Journal of the Egyptian Society of Parasitology, Vol.49, No. 2, August 2019
J. Egypt. Soc. Parasitol. (JESP), 49(2), 2019: 401 – 404

SEROLOGY OF THE SYSTEMIC LUPUS ERYTHEMATOSUS IN EGYPTIAN PATIENTS

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Abstract

Systemic lupus erythematosus (SLE) is a prototypic multisystem autoimmune disease with a vast spectrum of clinical presentations including almost all body organs and tissues. The incidence of lupus has tripled in the last 40 years, mainly due to the improved diagnosis. For more than 60 years the presence of autoantibodies in patients with SLE has been known. A great effort is being made for understanding the diagnostic, pathogenic and prognostic meaning of these autoantibodies. The study demonstrated the significance of autoantibodies test in Egyptian SLE patients. The results showed that 93.3% of patients have positive ANA test, 90% have positive anti-dsDNA. Anti-Sm antibodies were in 40% and 49.3% of patients have positive anti-nucleosome antibody test. Also, 30.6% of patients deve-loped a positive anti-histone test & 26% have positive anti-ribosomal test. Anti-phospholipids antibodies were found in 44.6% of patients**.** Levels of C3 & C4 (0.552±0.43 & 0.125±0.12, respectively) were lower than that of control group (1.2±0.35 & 0.42±0.54 for C3 & C4, respectively).

Key words: Egypt, Systemic lupus erythematosus, Autoantibodies test

Introduction

Systemic lupus erythematosus (SLE) is an autoimmune disease characterized by auto-reactive B and T cells and production of a broad and heterogeneous group of autoanti-bodies (Cozzani *et al*, 2014).

Antibodies that react with self-molecules occur in healthy individuals and are referred to as the natural antibodies or autoantibodies (Casali and Schettino, 1996). These self-antigens may be present in all types of cells or extensively specific for a specific cell type in one organ of the body. It may include proteins, nucleic acids, carbohydra-tes, lipids or various combinations of these. In systemic lupus, the dominant antigens are the ribonucleo-proteins (RNPs) or deoxyrib-onucleoproteins (Elkon and Casali, 2008).

Hargraves *et al.* (1948) demonstrated the presence of autoantibodies in SLE when they described lupus phenomenon. This was proven when it was understood that lupus was due to neutrophil phagocytosis of cell nuclei opsonized by autoantibodies. DNA antibodies were identified (Ceppellini *et al*, 1957) and Tan and Kunkel (1966) discover ed autoantibodies directed to antigens differ-ent from DNA and described the anti-Sm antibodies. These autoantibodies were used as useful biomarkers for SLE disease, and gave information about basic mechanisms of the loss of tolerance and inflammation (Elk-on and Casali, 2008). Sherer *et al.* (2004) reported that 116 autoantibodies were identi-fied in patients with SLE. In SLE, especially in the systemic form, auto-antibodies direct-ed to the nuclear (ANAs), cytoplasmtic, and cellular membrane antigens are considered the serologic hallmark. ANAs consisted of various types of auto-antibodies characteri-zed by the different antigen specificities. The nuclear antigens include the single strand (SS) and double strand (DS) DNA, histone proteins, nucleosome, centromere proteins, and extractable nuclear antigens (Smith antigen (Sm), Ro, La, ribonucleo-protein (RNP)…etc.). ANAs are present in about 95% of SLE patients with an active disease (Egner, 2000). In the patients with prevalent cutaneous lesions, ANAs have been found positive in 75% of cases. Hamdy *et al.* (2010) in Egypt, reported that most of SLE studied patients were characterized pre-dominantly by the muco-cutaneous and the

hematological manifestations, and the QoL of SLE patients with renal involvement as measured by SF-36 was poor than that of the healthy control in all the domains except emotional limitations.

The present study aimed to highlight the most promising and significant ones from the immuno-pathological and clinical perspectives point of view.

Patients and Methods

The study involved 150 patients with SLE attending the Department of Rheumatology and Rehabilitation, Cairo University Hospitals. All patients fulfilled the 1997 American College of Rheumatology criteria for SLE (Hochberg, 1997) and their disease activity was assessed by SLE Disease Activity Index or SLEDAI (Bombardier *et al*, 1992).

Qualitative analysis of antinuclear autoantibodies was done using anti-nuclear antibody immunoblot kit (Germany). The anti-double-stranded deoxyribonucleic acid antibody, anti-phospholipid antibodies, i.e., IgM & IgG anti-cardiolipin antibody, and IgM & IgG anti-β2 glycoprotein I antibody were measured using commercial ELISA kits and the omplement components C3 & C4 were measured by Nephelometry (Germany). The signed informed consent was obtained from all participants and the study was approved by the institute's Ethics Committee.

Results

The study included 150 patients with SLE (136 females & 14 males), whereas the apparent healthy controls were 150 individuals (130 females & 20 males) cross matched and ethnic background. Patients and controls were 30.8±5.6 & 29±6.5 years respectively. In SLE patients, disease onset was 29.3±6.8 years and duration was 12.5±1.2 years. The mean SLEDAI score was 13.9±9.6, indicated that all patients had active disease. The clinical pictures were hematological involvement (52%), lupus nephritis (52%), inflammatory arthritis (67%), and secondary anti-phospholipid antibody syndrome (12%). The complement components C3 & C4 were low in 39% and 31% of patients, respectively.

The autoantibody profile of patients C3 & C4 mean levels were summarized (Tab.1).

Table 1: Serology and complement C3 & C4 in SLE patients.

Autoantibody profiles	Number of patients (%)	No of controls (%)
ANA	140 (93.3%)	5 (3.3%)
Anti-dsDNA	135 (90%)	8 (0.53%)
Anti-nucleosome	74 (49.3%)	2 (1.3)%
APLA (β2GP1, LAC)	67 (44.6%)	1 (0.6%)
Anti-sm	60 (40%)	2 (1.3%)
Anti-histone	46 (30.6%)	1 (0.6%)
Anti-ribosomal	39 (26%)	3 (2%)
C3 (g/L)	0.552±0.43	1.2±0.35
C4 (g/L)	0.125±0.12	0.42±0.54

ANA: Anti-Nuclear Antibody, Anti-dsDNA: double strand (ds) DNA, APLA: antiphospholipids antibodies, anti-sm: anti- smith antibody, C3 and C4: complement proteins.

Discussion

Generally speaking, the systemic lupus erythematosus (SLE) is the chronic autoimmune disease affecting almost all organ systems, characterized by exacerbations (or flares) of the disease activity and the disease damages (Müller and Muir, 2014). The measures of disease activity include; SLE disease activity index, and British Isles Lupus Assessment group or BILAG disease activity index (Stoll *et al*, 1996) and SLE activity Measure or SLAM (Bae *et al*, 2000). Although 180 or more self-antigens were reported as targets in SLE yet, few were common, this subset, which includes the anti-Sm/RNP, anti-Ro/SS-A, anti-La/SS-B, anti-DS/DNA, and several others, consists primarily of the nucleic acid (DNA or RNA) associated proteins (Han *et al*, 2015).

In the present study, 93.3% of the patients gave positive ANA test; and 90% of them exhibited positive anti-DS/DNA. ANA are autoantibodies to the cells nuclei, 98% of all patients had a positive ANA test, making it

the most sensitive diagnostic test for SLE (Walravens, 1987). The ANAs were present in 5-10% of controls and people with other diseases; such as the rheumatoid arthritis (Grygiel-Górniak *et al*, 2018). Also, 20% of healthy women had a weakly positive ANA, and majority never developed lupus signs.

Anti-DS/DNA is a specific type of ANA antibody found in about 30% of SLE patients. Less than 1% of healthy individuals have Anti- DS/DNA, making it a helpful marker for confirming SLE. The presence of anti- DS/DNA antibodies often suggested more serious lupus, such as lupus nephritis. High amounts of anti-DNA antibodies were present in active lupus nephritis.

On the other hand, the anti-Sm antibodies were in 40% of patients. Anti-Sm is an antibody to a ribonucleoprotein found in the cell nucleus. It is found exclusively in people with lupus (Kurien and Scofield, 2006). It is present in 20% of patients with the disease, less than 1% of healthy individuals and rarely found in people with other rheumatic diseases. It proved helpful in diagnosis of systemic lupus. Unlike anti-DS/DNA, anti-Sm did not correlate with the presence of lupus nephritis.

In the present study, 49.3% of patients have positive anti-nucleosome antibody test. The first serological marker of the SLE described was anti-nucleosome antibodies. Nucleosomes are considered as the major autoantigen in SLE, played an important pathogenetic role and about 85% of patients have positive test (Bruns *et al*, 2000). Also, van der Vlag and Berden (2011) reported that nucleosome antibodies possess the important role in the pathogenesis of SLE, as being the first antibodies to appear in murine lupus models before the onset of other autoantibodies.

In the present study, 30.6% of the patients developed a positive anti-histone test. Anti-histone is an antibodies to histones, proteins that help to lend structure to DNA. It is usually found in both people with drug-induced lupus and people with systemic lupus (Cozzani *et al*, 2014). However, it was not specific enough to systemic lupus to be used as a diagnostic marker. But, 26% of patients have positive anti-ribosomal test. Nagai *et al.* (2005) reported that autoantibodies to ribosomal proteins were detected in 12-16% of SLE patients and have been associated with some disease manifestations, including the lupus psychosis and hepatitis.

In the present study, anti-phospholipids antibodies were found in 44.6% of patients, which were not confined to SLE patients only; but it was found in other autoimmune diseases, infections, malignant, and drug-induced disorders as well as in some apparently healthy individuals. In addition, Chu *et al.* (1988) reported that anti-phospholipids antibodies are positive in 30-40% of SLE patients. They added that only 1/3 of them developed clinical features of anti-PL syndromes such as the venous thrombosis, arterial thrombosis, recurrent pregnancy loss, hemolytic anemia and skin ulcers (Chu *et al*, 1988).

The present study reported 0.552±0.43 & 0.125±0.12 were for C3 & C4, respectively. These levels were lower than that of control group (1.2±0.35 & 0.42±0.54 for C3 & C4, respectively). The serum complement test measured the proteins consumed levels during the inflammatory process (Perrin *et al*, 1973). So, low complement levels reflect inflammation within the body. One of the diagnostic hallmarks of lupus is low C3 & C4 levels. Low C3 only is not specific to lupus; but combined low C3 & C4 is usually seen in lupus. Therefore, the patient's levels of C3 & C4 indicated the activity of the disease. Where, low levels of C3 and C4 suggested active disease and a current flare or a flare is imminent. Normal levels of C3 and C4 indicated that the disease is more likely calm; but it does not mean that the patient would not experience a flare. The consistently low level of C3 indicated the possibility of developing lupus nephritis in the future. In this case, the doctor must watch the kidneys and ask the patient to

return every two/three months for urine analysis (Walport, 2002).

Conclusion

Lupus occurs if immune system attacks healthy tissue (autoimmune disease). People with an inherited predisposition for lupus, when come into contact with environmental trigger lupus.

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