Original Article

Correlation between Interleukin-4 and Interleukin-6 and auto antibodies in Systemic Lupus Erythematosus.

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Summary:

Background: There is a general acceptance which illustrated that auto antibodies act as a central immunological disturbance in most of the auto immune diseases, among these auto immune disease lies the SLE

Patients and Methods: Thirty five patients with SLE were compared to twenty age and sex matched, control subjects and studied for the presence of auto antibodies, plus IL-4 and IL-6 using Elisa method and immune fluorescent method (for ANA only)

Results: Data showed that IL-6 detectable levels were statistically significant in patients with positive anti ds-DNA, but not significant statistically in ANA positive patients although it was detected in 24 (70.6%) of positive ANA patients, while there was no statistically significant correlation between IL-4 detectable level and autoantibodies production.

Conclusion: There is apparently a positive correlation between IL-6 and anti ds DNA production in lupus patients.

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Introduction:

There is a general acceptance, which illustrated that autoantibodies act as a central immunological disturbance in most of the autoimmune diseases, among this auto immune disease lies the SLE; where autoantibodies could be the actual pathogenic agent for the consequence of tissue damage in SLE (1). Antinuclear antibody (ANA) was the most characteristic and prominent antibody ever to be detected in lupus patients. It has been found in about 95% of those patients and they were included as serological marker for the diagnosis along with the anti ds DNA and anti smooth antibodies (2, 3). The production of antibodies often occurs as a result of hyper activity of B-cell which is increased in number in Lupus patients as well as increased number of plasma cell which secreting immunoglobulin in the peripheral blood (4).There is evidence that cytokines may play a role in the activation of these cells for Example B-cell in SLE patients are more easily differentiated by IL-6 than normal B-cell (5, 6). On the other hand, IL-4 is not detectable or low in SLE patients; mean while other study shows decrease IL-4 messenger RNA expression in peripheral blood mononuclear cell (7).

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Patients and methods:

Patients: Thirty-five lupus patients with age range from 9 to 45 years (mean age 30 years), were included in study. They were 6 males and 29 females. Those patients fulfilled 4 or more of the American College of Rheumatology (ACR) criteria for classification of SLE (8). Those patients attended the Rheumatology Department of Baghdad Teaching Hospital for the period from November 2001 to mid of January 2002.Twenty randomly selected, healthy subjects were included, as a control group, and they were 8 males and 17 females whose ages ranged from 16-45 years (mean age 27.9 years).

Methods: Each Lupus patient and control person was investigated for:

1-Detection of IL-4 and IL-6 using Elisa method.
a-Human IL-4 Elisa kit (IBL Cat No: MG 51042)
b- Human IL-6 Elisa kit (IBL Cat No: MG 51042)
2- Detection of ANA by ANA screen test using Elisa method and immunofluorescent.
3- Detection of anti ds DNA antibody by Elisa (anti ds DNA Elisa kit; Biomeghrib No. 80604)

Statistical Analysis:Data were translated into codes using especially designed coding sheet, and then into a computerized data base structure. Statistical analyses were done using SPSS version. Computer software (Statistical package for social science). Person's linear correlation coefficient was used to assess the strength and direction of linear correlation between two continuous variables. P values less than the 0.05 level of significance was considered statistically significant.

Results:

Serum Cytokines:There was a statistical significance of detectable levels of IL-6 in SLE patients (68.6%) compared to healthy control (0%). While there was no statistical significance of detectable levels of IL-4 in SLE patients and healthy control (Table -1).

Serum auto antibodies:Thirty-five data sera of SLE patients shows (97%) ANA positive by using both tests Elisa and immunofluorescent. While 74.3% of lupus patients showed positive ds DNA by using Elisa method. In control group, the test was negative for both ANA and Anti ds DNA (Table -2).

Regarding detectable levels of interleukin in auto antibodies positive patients, Table (3) shows that IL-6 was detected in (80.8%) of positive anti ds DNA antibody which is statistically significant, while it was detected in (70.6%) of ANA positive patient but this result was not statistically significant, while there was no statistical significance between IL-4 detectable levels and auto antibodies positive patients.

Table (1) Detection of IL-4 and IL-6 level in SLE patients and control groups.

Interleukin concentration (pg/dl)	SLE (n=35)		Healthy control (n=20)		P - value
	N	%	N	%	
IL-4 concentration Cutoff=85	2	5.7	0	0	0.4 (N.S)
IL-6 concentration Cutoff=160	24	68.6	0	0	< 0.001

Table (2) Auto antibodies detection in SLE and control.

Auto antibodies	SLE N (%)	Control N (%)
ANA Positive Negative	 34 (97.1) 1 (2.9)	0 (0) 20 (100)
Anti ds DNA Positive Negative	 26 (74.3) 9 (25.7)	 0 (0) 0 (0)

Table (3) Correlation between IL-4 and IL-6 and autoantibodies in SLE patients.

Prevalence of detectable interleukin levels						
	IL-4			IL-6		
	N	(%)	P(X^2)	N	(%)	P(X^2)
1.Antinuclear antibodies			0.44 (NS)			6.31 (NS)
Positive (n=34)	2	5.9		24	70.6	
Negative(n-1)	0	0		0	0	
2.Anti double stranded DNA antibodies			0.55 (NS)			0.02
Positive (n=26)	2	7.7		21	80.8	
Negative (n=9)	0	0		3	33.3	

Discussion:

The serological hallmark of systemic lupus erythematosus is the presence of circulating auto antibodies directed against a wide variety of antigens, this was quite obvious in the present study in which ANA and anti ds DNA antibodies were detected in 97% and 74% respectively which is comparable with another study by Worall et al (9). Autoantibodies production occurs in a setting of generalized immune cell abnormalities that involve the B-cell, T-cell and monocyte lineage. Together with cytokine appear to promote B-cell hyperactivity leading to hyperglobulineamia, increased number of antibodies producing cells and heightened responses to many antigens both self and non-self (10).

Regarding to IL-4 level, only two patients showed detectable level which may explain the elevated level of IL-6 in lupus patients, because one of normal biological activity of IL-4 is down regulation of IL-6 production by normal monocyte. Recent study showed that exogenous IL-4 down regulates IL-6 production by SLE derived peripheral blood monocyte cell (11).

Conclusions:

Serum IL-4 was undetectable in most of patients with SLE; this may reflect the role of this cytokine in the etiology of the disease, by failure to suppress secretion of IL-6 by macrophage. Positive auto antibodies (ANA and anti ds DNA) with their high frequency may reflect the action of Th subset derived cytokine as IL-6.

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