.49 μmol/L in SCLE, and 14.22±4.52 μmol/L in CCLE. The prevalence of hyperhomocysteinemia was 61.5% in ACLE (8/13), 54.5% in SCLE (12/22), and 43.8% in CCLE (7/16), with no significant differences between subtypes (χ^2 p=0.622).

Age and smoking effects

When stratified by age group (≤ 50 vs. > 50 years), patients did not differ significantly in Hcy (p=0.089), vitamin B12 (p=0.35), or folate (p=0.546). Nevertheless, Hcy tended to be higher in those aged 50 years or older (15.42±4.2 vs**.** 13.42±4.1 μmol/L). Correlation analysis demonstrated a significant positive association between age and Hcy (r=0.3025, p=0.031), while age was not significantly correlated with vitamin B12 (R=-0.2553, p=0.07) or folate (R=-0.0515, p=0.72). Smoking status was not associated with Hcy (p=0.451) or vitamin B12 (p=0.796). In contrast, folate levels differed significantly across smoking categories (p= 0.0145), driven by higher folate levels in former smokers compared **to** current smokers (post-hoc p=0.0148).

Correlations with disease activity

In primary correlation analyses, Hcy showed significant positive associations with both disease measures: SLEDAI-2K (r=0.3195, p=0.022) and CLASI (r=0.3226, p=0.021). Vitamin B12 demonstrated a significant inverse correlation with SLEDAI-2K (R=-0.4744, p=0.0004), whereas folate was not significantly correlated with SLEDAI-2K (R=-0.1008, p=0.48). Neither vitamin B12 (R=-0.1402, p=0.326) nor folate (R=-0.0295, p=0.837) showed a significant association with CLASI. Neither disease duration (Hcy: r=0.2562, p=0.07; B12: R=-0.1598, p=0.26; FA: R=0.0665, p=0.64) nor BMI (Hcy: r=0.0338, p=0.814; B12: R=-0.0915, p=0.52; FA: R=-0.2586, p=0.07) were significantly associated with any of the three biomarkers. Correlation results are summarised in Table 6.

Table 6. Correlation analyses: homocysteine, vitamin B12, and folic acid with disease activity and clinical variables

Variable pair	Coefficient	r / R	p-value
Primary outcomes			
Homocysteine vs CLASI	Pearson	r = 0.3226	p = 0.021
Homocysteine vs SLEDAI-2K	Pearson	r = 0.3195	p = 0.022
Vitamin B12			
Vitamin B12 vs SLEDAI-2K	Spearman	R = −0.4744	p = 0.0004
Vitamin B12 vs CLASI	Spearman	R = −0.1402	p = 0.326
Folic acid			
Folic acid vs SLEDAI-2K	Spearman	R = −0.1008	p = 0.48
Folic acid vs CLASI	Spearman	R = −0.0295	p = 0.837
Clinical determinants of homocysteine			
Age vs homocysteine	Pearson	r = 0.3025	p = 0.031
BMI vs homocysteine	Pearson	r = 0.0338	p = 0.814
Disease duration vs homocysteine	Pearson	r = 0.2562	p = 0.07
Clinical determinants of vitamin B12			
Age vs vitamin B12	Spearman	R = −0.2553	p = 0.07
BMI vs vitamin B12	Spearman	R = −0.0915	p = 0.52
Disease duration vs vitamin B12	Spearman	R = −0.1598	p = 0.26
Clinical determinants of folic acid			
Age vs folic acid	Spearman	R = −0.0515	p = 0.72
BMI vs folic acid	Spearman	R = −0.2586	p = 0.07
Disease duration vs folic acid	Spearman	R = 0.0665	p = 0.64

Discussion

In this cross-sectional study of 51 patients with lupus erythematosus, higher serum homocysteine was associated with greater systemic and cutaneous disease activity. Homocysteine correlated positively with both SLEDAI-2K (r=0.3195, p=0.022) and CLASI (r=0.3226, p=0.021), indicating that patients with more severe cutaneous and systemic involvement had higher circulating homocysteine levels. In parallel, vitamin B12 showed a moderate inverse correlation with systemic activity (R=-0.4744, p=0.0004), whereas folic acid was not significantly associated with either SLEDAI-2K or CLASI. These findings support the concept that disturbances in one-carbon metabolism - reflected by homocysteine and its key nutritional regulators - track with lupus inflammatory burden, including cutaneous severity as measured by a validated instrument.

Its metabolism proceeds either through folate- and vitamin B12-dependent remethylation back to methionine or through vitamin B6-dependent transsulfuration to cysteine [15,16]. The so-called homocysteine hypothesis of atherosclerosis - postulating that even moderately elevated homocysteine levels can drive atherosclerotic progression - was first formulated by McCully in 1969[27], while the first clinical evidence linking pathological homocysteine metabolism to coronary disease was described by Wilcken and Wilcken in 1976[28]. Hyperhomocysteinemia is now broadly recognized as an independent risk factor for coronary, cerebral, and peripheral atherosclerosis[17]. Mechanistically, homocysteine damages arterial cells through the release of cytokines, cyclins, and other mediators of inflammation and cell division. It also induces oxidative stress by impairing cellular respiration, leading to oxidation of low-density lipoproteins and other plaque components. Homocysteine may contribute to endothelial dysfunction through oxidative stress and impaired nitric oxide bioavailability [17,18]. In our cohort, where a majority of patients (52.9%) had hyperhomocysteinemia by the local laboratory threshold (> 13 μmol/L), these vascular mechanisms are directly relevant given the established cardiometabolic burden of CLE.

Beyond its vascular role, homocysteine has been proposed to function as a pro-inflammatory and immune-stimulatory molecule that can contribute to or initiate cellular

injury[19,20]. A bidirectional relationship appears to link homocysteine and the immunoinflammatory activation that characterizes autoimmune diseases: on one hand, immunoinflammatory activation may elevate homocysteine; on the other, elevated homocysteine may amplify inflammatory pathways. Evidence for this bidirectional model has been reported in rheumatoid arthritis and inflammatory bowel disease[20]. Furthermore, homocysteine can trigger autoimmune reactions through its capacity to bind and structurally modify specific proteins, thereby generating neoantigens potentially relevant to the onset or progression of cardiovascular and autoimmune disease[20]. Experimental evidence indicates that homocysteine can activate monocytes and induce the expression of pro-inflammatory cytokines, including IL-1β, IL-6, IL-8, and IL-12 [19]. The significant positive correlations we observed between homocysteine and both CLASI (r=0.3226, p=0.021) and SLEDAI-2K (r=0.3195, p=0.022) are consistent with this immunoinflammatory framework, supporting the concept that homocysteine reflects a shared metabolic–immune state relevant to lupus disease activity.

Our results align with the broader SLE literature. Two independent meta-analyses by Sam *et al*.,[21] and Tsai *et al*.,[22] demonstrated significantly higher serum homocysteine levels in SLE patients compared to healthy controls, suggesting an important role for homocysteine in lupus pathogenesis. Limitations of those analyses included the relatively small number of subjects and insufficient accounting for confounders, such as medications that affect homocysteine metabolism[21,22]. Our cohort similarly reflects the difficulty of controlling for all confounders in a real-world cross-sectional design. Nevertheless, the consistent finding across our study and the existing SLE literature that homocysteine is elevated and correlates with disease activity, strengthens the biological plausibility of this relationship. The mean homocysteine in our cohort (14.48±4.2 μmol/L) exceeded the local upper reference limit, and more than half of patients (52.9%) met criteria for hyperhomocysteinemia, a proportion comparable to rates reported in SLE cohorts.

The association between homocysteine and cutaneous activity is particularly significant given the very limited CLE-specific evidence. To date, only one study has directly evaluated homocysteine in CLE using CLASI as the disease activity measure: Bonciani *et al*.,[23] found that homocysteine levels were higher across different CLE subtypes and correlated positively with CLASI activity, leading those authors to suggest that hyperhomocysteinemia may be linked to the pathogenesis of lupus erythematosus. Our findings are consistent with those of Bonciani et al. and provide additional evidence from an independent cohort for an association between homocysteine and cutaneous and systemic disease activity. This is clinically meaningful because CLASI and SLEDAI-2K capture different, partially overlapping aspects of lupus activity, and a biomarker reflecting both dimensions simultaneously has potential utility across the CLE-SLE spectrum.

Parallels can also be drawn with other inflammatory skin diseases. Studies on psoriasis have reported higher homocysteine levels in patients than in healthy controls[29]; however, unlike our findings in CLE, these studies did not demonstrate a significant correlation between homocysteine levels and psoriasis severity[29]. By contrast, in vitiligo, serum homocysteine has been reported to correlate with disease severity, a finding that provided the rationale for using vitamin B12 and folic acid supplementation in those patients[30]. The positive correlation between homocysteine and CLASI severity in our cohort therefore aligns more closely with the vitiligo model than the psoriasis model, suggesting that in immune-mediated skin diseases driven by significant autoimmune activation (as in CLE and vitiligo), homocysteine may track more directly with disease burden than in conditions where inflammation is primarily keratinocyte-driven.

One proposed mechanism for elevated homocysteine in autoimmune diseases involves the depletion of folate and vitamin B12 due to increased consumption associated with

heightened oxidative stress and active proliferation of immunocompetent cells[31]. This creates a vicious cycle in which declining vitamin B12 levels lead to rising homocysteine, which in turn promotes inflammation and further consumption of folate and vitamin B12. Our data partially support this model: vitamin B12 showed a moderate, statistically significant inverse correlation with SLEDAI-2K (R=-0.4744, p=0.0004), indicating that lower B12 status is associated with greater systemic disease activity. This is consistent with the concept that systemic immune activation may deplete B12 through increased metabolic demand. In contrast, folic acid was not significantly associated with SLEDAI-2K (R=-0.1008, p=0.48) or CLASI (R=-0.0295, p=0.837), nor did either vitamin correlate significantly with CLASI. This divergence between B12 and folate may reflect differential sensitivity to systemic inflammation, differences in dietary intake, or the fact that folate is more readily repleted through diet in this population. Dietary folate intake has been inversely associated with circulating MCP-1 and IL-8 levels in patients with heart failure[32]. However, a randomized controlled trial found that lowering homocysteine did not significantly affect the inflammatory markers assessed [33]. Clinically, our data support systematic assessment of vitamin B12-and to a lesser extent folic acid-as potentially modifiable determinants of the homocysteine–inflammation axis in lupus patients.

Among the clinical determinants of homocysteine examined in this cohort, age showed a significant positive correlation with homocysteine (r=0.3025, p=0.031), consistent with well-established population trends reflecting age-related decline in renal function, reduced dietary intake, and altered vitamin metabolism[34]. Although the dichotomized age-group comparison (≤50 vs >50 years) did not reach statistical significance (p=0.089), patients older than 50 years tended to have higher homocysteine (15.42±4.2 vs**.** 13.42±4.1 μmol/L), and this subgroup analysis was likely underpowered given the modest sample size. Smoking status was not associated with homocysteine (p=0.451) or vitamin B12 (p=0.796), which contrasts with some population-based data showing that smoking is linked to higher homocysteine levels through effects on folate metabolism and oxidative stress. However, folic acid differed significantly across smoking categories (p=0.0145), driven by higher folate levels in former smokers compared to current smokers (post-hoc p=0.0148)-a finding that may reflect behavioral dietary changes following smoking cessation or residual confounding. Neither BMI nor disease duration was significantly associated with homocysteine, vitamin B12, or folate in this cohort, suggesting that in this patient group, age-related and immunoinflammatory factors were more closely linked to homocysteine variability than anthropometric measures or disease chronicity.

Clinical implications and future directions

From a clinical perspective, the observed associations suggest that homocysteine may serve as a readily available adjunct biomarker reflecting both systemic and cutaneous disease activity in lupus erythematosus, and that vitamin B12 status may be relevant to systemic inflammatory burden. Given the emerging evidence linking CLE to increased cardiometabolic risk, these findings further support consideration of homocysteine and nutritional determinants (vitamin B12/folate) within broader risk assessment frameworks, particularly in patients with more active disease. Prospective longitudinal studies are warranted to determine whether changes in homocysteine parallel changes in CLASI and SLEDAI-2K, and whether homocysteine adds predictive value for vascular and cardiometabolic outcomes in CLE and SLE. Interventional studies may also clarify whether correcting vitamin deficiencies meaningfully modifies disease activity or primarily lowers homocysteine without clinical impact.

Conclusion

Homocysteine was positively associated with both systemic and cutaneous disease activity in this cohort of 51 patients with lupus erythematosus, whereas vitamin B12 was

inversely associated with systemic activity. In combination with prior evidence of elevated homocysteine in SLE and limited CLE literature demonstrating an association with CLASI, our findings support homocysteine as a candidate biomarker spanning systemic and cutaneous lupus activity. Prospective longitudinal studies are needed to confirm whether homocysteine tracks with changes in CLASI and SLEDAI-2K over time and whether it adds predictive value for vascular outcomes in CLE/SLE populations.

Conflict of interest statement. None declared.

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