

Some Cytokines Levels (IL-6 and IL-10) in Sera Patients with Cutaneous Leishmaniasis in Nassiriya Province

Alaa Hussein Oleiwi*
Department of biology
College of Sciences, University of
Thi-Qar
Thi-Qar, Iraq
Alaah.bio@sci.utq.edu.iq

Fadhial Abbas Manshad
Department of biology
Faculty of Education for Pure
Sciences, University of Thi-Qar
Thi-Qar, Iraq

Ali Naeem Salman
Department of biology
Faculty of Education for Pure
Sciences, University of Thi-Qar
Thi-Qar, Iraq

Abstract— The present study was carried out in the Labs of collage of education for pure science, during period from January 2017 to endDecember of the same year. The immune status investigates for CLpatients by measuring the levels of cytokines (IL6and IL10) in sera using a technique enzyme-linked immune Sorbent adsorptive (ELISA). The study included 120 subjects with (60 CLpatientsL.majar and 60 CLpatients L. tropica with and (30) were healthy control. Increased mean Serum level of IL6 was in the observed in the total patients as compared to control Subjects (224.53pg/ml,70.70pg/ml), the result indicate there was significant difference at ($p<0.05$), such observation was consistent in the patient infected with L.majar and L. tropica (104 .90 pg/ml and 112.78 pg/ml) respectively. The results of the IL10 showed significant difference at ($p<0.05$)increased of mean Serum level in the total CL patients as compared to control Subjects(226.90 pg/ml 46.77pg/ml,).Ahighly significant difference at ($p<0.05$) increased observed in patients group infected with L.majar and followed by patients group infected L. tropica (112.78pg/ml and 114.12pg/ml) respectively. These results revealed that the excessive presence of cytokines might play a role in CL patients.

Keywords— Cutaneous Leishmaniasis, *L.majar*, *L. tropica*

I. INTRODUCTION

Leishmaniasis is a significant zoonotic disease caused by species of the genus *Leishmania*, it is a protozoan disease that affects approximately 12 million people worldwide, particularly in tropical and sub-tropical regions and endemic in near 100 countries worldwide. The protozoa *Leishmania* have digenetic lifecycles and can exist in two morphological forms; either as amastigotes inside the immune cells (macrophages) of mammals, or as flagellated promastigotes within the gut of a phlebotomine sand fly (Chappuis *et al.*, 2007; Gradoni, 2015). In CL, the infection is usually limited

to the skin and lymphatic system, but it may influence on deeper tissues in diffuse CL or penetrate into the mucous membranes in MCL. The life cycle is completed when sand flies feed near the skin lesions and the amastigotes enter the midgut of the sand fly where they subsequently develop into promastigote forms (Bailey and Lockwood, 2007; Kaye and Scott, 2011) There are many complexities in immunity against leishmaniasis. Interleukins are small protein molecules that signal specific cells to regulate the immune systems of organisms, they are primarily synthesized by T cells, monocytes, macrophages and endothelial cells (Gomes, 2017). The functions of IL include the facilitation of communication among immune system cells, regulation of transcription factors, and control of inflammation, cell differentiation, proliferation and antibody secretion (Salazar-Onfray *et al.*, 2007). several cytokines, such as interleukin- 10 (IL-10), have during the early immune response indeed, the function of a given cytokine is determined by its tissue levels, the nature of the target cell and activating signal, the timing and sequence of cytokine exposure and more generally (Cavaillon, 2001) IL-6 is a pleiotropic cytokine that acts as both a pro-inflammatory and anti-inflammatory cytokine. IL-6 is produced by several cell types, including macrophages DCs and T cells. Also, this cytokine acts as a B-cell growth factor. In this study, we investigated the immune status of CL patients infected with two species *L.majar* and *L. tropica* by studying the following immunological parameters by (IL-6 and IL-17) (Scheller *et al.*, 2011).

II. MATERIAL AND METHODS

This study was performed on 120 patients with cutaneous leishmania, who attended Imam Hussein Teaching Hospital, Shattrah general hospital, Rifya general hospital, Suq Al-Shayokh general hospital and Al-Chibayish general hospital the in Thi-Qar province in the period from the beginning of (October 2017 to end in May 2017 in the same year).

1- Collection Blood samples:

Interleukin	Subjects	Sample	Mean	Std. Deviation	t value	P value	
IL6	<i>L. major</i>	patients	60	104.90	38.46	3.738	0.001
		control	30	70.70	21.36		
	There is significant difference between groups at α 0.05						
	<i>L. tropica</i>	patients	60	119.63	57.27	4.017	0.0002
		control	30	70.70			
	There is significant difference between groups at α 0.05						
Total mean patients			224.53				
IL10	<i>L. major</i>	patients	60	112.90	31.33		0.0000
		control	30	46.77	11.20		
	There is significant difference between groups at α 0.05						
	<i>L. tropica</i>	patients	60	114.12	40.59	7.849	0.0000
		control	30	46.77	11.20	7.849	
	There is significant difference between groups at α 0.05						
Total mean patients			227.90				

Samples were collected by venipuncture from 120CL patients (60patients infected with *L. major* ,60patients infected with *L. tropica* and 30 controls.All patients were placed in a sterile plane tube and allowed to clot, then serum was separated by centrifugation at 4000 rpm for 15 minutes. The serum was stored at -20 C° freezing . These sera (120 patients and 30 controls) were used for estimating the concentration of interleukin (IL-6 and IL-10) .

The Nested PCR technique was performed for detection cutaneous leishmaniasis based on the kinetoplast DNA (kDNA) for detection *L. major* and *L. tropica*. This method was carried out according to the method described by Noyes *et al.*, (1998)

ELISA (technique enzyme-linked immune Sorbent adsorptive) kit areemploying the quantitative sandwich,were based on similar principle according to the Elabscience company (China,E-EL-H0101).

2- Statistical analysis:

Data were expressed as mean \pm standard deviation (SD) or median (interquintile range). Differences between groups were tested with the Student's t-test.The values of P < 0.05 were considered significant.

III. RESULTS AND DISCUSSION

In the current study the levels of human IL-6 and human IL-10 were measured in the patients sera using ELISA methods.The data analysis was done by SPSS and descriptive statistics were applied for the analysis of its results.Increased mean Serum level of IL6 was in theobserved in the total patients as compared to control

Subjects (224.53 pg/ml,70.70 pg/ml), the result indicate there was significant difference at (p<0.05) . such observation was consistent in the patient infected with *L. major* and *L. tropica* (104 .90 pg/ml and 119.63 pg/ml) respectively, as shown in table (1).

The results of the IL10 showed significant difference at (p<0.05)increased of mean Serum level in the total CL patients as compared to control Subjects(227.02pg/ml 46.77pg/ml).Ahighly significant difference at (p<0.05) increased observed in patients group infected with *L. major* and followed by patients group infected *L. tropica*(112 .78 pg/ml and 114.12 pg/ml) respectively,all these data summarized in table (1) .

During early invasion of Leishmania,T-cell and cytokines that they release play a critical role in determining the nature of the immune response and the outcome of the infection. (Mahmoodi *et al.*, 2005).These results were agreed with other studies.In Baghdad, study established by Al-Aubaidi (2011)increased Serum levels of IL-6 was significantly higher (p < 0.05) in patients with CL group than healthy subjects

TABLE.1. Serum interleukin levels among CL patients and control subjects

V. REFERENCE

Al-Aubaidi, I.K. (2011). Serum Cytokine Production in Patients with Cutaneous Leishmaniasis Before and After Treatment. IRAQI J MED SCI. 9(1).

Bailey, M.S.and Lockwood, D.N. (2007). Cutaneous leishmaniasis. Clin Dermatol, 25(2):203–11.

Cavaillon, J.M. (2001). Pro- versus anti-inflammatory cytokines: myth or reality. Cell Mol. Biol, 47, 695–702.

Chappuis, F.; Sundar, S.; Hailu, A.; Ghalib, H.; Rijal, S.; Peeling, R.W.; Alvar, J. and Boelaert, M. (2007). Visceral leishmaniasis: what are the needs for diagnosis, treatment and control. Nat. Rev. Microbiol. 5: 873- 882.

Gomes, K. (2017). IL-6 and type 1 diabetes mellitus: T cell responses and increase in IL-6 receptor surface expression. Ann Transl Med 5(1):16.

Gradoni, L. (2015). Canine Leishmania vaccines: still a long way to go. Vet Parasitol 208: 94-100.

Kaye, P.and Scott P.(2011). Leishmaniasis: complexity at the host-pathogen interface. Nat Rev Microbiol.,9(8):604–15.

Mahmoodi, M.; Rajabalian, S.; Fekri, A. and Esfandiarpour, I. (2005). Evaluation of In Vitro Production of IFN- γ , IL-10, IL-12 and IL-13 by Blood Cells in Patients with Cutaneous Leishmaniasis Lesions. IRANIAN JOURNAL OF ALLERGY, ASTHMA AND IMMUNOLOGY, 15. 4(1) .

Noyes , H. A.; Reyburn , H.; Bailey, J. W. and Smith, D.(1998). Anested PCR. Based Schizodeme method for identifying *Leishmania* kinetoplast minicircle classes directly from clinical samples and its application to the study of the epidemiology of *Leishmania tropica* in Pakistan. *J. of Clin. Microbiol.*; 36 (10) 2877 – 2881.

Salazar-Onfray, F.; Lopez, M.N. and Mendoza-Naranjo, A. (2007). Paradoxical effects of cytokines in tumor immune surveillance and tumor immune escape. *Cytokine & growth factor reviews* 18: 171-182.

Scheller, J.; Chalaris, A.; Schmidt-Arras, D. and Rose-John, S. (2011). The pro and anti-inflammatory properties of the cytokine interleukin-6. *Biochimica et Biophysica Acta (BBA). Mol Cell Res.*; 1813(5):878–88.