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Evaluation of Serum Interleukin-6 in Lichen Planus

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Abstract

Background: Lichen planus (LP) is animmune mediated inflammatorydiseaseof skin and mucous membranes without a clear etiology. Interleukin-6 (IL-6) is a pro-inflammatory cytokinewith variousbiologicaland pathological impactson immunity, acute phase response andinflammation. ExcessiveIL-6production can play a role in the pathogenesis of several pathological conditionslike psoriasis.

Aim:This study aimed to evaluate serum IL-6 levels inLP patients alongwithassessment ofitscorrelation with disease severity.

Patients and methods: Twenty-one adult patients with LP as well astwenty-one healthy adult controls were enrolled in the study. The total score of LP severity was estimated for all patientsby calculation of the affected body surface area in cutaneous LP patients and clinical scoring system with visual analogue scale for pain assessment for oral LP.Serum IL-6 levels were measured in all participants using enzyme-linked immunosorbent assay.

Results:The mean serumIL-6 levels were significantly higher in patients than controls (P < .002). A positive correlation wasnoticed between serum IL-6 with both cutaneous LP (P <0.004) and oral LP (P <0.005) severities.

Conclusion:Serum IL-6may be a potential biomarker for LP as it was greater in LP patients than controls and waspositivelycorrelated with disease severity.Further studies are necessary

to evaluate the therapeutic efficacy of IL-6 blockade that could be beneficial in LP management.

Key words:

Lichen planus, cutaneous, oral, interleukin-6, severity.

1. Introduction

Lichen planus (LP) is a chronic, inflammatory, T-cell mediateddisorder that can affect skin, mucosa, hair and nails. Classic LPis characterized by violaceous, flat or polygonal pruritic papules and plaques with the distinctivewhite reticular Wickham's stria. It usually affects middle-aged populations, with a slightpredilectionin women.**1**LP pathogenesis is not fully elucidated. Multiple factors were suggested to contribute to its pathogenesis,although immunological dysregulation is consideredas the most accepted triggerof damage.**2**

Interleukin-6 (IL-6) is a pleiotropic proinflammatory mediator produced by a broad spectrum of cells with variable roles in immunological responses, homeostasis, acute phase reactions and hematopoiesis. It is secreted temporarily as an adaptive response in cases of injuries and infections. **3**Previously, IL-6 had different names as B-cell stimulatory factor 2 (BSF-2) and hepatocyte stimulating factor (HSF) and Interferon-β2 (IFN-β2) but later all these compounds were proved to be the same by cloning so it was given the name of IL-6. **4**

Excessive IL-6 production can lead to higher expression of vascular endothelial growth factorin inflamed tissuesand tumors resulting in more angiogenesis and increase in vascular permeability. These eventsare typically seen in cases of inflammation and malignant conditions maintaining their chronicity and even progression **5**

Increased levels of IL-6 in serum were reported in several inflammatory diseases such as rheumatoid arthritis, Castleman's disease and Crohn's disease.**6**

Additionally, certain dermatological conditions weresuggested to have significantly higher serum IL-6 in patients compared to healthy individuals which include psoriasis **7**, atopic dermatitis**8** and acne vulgaris.**9**

Debates concerning the presence of a significant difference in IL-6 serum levels amongLP patients comparednormal individualswere reported by many studies.IL-6 serum as

well as salivarylevels were suggested to be higher in cases than controls. **10.** Additionally, serum IL-6 levels were observed to decrease after systemic treatment of LP patients with dexamethasone compared to higher levels before treatment. **11**

The variation of results may be due to ethnic difference because Caucasian and Asian populations showed different results being significant in Asians rather than in Caucasians.**12**

2. Patients and methods:

This study was approved by the Institutional Review Board of the Faculty of Medicine, Zagazig University, Egypt. An informed consent was obtained from each participant. All the data were kept private after assigning a code number to everyparticipant, only known by the researchers.

This case-control study was conducted at Zagazig University Hospital between February to December 2020. Data were collected consecutively from 21 adults, clinically proven, LP patients admitted to the Dermatology, Venereology and Andrology Department, Faculty of Medicine, Zagazig University, Egypt. The control group involved 21matched healthy individuals. Participants with infectious diseases, malignancy,autoimmune disorders,those treated for LP systemicallywithin the last2 months beforestudyas well asparticipants with positive covid-19 within the last 3 months were excluded from the study.

Allparticipants were subjected to detailed history taking, thorough physical examination and measurement of serum IL-6. In addition, LP severity was assessed for all LP patients.

Estimation of serum IL-6 by enzyme-linked immunosorbent assay method

Serum IL-6 levels were estimated based on the principle of double-antibody sandwich techniqueusing ELISA Kit for in vitro quantitative measurement according to the producer's protocol (SunRedBio, Shanghai, China).Five milliliter of venous blood samples were drawn into a sterile vial from all participants.The clotted samples arethen centrifuged for 10 minutesat 3,000 rpm. Then serum was transferred to labeled tubes and stored at -80°Cuntil analysis.

Standard and Streptavidin-HRP were mixed.Sample was added, with (IL-6 -antibody) and Streptavidin-HRP to the test wells. Then the membrane was sealed, gently shaken, incubated for 60 minutes at 37°C.A thirtytimes dilution was done with distilled water as standby. Then membrane was removedfollowed by liquid drainage, shakenaway the remaining water. Then, we added chromogen solution A followed by chromogen solution B to each well. Gently mixed and incubated for 10 min at 37°C away from light.Stop Solution was addedtoall wellsto stop the reaction(the blue changes into yellow immediately). The optical density (OD) was measured under 450 nm wavelength. According to standards' concentration and the corresponding OD values. Finally, the concentration of serum IL-6 in each sample was calculated from the standard curve relating the intensity of the color to the concentration of standards.

Assessment of disease severity for the involved cases:

- Cutaneous LP severity assessment was doneby calculation of the affected body surface area (BSA) where BSA is measured using the total palmar surface of hand, involving the five fingers, which isroughly considered 1% of total BSA. Patients were categorized on the basis of BSA into mild (<3% BSA affected), Moderate (3-10% affected) and severe (>10%affected) groups. This scale is primarily used in psoriasis with no similar scale for LP, it was useful practically and clinically for this study. **13**
- Severity assessment of OLP was doneusing a semiquantitative scoring system based on the site, area, and presence of erosions, known as clinical scoring system of OLP. It divides the mouth into 8 areas with score of severity between 0 and 2. Grading, based on scores, is divided into grade 0,1,2 and 3.Determination of severity,based on grade, is either mild, moderate or severe disease.**14**
- The visual analogue scale was used to assess pain in OLP with symptoms intensity on a scale from 0 to 10, 0 represent no symptoms while 10 represent the worst imaginable symptoms.**15**

3. Results:

Various characteristics of the study participants are shown in Table (1). Of the 21 patients with LP, 14 (66.7%) were females and 7 (33.3%) were males. The patients' age ranged between

29 and 68 years. And the same numbers for control group for males and females. Age ranged from 28to65 years. LP duration ranged from 1 week to 5 years.

Table (1): Characteristics of the studied groups:

Variables	Case group (n=21)	Control group (n=21)	Test of sig.	P
Age (years):			T	
Mean ± SD	50.2 ± 10.0	49.1 ± 12.2	0.3	0.8
Sex:				
Males	7 (33.3%)	7 (33.3%)	NA	NA
Females	14 (66.7%)	14 (66.7%)		
Duration:				
<1 year	9(42.9%)			
1-2 years	6 (28.6%)			
>2 years	6 (28.6%)			
Type of LP				
Cutaneous LP	11 (52.4%)			
Oral LP	4 (19.0%)			
Combined	6 (28.6%)			

N: number T: Independent t test χ^2: Chi square test NA: not applicable.

P <0.05: Significant P >0.05: non-significant. P<0.001: Highly significant.

SD: standard deviation. Sig.: significance.

The mean levels of serum IL-6 were significantly higher in patients than their healthy controls (P < .002; Table 2, Figure 1).Strong positive correlations between serum IL-6 and oral LP severity (P< .005)as well as withcutaneous LP were found (P < .004) as shown in table (3).

Table (2) Serum IL-6 in the studied groups:

Serum IL-6	Case group (n=21)	Control group (n=21)	MW	P
Mean ± SD	41.4 ± 35.5	18.5 ± 8.0		
Media	29.2	17.8	3.1	**0.002**
Range	3.7 – 125.7	1.8 – 33.1		**S**

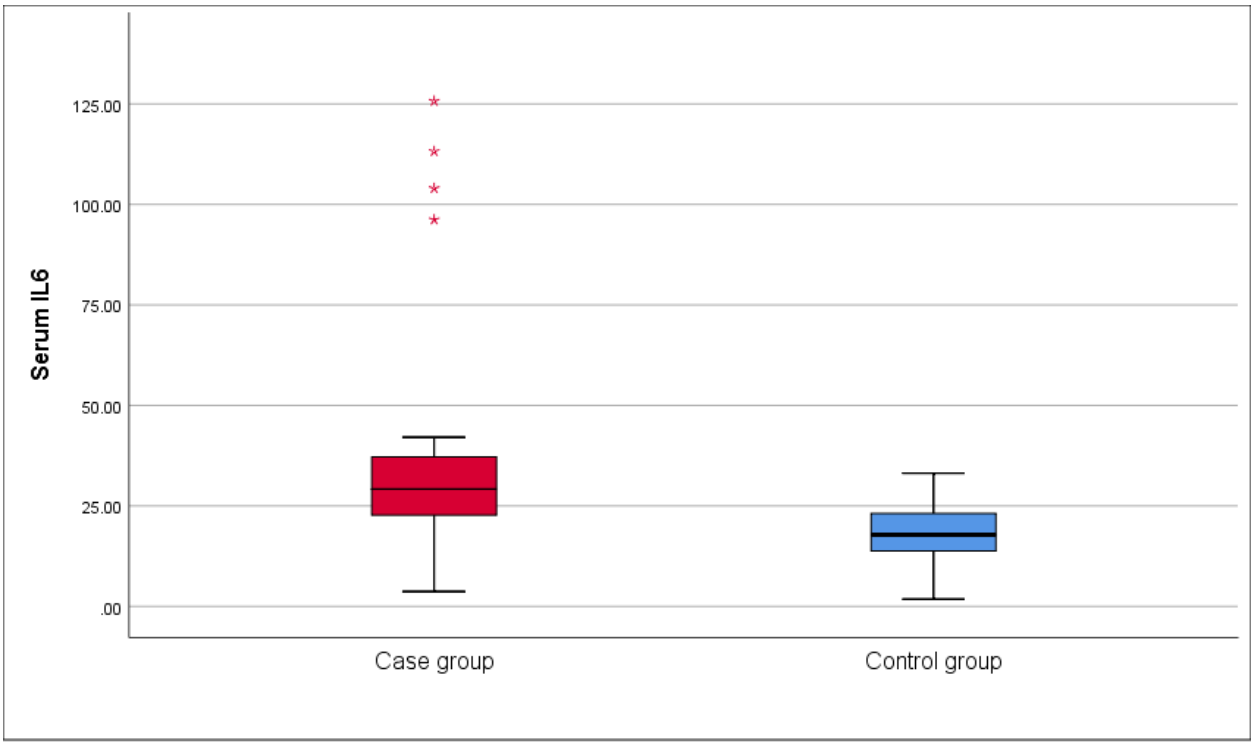

Figure (1): Serum IL-6 in the studied groups

Table (3)Correlation between serum IL-6 with severity of lesions in the studied patients

Variables	SerumIL-6	
	R	P
Cutaneous LP severity	0.660	**0.004**
Oral LP severity	0.809	**0.005**

4. Discussion

The current findings indicate that IL-6 serum levels were greater in LP patients than in controls and were significantly associated with cutaneous LP severity.Moreover, a strong positive correlation was observed between serum IL-6 levels and severity of oral LP.

Interleukin-6 is a pro-inflammatory cytokine with several functions including roles in humoral and cellular immunities:through promoting B cell differentiation, and stimulating immunoglobulin G secretion, T cell growth and differentiation, and cytotoxic T cell differentiation as well. **12**

Dysregulated production of IL-6 is known to play a pathogenic role in different inflammatory conditionslike rheumatoid arthritis and Castleman's disease IL-6 blocking therapy with an anti-IL-6 receptor antibody (tocilizumab) was reported toimprove clinical symptoms and the abnormal laboratory findingsin some inflammatory conditions as rheumatoid arthritis. **4**

Increased skin and serum IL-6 levels are a feature of psoriasis. Serum levels of IL-6 are regarded as a marker of the inflammatory activity in psoriasis as well as an indicator of treatment response. **7**

IL-6 was also found to be significantly higher in patients with atopic dermatitis**8** andacne vulgaris**9** comparedto healthy controls.

In the present study, we observed an association between LP and IL-6 serum levels in Egyptian population. We also found a statistically significant association between serum IL-6 levels and cutaneous LP severity. These findings could suggest a role of IL-6 in the pathogenesis of LP and might also be helpfuland simplebiomarker forevaluation of LP severity. IL-6 blockade by the humanized anti-IL-6 receptor antibody had been suggested as a successful measure in management of some inflammatory disorders mediated by IL-6. **16**Thisdrug could be tested in case of LP to judge its potential usefulness in its management.

5. Conclusion

In conclusion, serum levels of IL-6 were elevated in LP patients and was significantly correlated with cutaneous and oral LP severities; hence, it might be a potential biomarker for assessment of LP severity and subsequently could be used to monitor the therapeutic outcomes for better disease control. Further studies are suggested to verify our results and to assess the therapeutic effect of IL-6 receptor blockade, which could be advantageousin management of LP.

Conflict of interest

There are no conflicts of interest

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