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CD5+ B Lymphocytes in Systemic Lupus Erythematosus Patients: Relation to Disease Activity

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Abstract

Background: Systemic Erythematosus (SLE) is a chronic, complicated and challenging disease to diagnose and treat. The etiology of SLE is unknown, but certain risk factors have been identified that lead to immune system dysregulation with pathogenic autoantibody formation and immune complex deposition.

Am of Study: To assess blood concentration of CD5+ B cells in patients with SLE and to evaluate their relationship with SLE disease activity.

Patients and Methods: The present study included forty SLE patients who were selected from outpatient clinic of Rheumatology and Rehabilitation of Ain Shams University Hospital and diagnosed according to new EULAR and ACR classification criteria. Based on SLEDAI, the patients were selected and divided into two groups. The first group included 20 patients with inactive disease and the second group included 20 patients with active disease. They were matched with ten healthy individuals as a control group, and all were subjected to full history, clinical examination, ESR, CRP, serum complements, anti-dsDNA, ANA, serum creatinine twenty-four hours urinary proteins as well as CD5+ B lymphocytes by flow cytometric analysis.

Results: In the present study, the percentage of CD5+ B lymphocytes per total lymphocytes were significantly decreased in SLE patients compared to healthy individuals. Moreover, the percentage of CD5+ B lymphocytes per total B cells were significantly decreased in SLE patients compared to controls. We also have found a statistically highly significant decrease in the percentage of CD5+ B cells in active SLE patients compared to inactive patients. As regards the correlation studies, the results revealed a positive correlation between CD5+ B cells and each of platelets, C3 and C4. Moreover, the diagnostic performance of CD5+ B cells was evaluated and our results showed that CD5+ B cells can discriminate SLE patients from controls, and can predict the disease activity.

Conclusion: The proportions of CD5+ B cells were significantly decreased in SLE patients than normal people, and were significantly decreased in active SLE patients than inactive ones. These findings denote that CD5+ B cells may have a potential role in preventing autoimmunity development.

Key Words: *Systemic lupus erythematosus – CD5+ B cells – Disease activity.*

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Introduction

SYSTEMIC Lupus Erythematosus (SLE) is a prototypic autoimmune disease with diverse clinical manifestations in association to autoantibodies to components of the cell nucleus **[1]**.

Generalized immune cell abnormalities that involve the B cell, T cell, and monocyte lineages were found in SLE patients **[2]**. These immune cell abnormalities appear to promote B cell hyperactivity which is responsible for the production of an array of autoantibodies with immune-complex deposition and tissue injury **[3]**.

There are conflict data on B cell precursors that generate these autoantibodies. Indeed, numerous studies tried to localize or identify the cells that produce them in SLE patients **[4-6]**.

Peripheral blood B lymphocytes are generally divided into distinct B cell subsets which differ in their cellular markers **[7]**. CD5+ B cells are a small population of B lymphocytes which exhibit unique developmental, phenotypic and functional characteristics that differ from the majority of B cells **[8]**.

The role of CD5+ B cells in health and disease has long been a matter of debate. Although some would indicate that they are the source of these autoantibodies **[9]**, accumulating evidence has shown that high-affinity antibodies to double-stranded DNA (dsDNA) in SLE and Rheumatoid

Factor (RF) in Rheumatoid Arthritis (RA) are derived from CD5-negative B cells and not from CD5+ B cells [10,11].

It has been shown that CD5 + B cells spontaneously secrete natural antibodies which serve in removal of apoptic cells, and counter pathogenic IgG [12]. They also secrete interleukin 10 which has a regulatory effect on different immune population [13]. Furthermore, as long as CD5 co-receptor is expressed on B lymphocytes, the threshold of B-cell receptor is elevated and the cells are maintained in anergy. Subsequently, proliferation and antibodies production are limited [14]. These findings denote that CD5+ B cells have a protective role against autoimmunity, and that their reduced level might predispose to autoimmunity development [15].

Aim of the work:

The aim of the study is to assess blood concentration of CD5+ B cells in patients with SLE and to evaluate their relationship with SLE disease activity.

Patients and Methods

This is a case-control study which was carried out at the outpatient clinic and the inpatient of Physical Medicine and Rheumatology Department, Ain Shams University Hospital, Cairo, Egypt; during the period from May 2018 to January 2019.

The study was approved by the Ethics Committee of the Faculty of Medicine at Ain Shams University. An informed verbal consent was obtained from patients and controls after detailed discussion with him/her to explain the aim and the steps of the study.

Study population:

Patients group: The study included forty SLE patients. Diagnosis of SLE was based on new EULAR and ACR Classification criteria for SLE [16].

Exclusion criteria:

Patients with other associated autoimmune diseases e.g. Hashimoto's thyroiditis. Patients suffer from end-stage organ failure. Pregnant patients. Patients suffer from malignancy. Patients take Drug-Related Lupus (DRL) eg. Chloropromazine, hydralazine, isoniazid, methyldopa, momocycline, and procainamide.

Control group:

10 age and gender matched healthy individuals were randomly recruited to the study as control.

Study measurements:

All patients were subjected to the followings:

Clinical evaluation: Full history taking and complete clinical examination.

Laboratory investigations: CBC complete blood picture assayed by automated coulter. Erythrocyte Sedimentation Rate (ESR) by Westergren bolt method. C-reactive protein by latex method. Complement level (by radial immunodiffusion assay) C3, C4. Anti-double-stranded DNA antibodies (anti-dsDNA) by immune fluorescence. Anti-Nuclear Antibody (ANA) by immune florescence assay. Serum creatinine assayed on autoanalyzer Hitachi 917 and urea. Complete urine analysis. Twenty-four hours' urinary proteins.

Assay for CD5+ B Lymphocytes: By flow cytometric analysis using BECKMAN COULTER, France.

Flow cytometric analysis: Cells preparation and surface staining: Blood collection was performed in sterile Ethylene Diamine Tetra Acetate (EDTA)-filled blood collection tubes. One hundred microliter of blood was taken and added in Falcon tube with 20 gl of PerCp-conjugated anti-CD20 antibodies (clone: L27, BD) and 20 gl PE-conjugated anti-CD5 (clone: UCHT2, BD) and incubated for 20min at 4ºC. After surface staining, lysis of red blood cells was done by using 2ml of the lysis buffer (BD pharmingen) vortex to mix well and incubating for 10min at room temperature in the dark, followed by centrifugation for 5min at a speed of 1500rpm and discarding the supernatant. The pellets were washed twice with 2ml Phosphate Buffer Saline (PBS) then centrifuged for 5min at a speed of 1500rpm. Supernatant was removed and the pellet was resuspended in 500 gl of PBS for acquisition. The cells were acquired and analyzed by (BECKMAN COUTER, France). Lymphocytes were gated depending on both side and forward scatter. From the gated lymphocytes (A) Percentage of CD5+ B cells from total lymphocytes was calculated by using dot plot quadrants from the gated lymphocytes(A). CD5+ B cells were identified as double-positive cells for CD20 and CD5.

Assessment of disease activity:

Disease activity was assessed using Systemic Lupus Erythematosus Disease Activity Index (SLEDAI) [17]. Items were recorded when the

descriptor had been present at the time of the visit or in the preceding 10 days. Based on SLEDAI, patients were selected and divided into two groups. The first group included 20 patients with inactive disease (SLEDAI <4) and the second group included 20 patients with active disease (SLEDAI >4).

Statistical analysis:

The collected data was revised, coded, tabulated and introduced to a PC using Statistical package for Social Science (SPSS 15.0.1 for windows; SPSS Inc., Chicago, IL, 2001). Data was presented and suitable analysis was done according to the type of data obtained for each parameter.

Descriptive statistics:

Mean, Standard Deviation ($\pm$ SD) and range for parametric numerical data, while Median and Interquartile Range (IQR) for non-parametric numerical data. Frequency and percentage of non-numerical data.

Analytical statistics:

Student's "*t*" Test was used to assess the difference between two study group means. ANOVA test was used to assess the difference between more than two study group means. Correlation analysis: To assess the strength of association between two quantitative variables. The correlation coefficient defined the strength and direction of the linear relationship between two variables. The ROC Curve (receiver operating characteristic) provided a useful way to evaluate the Sensitivity and specificity for quantitative diagnostic measures that categorize cases into one of two groups. p-value: Level of significance: p>0.05: Non Significant (NS). p<0.05: Significant (S). p<0.01: Highly Significant (HS).

Results

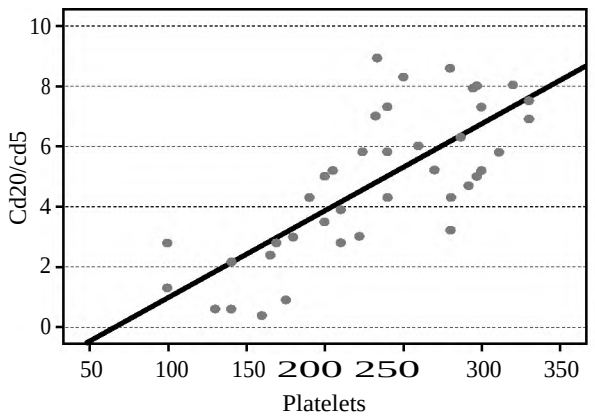

Fig. (1): Correlation between CD20/CD5 B cells of total lymphocytes and platelets.

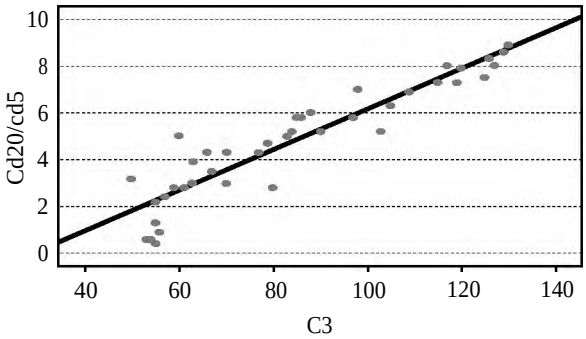

Fig. (2): Correlation between CD20/CD5 B cells of total lymphocytes and C3.

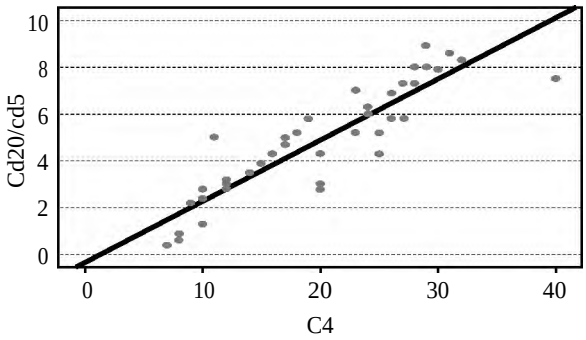

Fig. (3): Correlation between CD20/CD5 B cells of total lymphocytes and C4.

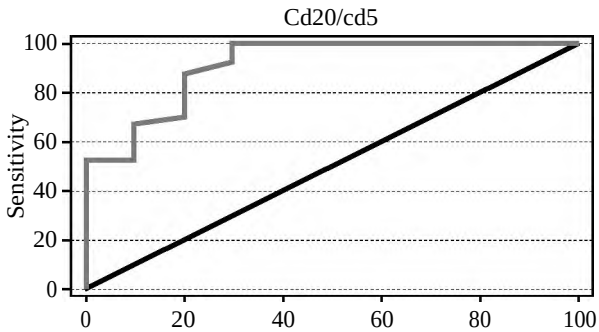

Fig. (4): Roc curve analysis showing the diagnostic performance of CD20/CD5 B cells to distinguish between SLE patients and controls.

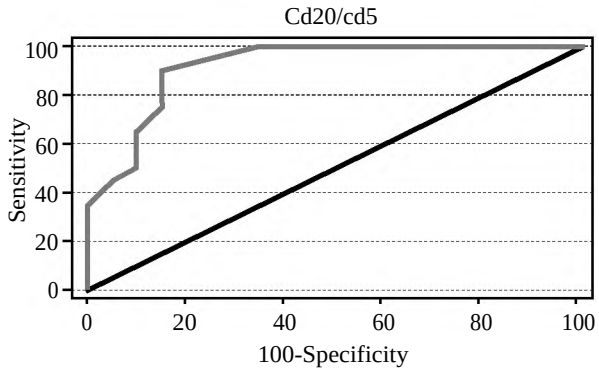

Fig. (5): Roc curve analysis showing the diagnostic performance of CD20/CD5 B cells to distinguish between active and inactive SLE patients.

Table (1): Demographic data in SLE patients and control group.

	Control group No.=10	Patients group No.=40	*p*-value	Sig.
Age:				
Mean ± SD	27.20±8.46	31.88±9.00	0.144	NS
Range	16-40	19-50		
Gender:				
Female	9 (90.0%)	36 (90.0%)	1.000	NS
Male	1 (10.0%)	4 (10.0%)		

Table (2): Clinical manifestations of SLE patients.

System affection	No. %
Constitutional	5 (12.50%)
Mucocutanous	18 (45.00%)
Musculoskeletal	15 (37.5%)
Neurology	1 (2.50%)
Lung affection	1 (2.50%)
Cardiac affection	1 (2.50%)
Renal affection	22 (55%)

Table (3): Laboratory data among the studied groups.

	Control group No.=10	Patient group No.=40	*p*-value	Sig.
TLC ($X10^3/mm^3$):				
Mean ± SD	5.75±0.91	7.36±2.39	0.043	S
Range	4.5-7	3.5-10.8		
Hemoglobin (gm/dl):				
Mean ± SD	12.70±0.92	10.41±1.15	0.001	HS
Range	11.5-14.2	8.5-13		
Platelets ($X10^3$):				
Mean ± SD	273.5±61.47	232.13±63.82	0.071	NS
Range	190-360	99-330		
ESR (mm/hour):				
Mean ± SD	16.5±4.70	54.33±30.04	0.000	HS
Range	11.0-24.0	10-120		
CRP (mg/dl):				
Mean ± SD	3.3±1.5	10.18±4.31	0.041	S
Range	1.5-6.3	5-15		
Anti-ds DNA:				
No. %	0 (0.0%)	26 (65.0%)	0.000	HS
ANA:				
No. %	0.0 (0.0%)	40 (100.0%)	0.000	HS
C3 (mg/dl):				
Mean ± SD	125.55±12.64	84.65±26.12	0.000	HS
Range	100-140	50-130		
C4 (mg/dl):				
Mean ± SD	35.73±11.82	19.75±8.35	0.008	HS
Range	20-50	7-40		
Urea (mg/dl):				
Median (IQR)	17.12 (14.98-21.4)	34.24 (19.26-60.03)	0.003	HS
Range	14.98-25.54	20-112		
Creatinine (mg/dl):				
Median (IQR)	0.6 (0.5-0.6)	0.9 (0.6-1.4)	0.007	HS
Range	0.3-0.7	0.5-2.2		
24h urine protein (mg/dl):				
Median (IQR)	90 (80-95)	123.8 (100-1078)	0.006	HS
Range	89-100	100-2082		

TLC : Total Leukocyte Count. Hgb : Hemoglobin.
ESR : Erythrocyte Sedimentation Rate. C3 : Complement 3.
Anti-ds DNA : Anti double stranded DN. C4 : Complement 4.

Table (4): Proportions of lymphocytes in SLE patients and controls.

	Control group No.=10	Patient group No.=40	*p*-value	Sig.
Total lymphocytes %:				
Mean ± SD	27.43±263	21.01±5.92	0.007	HS
Range	22.9-39.0	12.2-31.2		
B lymphocytes %:				
Mean ± SD	20.1±2.5	10.5±5.7	0.000	HS
Range	17.6-22.6	4.8-16.2		
CD20/CD5 B cells (%) of B lymphocytes:				
Mean ± SD	39.00±10.5	30.7±9.3	0.018	S
Range	28.5-50.5	21.4-40		
CD20/CD5 B cells (%) of total lymphocytes:				
Mean ± SD	11.1±4.9	4.80±2.42	0.000	HS
Range	5.09-16	0.4-8.9		

Table (5): Renal biopsy from lupus nephritis patients (n=22).

	N	%
Renal biopsy:		
Class II	5	23
Class III	7	32
Class IV	10	45

Table (6): SLEDAI in patient group.

	Patients group No.=40
SLEDAI:	
Median (IQR)	3.5 (2-8)
Range	0-20

Table (7): Comparison between inactive group and active groups as regard demographic data.

	Inactive group No.=20	Active group No.=20	*p*-value	Sig.
Age:				
Mean ± SD	32.75±8.76	31.00±9.39	0.288	(>0.05)
Range	20-48	19-50		NS
Gender:				
Female	16 (80.0%)	20 (100.0%)	0.108	(>0.05)
Male	4 (20.0%)	0 (0.0%)		NS
Duration:				
Median (IQR)	3.5 (2-10)	6.00 (3-10.5)	0.541	(>0.05)
Range	1-20	1-15		NS

Table (8): Comparison between active and inactive groups as regard system affection.

	Inactive group		Active group		*p*-value	Sig.
	No.	%	No.	%		
System affected	20	100.0	20	100.0		
Constitutional	0	0.0	5	25.0	0.017	(<0.05) S
Mucocutaneous	6	30.0	12	60	0.269	(>0.05) NS
Neurology	0	0.0	1	5.0	0.311	(>0.05) NS
Musculoskeletal	5	25.0	10	50.0	0.102	(>0.05) NS
Lung affection	0	0.0	1	5.0	0.311	(>0.05) NS
Cardiac affection	0	0.0	1	5.0	0.311	(>0.05) NS
Renal affection	12	60.0	10	50.0	0.069	(>0.05) NS

Table (9): Comparison between active and inactive groups as regard laboratory data.

	Inactive group No.=20	Active group No.=20	*p*-value	Sig.
TLC (X 10^3/mm^3):				
Mean ± SD	8.31±1.95	6.41±2.45	0.005	(<0.01)
Range	4.5-10.8	3.5-10.5		HS
Hemoglobin (gm/dl):				
Mean ± SD	10.79±1.13	10.03±1.07	0.030	(<0.05)
Range	8.9-13	8.5-12.5		S
Platelets (X10^3):				
Mean ± SD	253.15±56.98	211.10±64.68	0.021	(<0.05)
Range	160-33	99-311		S
Urea (mg/dl):				
Median (IQR)	30.26 (17.12-25.68)	53.5 (40.5-86.5)	0.003	(<0.01) HS
Range	20-60.64	25-112		
Creatinine (mg/dl):				
Mean ± SD	0.88±0.28	1.33±0.43	0.000	(<0.01)
Range	0.5-1.1	0.6-2.2		HS
ESR (mm/hour):				
Mean ± SD	27.50±20.04	30.33±19.00	0.239	(>0.05)
Range	13-110	10-120		NS
CRP (mg/dl):				
Mean ± SD	8.10±1.02	9.05±2.05	0.055	(>0.05)
Range	5-10	6-15		NS
C3 (mg/dl):				
Mean ± SD	104.95±20.01	64.35±11.49	0.000	(<0.01)
Range	66-130	50-86		HS
C4 (mg/dl):				
Mean ± SD	26.60±4.75	12.90±4.66	0.000	(<0.01)
Range	20-40	7-25		HS
ANA:				
No. %	20 (100%)	20 (100%)	NA	NA
Anti-dsDNA:				
No. %	9 (45%)	17 (85%)	0.008	(<0.01) HS

TLC : Total Leukocyte Count. C3 : Complement 3.
ESR : Erythrocyte Sedimentation Rate. C4 : Complement 4.
Anti-ds DNA : Anti double stranded DN. NA: Not Applicable

Table (10): Comparison between active and inactive groups as regard proportions of lymphocytes.

	Inactive group No.=20	Active group No.=20	*p*-value	Sig.
Lymphocytes %:				
Mean ± SD	23.45±5.61	18.57±5.29	0.004	(<0.0)
Range	13-31.2	12.2-27.7		(HS)
B lymphocytes %:				
Mean ± SD	14.3±3.8	11.2±6.6	0.048	<0.05
Range	9.6-16.2	4.8-17.1		(S)
CD20/CD5 B cells % of B Lymphocytes:				
Mean ± SD	31.7±11.2	23.5±10.8	0.024	<0.05
Range	21.4-42.9	18.1-40		(S)
CD20/CD5 B cells % of total Lymphocytes:				
Mean ± SD	6.51±1.75	3.10±1.69	0.000	<0.01
Range	2.8-8.9	0.4-5.8		(HS)

Table (11): Comparison between active and inactive groups as regard classes of lupus nephritis using Chi-square test.

	Inactive group		Active group		*p*-value	Sig.
	No.	%	No.	%		
Renal biopsy:						
Class II	5	41.6	0	0.0	0.007	<0.01
Class III	4	33.3	3	30.0		(HS)
Class IV	3	25.0	7	70.0		

Table (12): Comparison between active and inactive groups as regard SLEDAI.

	Inactive No.=20	Active No.=20	*p*-value	Sig.
SLEDAI:				
Median (IQR)	2.00(2-2)	8.00(6-14)	0.000	(<0.1)
Range	0-3	4-20		HS

Table (13): Correlation between CD20/CD5 B cells % of total lymphocytes and age and disease duration.

	Cd20/cd5		
	r	*p*-value	Sig.
Age	–0.279	0.054	>0.05 (NS)
Duration	–0.229	0.062	>0.05 (NS)

Table (14): Relation between CD20/CD5 B cells % of total lymphocytes and gender of patients.

Gender	Cd20/cd5		*t*-test	*p* value	Sig.
	Mean ± SD	Range			
Female	4.43±2.26	0.40-8.6	3.193	0.003	<0.01
Male	8.10±0.67	7.3-8.9			(HS)

Table (15): Relation between CD20/CD5 B cells % of total lymphocytes and system affection.

	Cd20/cd5		*t*-test	*p*-value	Sig.
	Mean ± SD	Range			
Musculoskeletal:					
Negative	5.87±1.37	1.30-8.90	1.679	0.111	(>0.05)
Positive	5.42±2.44	0.40-8.00			NS
Neurology:					
Negative	4.91±2.35	0.40-8.90	1.806	0.051	(>0.05)
Positive	1.60±0.00	1.60-1.60			NS
Mucocutaneous:					
Negative	6.06±2.08	2.40-8.90	1.843	0.120	(>0.05)
Positive	5.66±1.74	0.40-7.30			NS
Constitutional symptoms:					
Negative	3.55±1.62	0.40-8.90	1.283	0.071	(>0.05)
Positive	2.74±1.20	0.60-3.50			NS
Lung affection:					
Negative	4.71±2.45	0.40-8.90	1.889	0.060	(>0.05)
Positive	2.00±0.00	2.00-2.00			NS
Cardiac affection:					
Negative	4.67±2.36	0.40-8.90	1.900	0.056	(>0.05)
Positive	1.80±0.00	1.80-1.80			NS
Renal affection:					
Negative	4.26±2.13	0.60-8.90	2.099•	0.152	(>0.05)
Positive	4.75±2.03	0.40-8.60			NS

Table (16): Correlation between CD20/CD5 B cells % of total lymphocytes and patient labs.

	Cd20/cd5		
	r	*p*-value	Sig.
TLC ($X10^3/mm^3$)	+ 0.200	0.071	>0.05 (NS)
Hemoglobin (gm/dl)	+0.262	0.057	>0.05 (NS)
Platelets ($X10^3$)	0.757**	0.000	<0.01 (HS)
Urea (mg/dl)	–0.073	0.209	>0.05 (NS)
Creatinine (mg/dl)	–0.171	0.181	>0.05 (NS)
ESR (mm/hour)	–0.054	0.242	>0.05 (NS)
CRP (mg/dl)	–0.231	0.060	>0.05 (NS)
C3 (mg/dl)	+0.748	0.000	<0.01 (HS)
C4 (mg/dl)	+0.723	0.000	<0.01 (HS)
Total lymphocytes %	+0.630	0.000	<0.01 (HS)
B lymphocytes %	+0.622	0.000	<0.01 (HS)

TLC: Total Leukocyte Count. C3: Complement 3.
ESR: Erythrocyte Sedimentation Rate. C4: Complement 4.

Table (17): Correlation between CD20/CD5 B cells % of total lymphocytes and Anti-ds DNA, ANA.

	Cd20/cd5		*t*-test	*p*-value	Sig.
	Mean ± SD	Range			
Anti-ds-DNA:					
Negative (n=14)	5.81±2.71	0.4-8.90	2.019	0.061	<0.05
Positive (n=26)	4.25±2.11	0.6-8			(NS)
ANA:					
Negative (n=0)			NA	NA	NA
Positive (n=40)	4.80±2.42	0.40-8.90			

Anti-ds DNA : Anti double stranded DNA.
ANA : Anti-Nuclear Antibody.
NA : Not Applicable.

Table (18): Relation between CD20/CD5 B cells % of total lymphocytes and renal biopsy.

	Cd20/cd5		ANOVA test	*p*-value	Sig.
	Mean ± SD	Range			
Renal biopsy:					
Class II (n=5)	6.95±1.44	5.30-8.60	1.061	0.056	(>0.05)
Class III (n=7)	4.27±2.00	0.40-8.00			NS
Class IV (n=10)	4.01±1.08	1.40-6.30			

Table (19): Correlation between CD20/CD5 B cells % of total lymphocytes and SLEDAI.

	Cd20/cd5		
	r	*p*-value	Sig.
SLEDAI	–0.727	0.000	<0.01 (HS)

SLEDAI: Systemic Lupus Erythematosus Disease Activity Index.

Discussion

SLE is an idiopathic connective tissue disease with a spectrum covering wide array of clinical manifestations [1]. The etiology is multifactorial and B lymphocyte hyperactivity is thought to be one of these multiple factors [18]. Previously, it was believed that CD5+ B lymphocytes are the source of auto antibodies production and that their levels are increased in the blood of SLE patients [19]. Accumulating evidence suggests that CD5 negative B lymphocytes are the ones that produce these auto antibodies and not CD5+ B cells [20]. Many studies have revealed that CD5+ B cells have a role in preventing autoimmunity by raising the threshold required for activation of self-reactive B cells [21]. Also, CD5+ B cells are shown to produce high levels of IL-10 and natural IgM with low affinity and poly-reactivity for autoantigens that have a role in removing auto-antigens and apoptotic cells [20]. Therefore, these regulatory B cells are considered as inhibitors of inflammation and autoimmune responses [10].

Whether CD5+ B lymphocytes increase or decrease in the blood of SLE patients is still a controversial issue which we tried to find out by comparing CD5+ B cells in 40 SLE patients and 10 healthy controls. The forty patients were divided into active and inactive groups. The two groups were compared as regard CD5+ B cells to assess its relation to disease activity.

Our patients were 36 females (90%) and 4 males (10%), their age ranged from 19 to 50 years old with a mean of 31.88 ±9.00. These findings were in agreement with [22] Ginzler et al., (2015) who reported that about 90% of SLE cases occur in women frequently starting at childbearing age.

Most of our patients were females; as sex status is obviously of great importance in susceptibility to SLE, which is predominantly a disease of women, particularly during their reproductive years [23].

Essentially any organ system can be affected in SLE such as mucocutaneous, musculoskeletal, renal, Central Nervous System (CNS), pulmonary and cardiac systems [23].

In our study, (45%) of patients were found to have mucocutaneous manifestations in the form of malar rash, oral ulcers, discoid lupus, alopecia or photosensitivity. This was less than the percentages reported by El-Sadek (2002) [24], Bujan et al., (2003) [25], Carter et al., (2005) [26], and Buyon (2008) [23] who reported cutaneous manifestation in 85%, 75%, 80% and 85% of their patients, respectively.

Arthralgia was found in 37.5% of our patients. This percentage was lower than that reported by Carter et al., (2005) who found that 56% of his studied SLE patients' samples had arthralgia [26]. Buyon (2008), also reported a higher percentage than ours (76% to 100%) [23].

Only one patient (2.5%) had neurological affection in the form of severe lupus headache. This was lower than the percentage reported by Zakaria (2004) who found neuropsychiatric manifestation in 50% of his patients [27], and lower than results reported by Annese et al., (2006) who noted that 10% of his lupus patients had suffered from headache only [28].

Of the forty patients, only one patient (2.5%) had pleural effusion. This was lower than the results reported by Zakaria et al., (2004) [27] and Buyon (2008) [23] as the pleural effusion was found in 45% and 30% of their patients respectively. This difference could be explained by the smaller number of patients included in our study.

Cardiac affection in the form of pericarditis was found in only one patient (2.5%). This was lower than the percentage reported by Buyon (2008) where he supposed that percentage of cardiac affection in SLE patients is 45% [23].

Renal affection in our study was found to be in 55% of our patients. This come in line with the percentage reported by Tassiulas I.O., and Boumpas D.T. (2009) who stated a percentage of 40-70% [29], but lower than percentage reported by Buyon (2008) who stated a percentage of 65% [23].

The difference between their results and ours concerning clinical manifestations may be due to the small number of patients included in the study.

Our results have demonstrated a statistically highly significant difference between patient and healthy groups as regard ESR ($p<0.01$), but no statistically significant difference was found between active and inactive groups ($p=0.239$). These results are consistent with those reported by Haq et al., (2002) who noted that ESR test cannot distinguish a lupus flare from an infection and the level doesn't directly correlate with lupus disease activity [30]. Our results also come in accordance with the results reported by Buyon (2008) who stated that ESR is frequently elevated in SLE and is generally not considered reliable marker of clinical activity [23]. Therefore, they think that ESR may not be a useful test for monitoring SLE disease activity.

Moreover, Akbarian et al., (2009) stated that ESR showed a weak correlation to disease activity and was influenced by a multiplicity of factors [31].

We found a statistically significant difference between patients and controls as regard the CRP ($p=0.041$). No statistically significant difference was found between active and inactive groups ($p=0.055$). Our results are consistent with Williams et al., (2005) who stated that very high proportion of uninfected lupus patients was found to have clinically significant CRP elevations, which did not, however, correlate with disease activity [32].

Our results are different from Ter Borg EJ et al., (2000) who noted that CRP is not usually elevated in SLE patients even in the disease activity unless infection or serositis has been established [33]. On the other hand, Mok et al., (2003) reported that CRP is elevated with activity of lupus and positively correlates with lupus disease activity index [34].

Each of the cellular elements of blood can be affected in SLE. Accordingly, CBC is a critical part in the diagnosis and continued evaluation of all lupus patients [23]. In our study hemoglobin level ranged from 8.5 to 13 with a mean of 10.41 ± 1.15, and a statistically highly significant difference was found between patients and controls ($p<0.01$). These findings are in agreement with Sharaf El Din (2002) [35] and Zakaaria (2004) [27] who stated that anemia is a clinical feature in most of SLE patients. Also, Buyon (2008) noted that SLE patients may have non-specific anemia reflecting the chronic condition of the disease [23]. Furthermore, we have found a statistically significant difference between active and inactive groups ($p=0.030$), and this come in accordance with Stojan et al., (2013) who reported that anemia is associated with disease activity [36].

As regard the Total Leucocytic Count (TLC), we recorded a statistically significant difference between patients and healthy groups ($p=0.043$), and a highly statistically significant difference between active and inactive patients ($p=0.005$). These finding are consistent with Stojan et al., (2013) who reported that leucopenia is common in SLE patients and may reflect active disease [36].

As regard platelets count, our results revealed that there is a statistically significant difference between active and inactive groups ($p=0.021$) which are in agreement with Amaylia et al., (2013) who reported that thrombocytopenia is associated with disease activity [37].

Anti-dsDNA, a protein directed against double-stranded DNA, plays a role in SLE pathogenesis, especially in kidney damage. The detection of anti-dsDNA antibodies in the circulation of patients is one of the major criteria for diagnosis of SLE. Moreover, exacerbation of the disease is proceeded

by increasing anti-DNA levels and the development of lupus nephritis. Lupus nephritis is one of the most serious complications and correlates strongly with the presence of high avidity anti-DNA [38].

The preceding note complies with our results where anti-dsDNA were negative in all our healthy individuals and positive in 65% of our patients. This percentage is close to percentages reported by Svenungsson E. et al., (2003) [39] and Buyon (2008) [23] where they have detected anti-ds DNA in 60% of their patients, but lower than those found by El Dafrawy (2003) [40] and Bujan et al., (2003) [25] (100%), and higher than those found by Doria et al., (2005) (47%) [41]. The difference between all these results may be related to the different number of patients included in each study and the sensitivity of the laboratory method used. Furthermore, we have found a statistically highly significant difference between active and inactive groups as regard the Anti-ds DNA (p=0.008). This was in agreement with Abd-Elhafeez et al., (2017) who reported that rising titers of Anti-ds DNA can be used to confirm SLE disease activity [42].

It is of well-known that ANA test is the most sensitive diagnostic test for confirming the diagnosis of SLE. Indeed, all our patients were positive for ANA test (100%). This percentage is the same found by El Dafrawy (2003) [40] and Bujan et al., (2003) [25], and close to that found by Buyon (2008) (more than 90%) [23].

Complement component 3, often simply called C3, is a protein of the immune system and plays a central role in the innate immunity and complement system. Another protein involved in the intricate complement system is complement component 4 (C4) which originate from the Human Leukocyte Antigen (HLA) system and serves a number of critical functions in immunity, tolerance, and autoimmunity. The cause of complement depletion in SLE is the formation of immune complexes, which in turn activate complement, predominantly by means of the classical pathway leading to complement consumption. Complement activation is normally measured in clinical practice by estimation of levels of both C3 and C4 [43]. In the current study, the complement levels were lower in patients than controls. We recorded a highly statistically significant difference between patients and controls regarding C3 and C4 as (p= 0.000, 0.008 respectively) and a highly statistically significant difference between active and inactive groups regarding C3 and C4 as (p<0.01). Our results are in agreement with Nived O., and Sturfelt G. (2004) who noted that low complement concentrations and high complement system activation are characteristic findings in active SLE [44].

Regarding serum creatinine and urea, a highly statistically significant difference was found between patients and controls (p=0.007, 0.003 respectively). Impaired renal function in our patients could be due to the result that 55% of our patients are lupus nephritis. We also recorded a highly statistically significant difference between active and inactive groups regarding creatinine and urea as (p=0.000, 0.003 respectively). Our findings are consistent with Borchers A. et al., (2012) who reported that elevated renal function tests are signs of SLE activity [45].

Renal biopsies were taken from lupus nephritis patients and revealed that 23% were class II, 32% were class III, 45% were class IV. Our results are consistent with Tassiulas I.O. and Boumpas D.T. (2009) who reported that class IV is the most frequent pathologic type in lupus nephritis patients [29].

Disease activity was assessed using Systemic Lupus Erythematosus Disease Activity Index (SLEDAI). SLEDAI score ranged from 0 to 20 with a median (IQR) of 3.5 (2-8).

The study was conducted on 20 inactive and 20 active patients. There was a highly statistically significant difference between the two groups regarding SLEDAI score as (p<0.01).

As regard the proportion of lymphocytes in our patients, total lymphocytes percentage ranged from 12.2 to 31.2 with a mean of 21.01 ±5.92%. There was a highly statistically significant difference between patients and controls as (p=0.007) and between active and inactive ones (p=0.004). Our results are consistent with Amaylia et al., (2013) who reported that lymphopenia is common in SLE patients and it's a chief finding in lupus fluctuations reflecting case activity [37].

Percentage of total B lymphocytes were highly significantly decreased in our patients compared to controls as (p<0.01). Furthermore, we found a statistically significant difference between active and inactive patients as (p=0.048). Our results are consistent with Odendahl et al., (2000) who reported that SLE patients had significant B cell lymphopenia with some disturbances and impairments in all B cell types which are naïve, memory B cells and plasma cells [46].

We assessed the percentage of CD20+/CD5+ B lymphocytes per total B cells, and they were

significantly decreased in SLE patients (30.7 ±9.3) % compared to controls (39.00±10.5) % (p=0.018).

We also assessed the percentage of CD20+/ CD5+ B lymphocytes per total lymphocytes, and it was significantly decreased in SLE patients (4.80 ±2.42) % compared to healthy individuals (11.10± 4.9) % (p<0.01).

These findings are consistent with the data obtained from Vernino L.A. et al., (1992) who demonstrated that the percentage of CD5 + B cell was 24% in ten normal subjects and it was about 17% in 16 SLE patients [47].

Our results also come in accordance with Hassan et al., (2017) who found that the percentage of CD5 + B cell was 10.8% in 100 normal subjects and it was about 4.1% in 100 SLE patients [48].

Unlike our findings, Markeljevic et al., (1994) measured the percentage of CD5-expressing B cells in peripheral blood of 28 SLE patients in the remission phase and found that relative to healthy control subjects, the blood CD5+ B cell subset tended to be elevated in SLE patients [49]. However, they did not measure the percentage of CD5-expressing B cells in active SLE patients which we think that it would add valued data about the proportion of CD5-expressing B cells in the entire SLE patients and not in patients in the remission phase only.

On the other hand, Böhm in 2004, conducted a study on 24 SLE patients which showed that SLE patients had increased percentages of CD5+ B cells compared to controls. However, patients included in his study were mainly cutaneous lupus patients [50]. The differenc between our results and those reported by Böhm may be explained by the difference in the affected systems.

Garaud et al., (2009) reported that the percentage of CD5+ B cell was similar in both 25 healthy control and 25 SLE patients. However, they measured the membrane density of CD5 on B cells and it was lower in SLE patients than controls [51]. The decreased membrane density of CD5 on B cells in SLE goes in accordance with the new concept that membrane CD5 elevates the threshold of BCR mediated signaling and prevents B lymphocyte expansion and auto antibody production [18].

We reported a statistically highly significant decrease in the percentage of CD20/CD5+B cells per total lymphocytes in active SLE patients (3.10 ± 1.69) % compared to inactive patients (6.51 ±1.75) % (p<0.01). Our findings support that decreased CD5 B cells may have a role in disease activity.

In contrast to our results, Ebo et al., (1994) did not find any association between CD5+ B cells and SLE activity [52]. This difference between our results and those reported might be explained by the difference in the criteria used for diseases activity categorization for SLE patients.

We also found a positive correlation between CD5+ B cells % of total lymphocytes and platelets (r=0.757 * *, p<0.01).

We have found a positive correlation between CD5+ B cells of total lymphocytes and each of C3 (r=0.748 * *, p<0.01) and C4 (r=0.723 * *, p<0.01). Our results are in agreement with Hassan et al., (2017) who reported a significant positive correlation between CD5 B cells of total lymphocytes and both of C3 and C4 [48]. The positive correlation with complement levels may be due to the role of complement receptors in selection, expansion, and maintenance of B-1 cells [53]. Also, B1 cells are positively selected in early development by cognate antigens and that interaction requires complement receptors.

Our results revealed a statistically highly significant correlation between CD20/CD5+ B cells of total lymphocytes and each of percentage of total lymphocytes and percentage of B lymphocytes as (r=0.630**, 0.622** respectively) (p<0.01) our results are different from results reported by Hassan et al., who did not find significant correlation between CD5+ B cells of total lymphocytes and any of them [48].

We did not find a significant correlation between CD5+ B cells and age of patients, duration of the disease, ESR, CRP, other laboratory characteristics or system affection.

Conclusion:

In this study, CD5+ B cells were significantly decreased in SLE patients than control group, and highly significantly decreased in active SLE patients than inactive subgroup.

An important conclusion is that CD5+ B cells may have a potential role in preventing autoimmunity development and could be used as marker of SLE disease activity.

Thus, our results might be important as they suggest a novel approach to the clinical management of lupus patients.

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References

1- D'CRUZ D., KHAMASHTA M. and HUGHES G.: Systemic lupus erythematosus. Lancet, 369: 587-96, 2007.

2- MAGEED R. and PRUD'HOMME G.: Immunopathology and the gene therapy of lupus. Gene Therapy, 10: 861-74, 2003.

3- CHAN O.T., MADAIO M.P. and SHLOMCHIK M.J.: The central and multiple roles of B cells in lupus pathogenesis. Immunol. Rev., 169: 107-21, 1999.

4- ODENDAHL M., JACOBI A., HANSEN A., et al.: Disturbed peripheral B lymphocyte homeostasis in systemic lupus erythematosus. J. Immunol., 165: 5970-9, 2000.

5- JACOBI A.M., ODENDAHL M., REITER K., et al.: Correlation between circulating CD27 high plasma cells and disease activity in patients with systemic lupus erythematosus. Arthritis Rheum., 48: 1332-42, 2003.

6- CHANG N.H., McKENZIE T., BONVENTI G., et al.: Expanded population of activated antigen-engaged cells within the naive B cell compartment of patients with systemic lupus erythematosus. J. Immunol., 180: 1276-84, 2008.

7- KANTOR A.B. and STALL A.M.: Differential development of progenitor activity for three B-cell lineages. Proc. Natl. Acad Sci., 89: 3320-4, 2000.

8- BERLAND R. and WORTIS H.H.: Origins and functions of B-1 with notes on the role of CD5. Annu. Rev. Immunol., 20: 253-300, 2002.

9- MURAKAMI M., YOSHIOKA H., SHIRAI T., et al.: Prevention of autoimmune symptoms in autoimmune-prone mice by elimination of B-1 cells. Int. Immunol., 7 (5): 877-82, 1999.

10- BOUAZIZ J., YANABA K., TEDDER T., et al.: Regulatory B cells as inhibitors of immune responses and inflammation. Immunol. Rev., 224: 201-14, 2008.

11- REID R.R., WOODCOCK S., SHIMABUKURO-VORNHAGEN A., et al.: Functional activity of B-1 a cells. J. Immunol., 187: 8403-5340, 2002.

12- HUREZ V., KAZATCHKINE M.D., VASSILEV T., et al.: Pooled normal human polyspecific IgM contains neutralizing anti-idiotypes to IgG autoantibodies of autoimmune patients and protects from experimental autoimmune disease. Blood, (10): 4004-13, 1997.

13- BURDIN N., ROUSSET F. and BANCHEREAU J.: B-cell-derived IL-10: Production and function. Methods, 11: 981-12, 2007.

14- YOUINOU P.: Renaudineau Y.CD5 Expression in B cells from patients with systemic lupus erythematosus. Crit. Rev. Immunol., 31: 31-42, 2011.

15- BURDIN N., ROUSSET F. and BANCHEREAU J.: B-cell-derived IL-10: Production and function. Methods, 11: 981-12, 2007.

16- ARINGER M., COSTENBADER K., DAIKH D., et al.: Ann. Rheum. Dis., 76 (S2): 454-7, 2017.

17- ISENBERG D., APPEL G.B. and CONTRERAS G.: SLE activity measures. Rheumatology (Oxford), 49 (1): 128-40, 2010.

18- RENAUDINEAU Y., HILLION S., SARAUX A., et al.: An alternative exon 1 of the CD5 gene regulates CD5 expression in human B lymphocyte. Blood, 106: 2781-9, 2005.

19- SMITH H.R.: Olson RR.CD5+ B lymphocytes in systemic lupus erythematosus and rheumatoid arthritis. J. Rheumatol., 17: 833-5, 1990.

20- DUAN B. and MOREL L.: Role of B-1a cells in autoimmunity. Autoimmun. Rev., 5: 403-8, 2006.

21- HIPPEN K.L., TZE L.E. and BEHRENS T.W.: CD5 maintains tolerance in anergic B cells. J. Exp. Med., 191: 883-90, 2000.

22- GINZLER E. and TAYAR J.: Fast Facts of Lupus. J. Immunol., 150 (12): 730-3, 2015.

23- Buyon J.P.: Systemic lupus erythematosus. In: Klippel, J.M., Stone, J.H., Crofford, L.J. et al., Primer on the Rheumatic disease, 13th edition. Atlanta, Georgia, Arthritis foundation, 2008.

24- EL SADEK M.A., MAURI M., MATAS L., et al.: Anti Topoisomerase one antibodies in systemic lupus erythematosus patients. Thesis submitted for master degree to Faculty of Medicine, Ain Sham University, 2002.

25- BUJAN S., ORDI-ROS J., PAREDES J., et al.: Contribution of the initial features of systemic lupus erythematosus to the clinical evolution and survival of a cohort Mediterranean patients. Ann. Rheum. Dis., 62 (9): 859-65, 2003.

26- CARTER R., ZHAO H., LIU X., et al.: Expression and occupancy of BAFF-R on B cells in systemic lupus erythematosus. Arthritis Rheum., 52: 3943-54, 2005.

27- ZAKARIA M.A., D'AGOSTINI S., FERRACCIOLI G., et al.: Early detection of articular inflammatory activity in patients with systemic lupus erythematosus using Power Doppler Ultrasonography. Thesis submitted for master degree to Faculty of Medicine, Ain Shams University, 2007.

28- ANNESE V., TOMIETTO P., VENTURINI P., et al.: Migraine in SLE: Role of antiphospholipid antibodies and Raynaud's phenomenon. Reumatismo., 58: 50-8, 2006.

29- TASSIULAS I.O. and BOUMPAS D.T.: Clinical picture of systemic lupus erythematosus, In Harris, E. D (ed.), Kelly's textbook of rheumatology. 8th ed. Philadelphia, Elsevier, 20093.

30- HAQ I. and ISENBERG D.A.: How does one assess and monitor patients with systemic lupus erythematosus in daily clinical practice? Best Pract. Res. Clin. Rheumatol., 16: 181-94, 2002.

31- AKBARIAN M., GHARIBDOOST F., HADJALILOO M., et al.: Assessment of Serum Thrombomodulin in Patients with Systemic Lupus Erythematosus in Rheumatology Research Center. Acta Medica Iranica., 47 (2): 97-102, 2009.

32- WILLIAMS R.C., HARMON M.E., BURLINGAME R., et al.: Studies of serum C-reactive protein in systemic lupus erythematosus. J. Rheumatol., 32: 454-61, 2005.

33- TER BORG E.J., HORST G., LIMBURG P.C., et al.: C-reactive protein levels during disease exacerbations and infections in systemic lupus erythematosus: A prospective longitudinal study. J. Rheumatol., 17: 1642-8, 2000.

34- MOK C.C. and LAU C.S.: Pathogenesis of systemic lupus erythematosus. J. Clin. Pathol., 56: 481-, 2003.

35- SHARAF EL DIN A.R., GAD-ALLAH N.A., ABDEL HAKIM M.R., et al.: Serum leucine aminopeptidase level in systemic lupus erythematosus and rheumatoid arthritis. Thesis submitted for Faculty of Medicine, Ain Shams University, pp. 56, 2002.

36- STOJAN G., FANG H., MAGDER L., et al.: Erythrocyte sedimentation rate is a predictor of renal and overall SLE disease activity. Lupus, 22: 827, 2013.

37- AMAYLIA O.E., HENDARSYAH S., SUMARTINI D., et al.: The Role of Neutrophil Lymphocyte Count Ratio as an Inflammatory Marker in Systemic Lupus Erythematosus, Acta Medica Indonesiana. The Indonesian Journal of Internal Medicine, 170-4, 2013.

38- CHAIM P. and PUTTERMAN A.: New approaches to the renal pathogenicity of anti-DNA antibodies in systemic lupus erythematosus: A diagnostic and prognostic tool for systemic lupus erythematosus. MERONI1 Journal of Anti-DNA antibodies, 3 (2): 7-11, 2004.

39- SVENUNGSSON E., GUNNARSSON I., FEI G., et al.: Elevated triglycerides and low levels of high-density protein as markers of disease activity in association with up-regulation of the tumor necrosis factor alpha/tumor necrosis factor receptor system in systemic lupus erythematosus. Arthritis Rheum., 48 (9): 2533-40, 2003.

40- EL DAFRAWY D.M.: Antinucleosome IgG antibodies in systemic lupus erythematosus: Correlations with disease activity and lupus nephritis. Thesis submitted for master degree to Faculty of Medicine, Ain Shams University, 2003.

41- DORIA A., IACCARINO L., SARZI-PUTTINI P., et al.: Cardiac involvement in systemic lupus erythematosus. Lupus, 14: 683-6, 2005.

42- ABD-ELHAFEEZ H.A., EL-MEGHAWRY E.S., OMRAN T.M., et al.: Study of Neutrophil Lymphocyte Ratio and Platelet Lymphocyte Ratio as Inflammatory Markers in Systemic Lupus Erythematosus Egyptian Patients. Electronic J. Biol., 13: 3, 2017.

43- BIRMINGHAM D., IRSHAID F., NAGARAJA H., et al.: The complex nature of serum C3 and C4 as biomarkers of lupus renal flare. Lupus, 19 (11): 1272-80, 2010.

44- NIVED O. and STURFELT G.: ACR classification criteria for systemic lupus erythematosus: Complement components. Lupus, 13: 1-3, 2004.

45- BORCHERS A., LEIBUSHOR N., NAGUWA S., et al.: Lupus nephritis: A critical review. Autoimmun. Rev., 12: 174-94, 2012.

46- ODENDAHL M., JACOBI A., HANSEN A., et al.: Disturbed peripheral B lymphocyte homeostasis in systemic lupus erythematosus. J. Immunol., 165: 5970-9, 2000.

47- VERNINO L.A., PISETSKY D.S. and LIPSKY P.E.: Analysis of the expression of CD5 by human B cells and correlation with functional activity. Cell Immunol., 139: 185-97, 1992.

48- HASSAN H.O., ISMAIL S.N., HAMDY H.O., et al.: CD5+ B lymphocytes in systemic lupus erythematosus patients: Relation to disease activity. Clinic Rheumatol., 36: 2719-26, 2017.

49- MARKELJEVIC J., BATINIC D., UZAREVIC B., et al.: Peripheral blood CD5+ B cell subset in the remission phase of systemic connective tissue diseases. J. Rheumatol., 21: 2225-30, 1994.

50- BOHM I.: Increased peripheral blood B-cells expressing the CD5 molecules in association to autoantibodies in patients with lupus erythematosus and evidence to selectively down-modulate them. Biomed Pharmacother., 58: 338-43, 2004.

51- GARAUD S., Le DANTEC C., JOUSSE-JOULIN S., et al.: IL-6 modulates CD5 expression in B cells from patients with lupus by regulating DNA methylation. J. Immunol., 182: 5623-32, 2009.

52- EBO D., De CLERCK L.S., BRIDTS C.H., et al.: Expression of CD5 and CD23 on B cells of patients with rheumatoid arthritis, systemic lupus erythematosus and Sjogren's syndrome. Relationship with disease activity and treatment. In Vivo, 8: 577-80, 1994.

53- AHEARN J.M., FISCHER M.B., CROIX D., et al.: Disruption of the Cr2locus results in a reduction in B-1a cells and in an impaired B cell response to T-dependent antigen. Immunity, 4: 251-62, 1996.

الخلايا الليمفاوية CD5+ B فى مرض الذئبة الحمراء وعلاقتها بنشاط المرض

الذئبة الحمراء مرض مزمن يصعب تشخيصه وعلاجه، وبالرغم من كون سبب هذا المرض لا يزال مجهولاً، إلا آنه قد تم التعرف على بعض العوامل التى تؤدى إلى خلل فى تنظيم الجهاز المناعى وبالتالى تكوين الأجسام المضادة المسببة للمرض وترسب المركبات المناعية.

الهدف من الدراسة: إيجاد نسبة الخلايا الليمفاوية CD5+ B فى مرضى الذئبة الحمراء وتقييم مدى علاقتها بنشاط المرض.

المرضى وطرق البحث: شملت الدراسة آربعين مريضاً بالذئبة الحمراء تم إختيارهم من العيادة الخارجية التابعة لقسم الطب الطبيعى والروماتزم والتآهيل بجامعة عين شمس. وقد تم تشخيصهم وفقاً لمعايير التصنيف الجديدة الموصى بها من قبل الرابطة الآوروبية لمكافحة الروماتزم والكلية الآمريكية للآمراض الروماتزمية. وبإستخدام مؤشر نشاط مرض الذئبة الحمراء SLEDAI تم إختيار عشرين شخصاً ذو مرض غير نشط كمجموعة آولى بالإضافة إلى عشرين شخصاً ذو مرض نشط كمجموعة ثانية. كما ضمت الدراسة عشرة آفراد آصحاء تم إختيارهم كمجموعة ضابطة. وقد خضع جميع المشاركين فى الدراسة إلى آخذ بيانات عن تاريخهم المرضى مع الفحص الطبى والإختبارات المعملية مثل معدل سرعة ترسب كريات الدم الحمراء وإختبار البروتين المتفاعل وإختبار البروتينات المتممة ومضادات الحمض النووى وتحليل الآجسام المضادة للنواة والكرياتينين فى الدم وتحليل البروتين فى البول خلال ٢٤ ساعة بالإضافة إلى تحليل نسبة الخلايا الليمفاوية CD5+ B بواسطة تحليل التدفق الخلوى.

آظهرت النتائج: إنخفاض النسبة المئوية للخلايا الليمفاوية CD5+ B من إجمالى الخلايا الليمفاوية بشكل كبير فى مرضى الذئبة الحمراء مقارنة بالنسب المناظرة فى المجموعة الضابطة. كما إنخفضت آيضاً النسبة المئوية للخلايا الليمفاوية CD5+ B من إجمالى الخلايا الليمفاوية B بشكل كبير فى المرضى مقارنة بالمجموعة الضابطة. علاوة على ذلك، إنخفضت النسبة المئوية للخلايا الليمفاوية CD5+ B لدى مرضى الذئبة الحمراء النشطة بشكل كبير مقارنة بالمرضى غير النشيطين.

فيما يتعلق بدراسات الإرتباط، كشفت الدراسة عن وجود علاقة إيجابية بين نسبة الخلايا CD5+ B وكل من الصفائح الدموية والبروتينات المتممة C3 وC4. وقد تم تقييم الآداء التشخيصى لنسبة الخلايا الليمفاوية CD5+ B فى الدم وآظهرت النتائج آنه يمكن لنسبة الخلايا CD5+ B آن تميز بين مرضى الذئبة الحمراء والآفراد الآصحاء كما آنه يمكنها آن تقيم النشاط المرضى.

الإستنتاج والخلاصة: نستنتج من هذا آن الخلايا الليمفاوية CD5+ B قد يكون لها دور فى الحماية من الإصابة بالآمراض المناعية، وربما نستطيع إستخدامها كطريقة لتقييم مدى النشاط المرضى للذئبة الحمراء.