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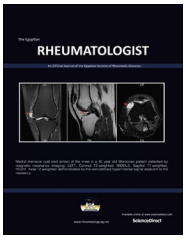

SHORT COMMUNICATION

Association between anti-Ro 60 kDa (SS-A) autoantibodies and hypocomplementemia in systemic lupus erythematosus patients from Algiers prefectures

K. Kerboua [a,*], **S. Chaib** [b], **F. Haiba** [c]

[a] *Immunology Unit, Military University Hospital of Oran, HMRUO, Oran, Algeria*
[b] *Immunology Department, Central Military University Hospital, Algiers, Algeria*
[c] *Scientific Council, Military University Hospital, HMRUO, Oran, Algeria*

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KEYWORDS

Anti SSA;
Circulating immune complexes;
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Abstract *Background:* Systemic lupus erythematosus (SLE) is characterized by a vicious cycle maintaining systemic inflammation. It starts by autoantibody production, immune complex formation and complement activation that contribute to inflammation, tissue damage and further autoantibody production.

Aim of the work: To evaluate the association between the auto-antibodies (abs), circulating immune complexes (CIC), and complement activity in SLE patients.

Patients and methods: This study involved 30 female SLE patients analyzed for autoantibodies, complement profile including complement hemolytic 50 (CH50), alternative pathway 50, factor B, C1q, C2, C3 and C4 as well as C1q-CIC. SLE disease activity was assessed by the SLE Disease Activity Index (SLEDAI).

Results: The age of patients was 34 ± 12.8 years, disease duration was 5.2 ± 3.2 years and their mean SLEDAI was 9.96 ± 4.2. Anti-SSA, anti-dsDNA, anti-C1q abs, and CIC were detected in 36.7%, 50%, 50% and 30% of patients, respectively. Anti-SSA were higher in patients with lower compared to normal CH_{50} activity and C3 level (24.7 vs 88.6 U/ml; $p = 0.002$ and 118.6 ± 25.18 U/ml vs 15.9 ± 7.3; $p < 0.0001$ respectively) than the other autoantibodies. Increased CIC were higher in patients with lupus nephritis and were associated with anti-SSA, anti-SSB, anti-C1q, anti-Sm and in patients with low CH50 activity. The CIC significantly correlated with anti-C1q ($r = 0.69$, $p < 0.0001$), anti-SSA ($r = 0.5$, $p = 0.005$) and negatively with CH_{50} ($r = -0.4$, $p = 0.046$).

Conclusions: The current study confirms that the etiopathogenic anti-SSA autoantibodies are the most associated with hypocomplementemia in SLE. This would stimulate future researches for

* Corresponding author. Mobile: +213 (0) 541 23 91 63.
E-mail address: k.k.eddine@gmail.com (K. Kerboua).

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validation of predictive biomarkers earlier than hypocomplementemia which is still the major unmet need in lupus research and patient care.

1. Introduction

Systemic lupus erythematosus (SLE) is an autoimmune disease characterized by disturbed tolerance to self antigens and development of auto-antibodies leading to the formation of immune complexes (ICs) [1]. These ICs deposit in the tissues initiating an immune response by activating the complement cascade and recruiting inflammatory cells [2]. The etiology of SLE is still not fully understood but the autoantibodies [3] and interplay of multiple factors including cytokine over production [4,5] and genetic predisposition [6] have been involved in the abnormal immune responses and pathogenesis of SLE. Furthermore, apoptosis [7] and oxidative stress [8] play central roles in the development of the disease.

Ro60 (also called 60 kDa SS-A) is an RNA-binding protein of 60 kDa that takes its name from the initials of a systemic lupus erythematosus (SLE) patient "Ro" [9] and the other nomenclature related to its association with Sjögren's syndrome (SS) [10]. It is proposed to function in a clearance pathway in which imperfect noncoding RNAs are targeted for decay, for cell survival; and ultimately, prevention of autoimmunity [11]. In autoimmune rheumatic disorders, among a variety of autoantibodies, the Ro/SSA abs are the most prevalent extractable nuclear antigen (ENA) specificity identified in laboratories. Sera from SLE patients have anti Ro antibodies (abs) in 24–60% and in 70–100% of patients with subacute cutaneous lupus erythematosus [12,13].

Needless to a reminder that releasing high amount of auto-abs is followed by intensive consumption of the classical complement compounds [14]. This latter is considered the best predictive biomarker for SLE flare and is included in SLE diagnosis criteria [15]. Hence, it is vital to detect an antibody that would be associated the most with hypocomplementemia to detect earlier SLE flare and help to monitor SLE activity. In this way, McClain and colleagues have taken an important step in answering these questions by determining that anti SSA auto-abs appeared nearly 4 years before the diagnosis of lupus [16].

The aim of this study was to evaluate serum autoantibodies with special attention to anti-Ro/SSA abs and assess their relation to hypocomplementemia and circulating immune complexes (CIC) in Algerian SLE patients.

2. Patients and methods

This study involved 30 consecutive female SLE patients attending different wards of the Central Military University Hospital, Algiers, Algeria. All patients fulfilled the 1997 American College of Rheumatology (ACR) criteria [17]. The SLE Disease Activity Index (SLEDAI) was done for each patient [18]. The absence of flare was defined by SLEDAI = 0 after subtraction of anti-double stranded deoxyribonucleic acid (anti-dsDNA) and complement points from SLEDAI index. All patients gave written informed consent, and the study protocol was fully approved by the ethics committee of the Central Military University Hospital, Algiers, Algeria. The plasma and sera were separated and stored at −80 °C until analysis of the autoantibodies and complement as well as the CIC.

2.1. Autoantibodies and CIC analysis

Antinuclear antibodies (ANA) were detected by indirect immunofluorescence (IIF) using the HEp-2000 test (BioRad, Germany). Titers equal or exceeding 1:160 were considered positive. Qualitative estimation of autoantibodies against 12 antigens: ribonucleoprotein (RNP), Smith (Sm), SS-A, SS-B, Tripartite Motif Containing 21 (TRIM21), scleroderma-70 (Scl-70), polymyositis-scleroderma (PM-Scl), Jo-1, centromere B, Proliferating cell nuclear antigen (PCNA), ds-DNA, nucleosome, histone and ribosomal protein P by bead cytometry Luminex (Austin, Texas, USA) using FIDIS™ connective 10 FIDIS (Biomedical diagnostics, 'BMD', Belgium). Three different assays were used to quantify anti-dsDNA: IIF by Crithedia lucilaea (Bio-RAD), ELISA (BioSystems, Spain) and Luminex. ELISA was used for anti-nucleosome (BioSystems, Spain), anti-phospholipids [IgG, IgM anti-Cardiolipin and anti B_2GPI] (VARELIS), anti histones abs (EUROIMMUN, Germany), and to quantify C1q-CIC (BINDAZYME™ CIC Human Anti-C1q). *Normal values* were considered according to the manufacturer's instructions: anti ds-DNA abs ($\geqslant$50 UI/ml by ELISA and $\geqslant$40 UI/ml by Luminex), anti-nucleosome abs ($\geqslant$10 AU/ml), anti C1q abs ($\geqslant$20 AU), CIC ($\geqslant$10.8 µg Eq) and for anti histone abs ($\geqslant$20 U).

2.2. Complement analysis

The classical and alternative complement pathways hemolytic activities were assessed by the previously described CH_{50} and AH_{50} [19]. Antigenic C1q, C2 and complement factor B (CfB) were assayed by radial immunudiffusion (The Binding Site Birmingham, UK), while light-scatter techniques nephelometry was used for C3 and C4 (nephelometry on BN Prospec® by Siemens). Detection of anti-C1q Abs (IgG) was performed by ELISA with purified C1q (The Binding Site, Birmingham, UK).

2.3. Statistical analysis

Correlations between complement parameters, C1q-CIC, and auto-abs were done by the nonparametric Spearman's Rank. Non-parametrical Mann–Whitney *U* test was used for analysis of statistical relations between patient groups and immunological markers. Results were considered significant when $p < 0.05$. Statistical analysis was performed using SPSS for

Windows 20.0 statistical software (SPSS, Inc, IL, Chicago, USA).

3. Results

The mean age of 30 patients was 34 ± 12.8 years, the mean disease duration was 5.2 ± 3.2 year their mean SLEDAI was 9.96 ± 4.2 (Table 1). Regarding ACR criteria for SLE, arthritis was detected in more than 80% of patients, followed by photosensitivity and hematological disorders. The frequency of ANA was 73.3%. The clinical features of the studied patients did not differ from the ACR one (Table 2).

The current study exhibited high frequencies for anti-nucleosome, anti ds-DNA and anti C1q abs. Anti nuclear abs were positives in 72.2%. For the ENAs; anti Sm, anti U1-RNP, anti SSA60 and anti SSB were positives at 6.7%, 16.7%, 36.7%, 16.7% respectively. There were no positive sera for anti centromere-B or anti Jo1 abs. Otherwise, for the non-ENAs: the frequency for anti histones was 20% and 86.7% for anti nucleosome, while anti ds-DNA was found in 30% by IIF, 46.7% by ELISA and 50% by Luminex. The frequency of positive antiphospholipid abs was totally 16.7% and CH_{50} activity was decreased in 80% of SLE patients (Table 3).

None of the anti-SSA positive patients had any known homozygous deficiency for C2 or C4. Among the autoimmunity panel, only anti-dsDNA assessed by Luminex and anti-SSA abs were associated with complement consumption. Anti-SSA were higher in patients with lower CH_{50} and C3 (24.7 vs 88.6 U/ml; $p = 0.002$ and 118.6 ± 25.18 U/ml vs 15.9 ± 7.3; $p < 0.0001$ respectively). Of interest, there was no significant difference in the detection of anti-dsDNA in patients' sera with low and normal C3 (529.6 ± 175.5 IU/ml vs 463.5 ± 53.9 IU/ml, $p = 0.1$), although the values differed significantly between those with low and normal CH_{50} (874 ± 430.48 UI/ml vs 137 ± 39.38 UI/ml, $p = 0.000$) (Fig. 1).

Increased C1q-CIC levels were higher in patients with lupus nephritis (14.8 ± 4.9 µg Eq/ml vs 4.99 ± 0.9 µg Eq/ml; $p = 0.03$) and were associated with anti-SSA 60 kDa (15.4 ± 5.4 µg Eq/ml vs 5.7 ± 1.5 µg Eq/ml; $p = 0.04$), anti-SSB (25 ± 10.4 vs 6.1 ± 1.3; $p = 0.001$), anti-C1q (13.9 ± 3.6 µg Eq/ml vs 3.1 ± 0.9 µg Eq/ml; $p = 0.02$), anti-Sm (21.5 ± 9.2 µg Eq/ml vs 7.4 ± 2.1 µg Eq/ml; $p = 0.04$) and in patients with low CH50 activity (36.8 ± 12.8 µg Eq/ml vs 6.18 ± 1.27; $p < 0.0001$) (Fig. 2). The CIC-C1q level significantly correlated with the anti-C1q and anti-SSA 60 kDa abs and negatively with the CH_{50} (Table 4).

Table 1 Age, disease duration, number of ACR criteria and disease activity in SLE patients.

Characteristics	Mean ± SD
Age (years)	34 ± 12.77
Disease duration (years)	5.22 ± 3.18
ACR criteria (n°)	6.66 ± 2.39
SLEDAI	9.96 ± 4.2
Renal SLEDAI	2.086 ± 1.3

ACR: American college of rheumatology, SLEDAI: systemic lupus erythematosus disease activity index.

Table 2 Clinical features according to the ACR criteria for SLE in the patients.

SLE criteria	*n*	%
Skin lesions	21	(70)
Discoid rash	11	(36.7)
Photosensitivity	23	(76.7)
Oral ulcers	14	(46.7)
Arthritis	25	(83.3)
Serositis	17	(56.7)
Renal involvement	13	(43.3)
Neurological disorders	6	(20)
Hematological disorders	23	(76.7)
Antinuclear antibodies	22	(73.3)

ACR: American college of rheumatology, SLE: systemic lupus erythematosus.

Table 3 Autoantibodies, complement profile and circulating immune complexes (CIC) in SLE patients.

Test	SLE patients ($n = 30$)	
	+ve finding (%)	Cutoff
Autoantibodies		
ANA (IIF) positivity	73.3	1:160
Anti-dsDNA		
IIF positivity	30	1:10
ELISA (IU/ml)	46.7	50
Luminex (IU/ml)	50	40
Anti-C1q (U/ml)	50	20
Anti-nucleosome (U/ml)	86.7	10
Anti-histone (U/ml)	20	20
Anti SSA 60 (U/ml)	36.7	40
Anti SSB (U/ml)	16.7	40
Anti Sm (U/ml)	6.7	40
Anti U-RNP (U/ml)	16.7	40
Anti centromere (U/ml)	0	40
Anti Jo1 (U/ml)	0	40
Anti TRIM21 (U/ml)	33.3	40
Anti PLs IgG (GPL)	16.7	10
IgM (MPL)		10
Complement components		
CH50 (U)	7.9	80% NP
C1q (mg/dl)	7.9	80% NP
C2 (mg/dl)	13.2	80% NP
C3c (mg/dl)	7.9	
C4 (mg/dl)	15.8	
AP50 (U)	7.9	80% NP
C fB (mg/dl)	7.9	80% NP
Circulating immune complexes		
C1q-CIC (µg Eq/ml)	30	4.4–10.8

SLE; systemic lupus erythematosus, ANA: antinuclear antibodies, IIF: indirect immunofluorescence, ELISA: enzyme linked immunosorbent assay, SS: Sjögren syndrome, Sm: Smith, RNP: ribonucleoprotein, TRIM: Tripartite Motif Containing 21, PLs: phospholipids, Ig: immunoglobulins, CH50: complement hemolytic 50, C; Complement, AP: alternative pathway, C fB: complement factor B, CIC: circulating immune complexes, NP: normal population.

4. Discussion

A conserved RNA-binding protein, the Ro 60 kDa (Ro60) auto-antigen, is a major target of autoantibodies in patients

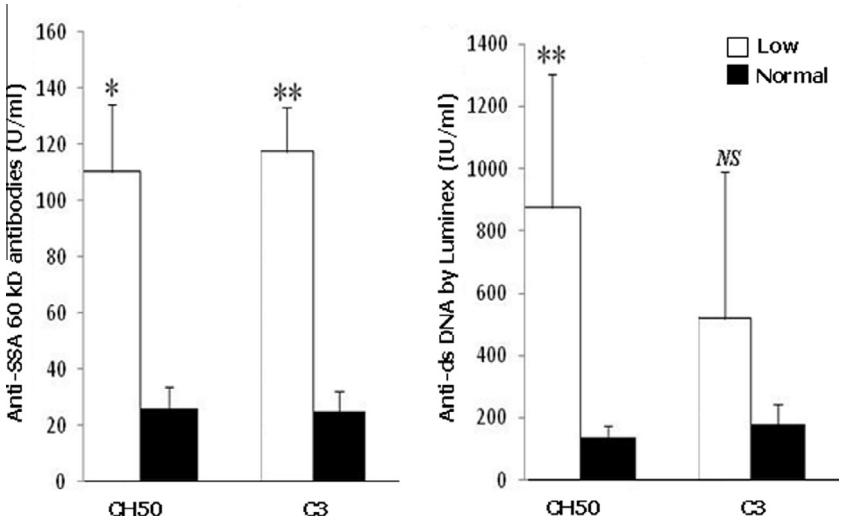

Figure 1 Anti ds-DNA and anti-SSA according to low and normal C3 and CH50 levels in SLE patients. * = significant at $p < 0.05$, ** = significant at $p < 0.001$. CH50: complement hemolytic 50, C: complement, NS: not significant.

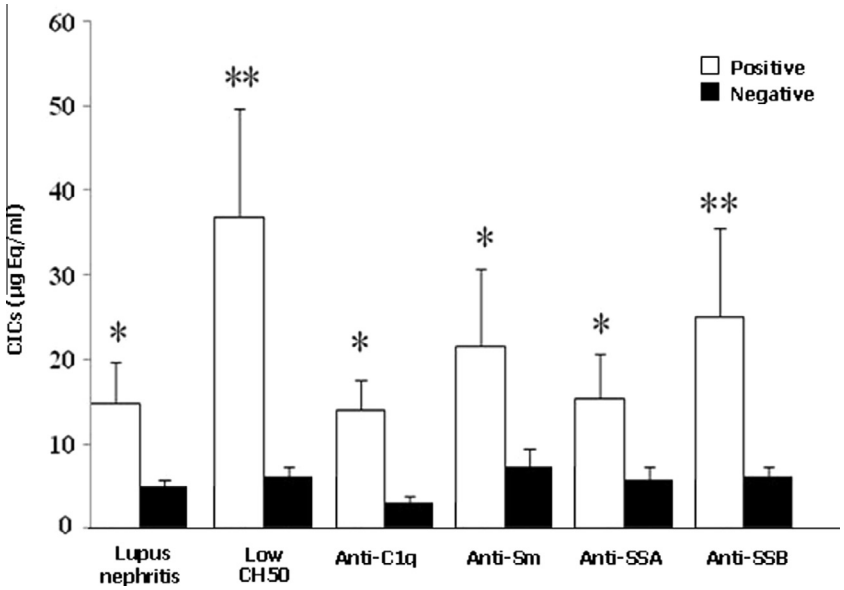

Figure 2 The circulating immune complex (CIC) titers according to the presence of lupus nephritis, low CH50 activity and autoantibodies. * = significant at $p < 0.05$, ** = significant at $p < 0.001$. CH50: complement hemolytic 50, C: complement, Sm: Smith, SS: Sjögren syndrome.

Table 4 CIC titer correlations with anti-SSA, anti-C1q antibodies and CH50 in SLE patients.

Parameters r (p)	CIC	
Anti-SSA	0.5	(0.005)
Anti-C1q	0.69	(<0.0001)
CH50	−0.4	(0.046)

SS: Sjögren syndrome, CIC: circulating immune complexes, CH50: complement hemolytic 50.

suffering from the rheumatic diseases including SS, SLE, subacute cutaneous lupus erythematosus and neonatal lupus erythematosus. This protein binds misfolded noncoding RNAs in vertebrate cells and likely functions in a pathway by which defective RNAs are recognized and targeted for degradation [20]. Besides, SLE is known for its high apoptotic cell load; and consequently, the extensive exposition of SSA may induce anti-SSA abs and autoimmunity. Otherwise, the pathogenesis of active SLE involved reduced serum complement with abundant CIC levels [21]. Therefore, the *immune complex theory* postulated that immune complexes of autoantibodies and their related auto-antigens activate complement, resulting in complement consumption and tissue damage. Moreover, possible roles for complement proteins, particularly C1q in the regulation of cytokine release have also been suggested in SLE which drew more attention for anti-C1q involvement in this disease [22,23]. Given these observations, it has been proposed that

among SLE biomarkers, anti-SSA, anti C1q, CIC and CH50 activity are of high interest and the current study was designed to investigate their association.

The present results revealed that CH_{50} activity is decreased in about 80% of SLE patients. Nonetheless, only anti-dsDNA and anti-SSA were associated with hypocomplementemia. However, anti-SSA abs levels were significantly increased in $C3_{low}$ sera, which is in disagreement with the results obtained by Mok et al. who have shown that the serum levels of anti-dsDNA abs correlated negatively with C3 and C4 levels [24]. A variety of tests that can detect anti-dsDNA abs have been described and only a few of them exhibit a reliable clinical and scientific usefulness. The current study confirmed that anti-dsDNA abs assessed by Luminex (not by ELISA or IIF), were associated with hypocomplementemia. Of note, anti-dsDNA abs, albeit they have less specificity, are the most sensitive for SLE. They are found in the sera of 55% to 80% SLE patients, which concords with our results.

As in our study (36.7%), anti-Ro 60 abs are the second most frequent SLE associated auto-abs [25,26]. Both classically complement function and the occurrence of anti-SSA influenced circulating IC levels, with a strong relation between decreased complement function and the occurrence of anti-SSA abs [27]. In contrast to the study of Hassan et al. [27], the levels of anti-SSA exhibit variations in relation to low C3, low CH_{50} and correlate significantly with CICs. This is in accordance with another study showing higher significance [28]. Furthermore, cytoplasmic (Ro) antigens could be involved in immune complexes [1,29] and are capable of fixing the complement [1]. This latter is employed clinically as an indirect and early indicator of the presence of immune-complex-mediated tissue damage, as well as a useful guide for the suppression of disease activity by immunosuppressive agents.

Impairment of IC handling is believed to be pathogenic and relevant to the complement system in SLE patients [30]. Nevertheless, these data don't follow the previous studies about the tendency of anti SSA60 to mediate antibody dependent cellular cytotoxicity (ADCC)-induced damage, because the most cogent explanation for this hypocomplementemia is the complement dependant cytotoxicity (CDC). In other words, immune complexes consist of antibodies associated with their corresponding antigens to provoke type III hypersensitivity that involves the classic complement pathway [31]. Noteworthy, persistent IC induce the production of IL10 and IL6 that might contribute to increased specific production of anti-SSA, further IC production, and thereby contribute to a vicious cycle in SLE maintaining systemic inflammation [1]. None of the anti-SSA positive patients had any known homozygous deficiency for C2 or C4 as this could have caused a bias in the analysis.

In accordance to previous studies (30–60%) [32,33], the current study found an autoimmune response directed to C1q in 50% of SLE patients. Besides, our results show that serum anti C1q antibodies levels correlated with serum C1q-CIC concentrations. After Eugenia Bălănescu et al. this positive correlation could be explained by the fact that decreased CIC binding to erythrocytes and platelets may increase their serum half-life [34]. It is possible that CIC binding of C1q determines C1q conformational changes similar with those described when binding to cryoglobulins [35]. Moreover, we found that CIC was significantly associated with low CH_{50} and correlated negatively with it, which is in accordance with previous data showing the important role of complement, in particular C3, in the clearance of ICs [36]. Small CIC are common activators of the classic pathway that cause generally hypocomplementemia, the hallmark of SLE. Abnormal clearance of these complexes was suggested to play a major etiologic influence in SLE [37]. Removal failure, possibly related to abnormalities in the interaction of these apoptotic bodies with classic pathway components may sustain the presence of CIC within tissues and potentially evoke an autoimmune response that may target C1q thus amplifying this process [38,39].

Anti-SSA antibodies are regarded as earlier SLE markers, but have not previously been associated with disease activity. The results of the present study indicate that these antibodies might be of importance in the formation of CIC and hypocomplementemia. The current data suggest that the measurement of anti-SSA abs may be a helpful tool in the assessment of patients with biologically active SLE. Of note, anti dsDNA may be used to monitor SLE activity, albeit it depends on the methods variability and availability. Hereby, only anti dsDNA assessed by Luminex was associated with hypocomplementemia. Conducting the study longitudinally and on a larger scale is recommended to confirm our results and study the possible relations with other autoantibodies, relation to medications and disease damage.

Conflict of interest

None.

References

[1] Pieterse E, van der Vlag J. Breaking immunological tolerance in systemic lupus erythematosus. Front Immunol 2014;5:164.
[2] Wu J, Ekman C, Jonsen A, Sturfelt G, Bengtsson AA, Gottsater A, et al. Increased plasma levels of the soluble Mer tyrosine kinase receptor in systemic lupus erythematosus relate to disease activity and nephritis. Arthritis Res Ther 2011;13(2):R62.
[3] Kamen DL. Environmental influences on systemic lupus erythematosus expression. Rheum Dis Clin North Am 2014;40(3):401–12.
[4] Mohsen MA, Abdel Karim SA, Abbas TM, Amin M. Serum interleukin-18 levels in patients with systemic lupus erythematosus: relation with disease activity and lupus nephritis. Egypt Rheumatol 2013;35(1):45–51.
[5] Gaber W, Sayed S, Rady NM, Mohey AM. Interleukin-27 and its relation to disease parameters in SLE patients. Egypt Rheumatol 2012;34(3):99–105.
[6] Azkalany GS, Gheita T, Gaber W, Mohey A. Clinical significance of serum TNFα and -308 G/A promoter polymorphism and serum Il-6 and -174 G/C promoter polymorphism in systemic lupus erythematosus patients. Egypt Rheumatol 2012;34(3):119–25.
[7] Gheita TA, Bassyouni IH, Bassyouni RH. Plasma concentrations of growth arrest specific protein 6 and the soluble form of its tyrosine kinase receptor Axl in patients with systemic lupus erythematosus and Behçets disease. J Clin Immunol 2012;32(6):1279–86.
[8] Hassan SZ, Gheita TA, Kenawy SA, Fahim AT, El-Sorougy IM, Abdou MS. Oxidative stress in systemic lupus erythematosus and rheumatoid arthritis patients: relationship to disease manifestations and activity. Int J Rheum Dis 2011;4:325–31.
[9] Clark G, Reichlin M, Tomasi TB. Characterization of a soluble cytoplasmic antigen reactive with sera from patients with systemic lupus erythematosus. J Immunol 1969;102:117–22.

[10] Alspaugh MA, Tan EM. Antibodies to cellular antigens in Sjögren's syndrome. J Clin Invest 1975;55(5):1067–73.
[11] Chen X, Wolin SL. The Ro 60 kDa autoantigen: insights into cellular function and role in autoimmunity. J Mol Med (Berl) 2004;82(4):232–9.
[12] Inagaki Y, Jinno Y, Hamasaki Y, Ueki H. Higher incidences of anti-SS-A/Ro and anti-SS-B/La autoantibodies in Japanese patients with autoimmune disorders–studies of antigens and antibodies using the immunoblotting method. Arch Dermatol Res 1989;281(2):89–94.
[13] Franceschini F, Cavazzana I. Anti-Ro/SSA and La/SSB antibodies. Autoimmunity 2005;38(1):55–63.
[14] Stoyanova V, Petrova S, Tchorbadjieva M, Deliyska B, Vasilev V, Tsacheva I. New insight into the autoimmunogenicity of the complement protein C1q. Mol Immunol 2011;48(4):678–82.
[15] Petri M, Orbai AM, Alarcón GS, Gordon C, Merrill JT, Fortin PR, et al. Derivation and validation of the Systemic Lupus International Collaborating Clinics classification criteria for systemic lupus erythematosus. Arthritis Rheum 2012;64(8):2677–86.
[16] McClain MT, Heinlen LD, Dennis GJ, Roebuck J, Harley JB, James JA. Early events in lupus humoral autoimmunity suggest initiation through molecular mimicry. Nat Med 2005;11:85–9.
[17] Hochberg MC. Updating the American College of Rheumatology revised criteria for the classification of systemic lupus erythematosus. Arthritis Rheum 1997;40:1725.
[18] Formiga F, Moga I, Pac M, Mitjavila F, Rivera A, Pujol R. Mild presentation of systemic lupus erythematosus in elderly patients assessed by SLEDAI. SLE Disease Activity Index. Lupus 1999;8:462–5.
[19] Nilsson UR, Nilsson B. Simplified assays of hemolytic activity of the classical and alternative complement pathways. J Immunol Methods 1984;72:49–59.
[20] Wolin SL, Reinisch KM. The Ro 60 kDa autoantigen comes into focus: Interpreting epitope mapping experiments on the basis of structure. Autoimmun Rev 2006;5(6):367–72.
[21] Lachmann PJ, Müller-Eberhard HJ, Kunkel HG, Paronetto F. The localization of in vivo bound complement in tissue section. J Exp Med 1962;115:63–82.
[22] Yamada M, Oritani K, Kaisho T, Ishikawa J, Yoshida H, Takahashi I, et al. Complement C1q regulates LPS-induced cytokine production in bone marrow-derived dendritic cells. Eur J Immunol 2004;34(1):221–30.
[23] Rönnblom L, Eloranta ML, Alm GV. Role of natural interferon-alpha producing cells (plasmacytoid dendritic cells) in autoimmunity. Autoimmunity 2003;36(8):463–72.
[24] Mok CC, Lau CS. Pathogenesis of systemic lupus erythematosus. J Clin Pathol 2003;56:481–90.
[25] Ben-Chetrit E, Eilat D, Ben-Sasson SA. Specific inhibition of the DNA-anti-DNA immune reaction by low molecular weight anionic compounds. Immunology 1988;65:479–85.
[26] Ioannidis JP, Boki KA, Katsorida ME, Drosos AA, Skopouli FN, Boletis JN, et al. Remission, relapse, and re-remission of proliferative lupus nephritis treated with cyclophosphamide. Kidney Int 2000;57(1):258–64.
[27] Hassan AB, Lundberg IE, Isenberg D, Wahren-Herlenius M. Serial analysis of Ro/SSA and La/SSB antibody levels and correlation with clinical disease activity in patients with systemic lupus erythematosus. Scand J Rheumatol 2002;31:133–9.
[28] Mathsson L, Ahlin E, Sjöwall C, Skogh T, Rönnelid J. Cytokine induction by circulating immune complexes and signs of in-vivo complement activation in systemic lupus erythematosus are associated with the occurrence of anti-Sjögren's syndrome A antibodies. Clinical Exp Immunol 2007;147:513–20.
[29] Tan EM, Kunkel HG. Characteristics of soluble nuclear antigen precipitating with sera of patients with systemic lupus erythematosus. J Immunol 1966;96:464–71.
[30] Schifferli JA, Taylor RP. Physiological and pathological aspects of circulating immune complexes. Kidney Int 1989;35 (4):993–1003.
[31] Tang S, Lui SL, Lai KN. Pathogenesis of lupus nephritis; an update. Nephrology (Carlton) 2005;10:174–9.
[32] Seelen MA, Trouw LA, Daha MR. Diagnostic and prognostic significance of anti-C1q antibodies in systemic lupus erythematosus. Curr Opin Nephrol Hypertens 2003;12:619–24.
[33] Marto N, Bertolaccini ML, Calabuig E, Hughes GR, Khamashta MA. Anti-C1q antibodies in nephritis: correlation between titres and renal disease activity and positive predictive value in systemic lupus erythematosus. Ann Rheum Dis 2005;64(3):444–8.
[34] Bălănescu E, Tănăsescu C, Bălănescu P, Olteanu R, Badea C, Petruţescu B, et al. Anti C1q antibodies in cutaneous lupus erythematosus. Rom J Intern Med 2010;48(2):159–63.
[35] Terrier B, Derian N, Schoindre Y, Chaara W, Geri G, Zahr N, et al. Restoration of regulatory and effector T cell balance and B cell homeostasis in systemic lupus erythematosus patients through vitamin D supplementation. Arthritis Res Ther 2012;14(5):R221.
[36] Qing X, Zavadil J, Crosby MB, Hogarth MP, Hahn BH, Mohan C, et al. Nephritogenic anti-DNA antibodies regulate gene expression in MRL/lpr mouse glomerular mesangial cells. Arthritis Rheum 2006;54(7):2198–210.
[37] Mevorach D. Clearance of dying cells and systemic lupus erythematosus: the role of C1q and the complement system. Apoptosis 2010;15(9):1114–23.
[38] Walport MJ. Complement and systemic lupus erythematosus. Arthritis Res 2002;4(Suppl. 3):S279–93.
[39] Smykał-Jankowiak K, Niemir ZI, Polcyn-Adamczak M. Do circulating antibodies against C1q reflect the activity of lupus nephritis? Pol Arch Med Wewn 2011;121(9):287–95.