

The role of serum IL-6 changes in evaluation of endometriosis

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Summary:

Background:- Endometriosis is a common chronic gynecological disorder characterized by the presence and proliferation of functional endometrial gland and stroma outside the uterine cavity, affecting approximately 10% of reproductive age women. IL-6 level increased in the patient with endometriosis.

Objective:- Evaluate the correlation of IL-6 levels with the degree of severity of endometriosis

Patients and methods:- In this study, 84 samples 51women with endometriosis (20 newly diagnosed and 31 treated cases) and 33 of healthy control group, were collected starting from November 2011 to end of January 2012, to investigate the IL-6 serum level by using ELISA kit.

Results :- Endometriosis patients mainly presented at the age between (28-37). IL-6 serum levels were significantly decrease in those with stage IV endometriosis. Also appear to be significantly affected by the use of danazol or zoladex treatment of endometriosis and remain low even in treated patients. Serum IL-6 test was highly specific but with low sensitivity.

Conclusion:- Serum IL-6 concentration seem to be the highly specific test that can be used as an early detection marker for endometriosis.

Keywords: Endometriosis, IL-6.

J Fac Med Baghdad
2013; Vol.55, No .2
Received Jan .2013
Accepted April.2013

Introduction:-

Endometriosis is a common chronic gynecological disorder characterized by the presence and proliferation of functional endometrial gland and stroma outside the uterine cavity, affecting approximately 10% of reproductive age women [1,2]. It is one of the most complicated and baffling disease with debilitating symptoms of cyclical pelvic pain, which may render the patients life unbearable if left untreated [3]. Cells in endometriosis in that are normally found lining the uterus are also found on the pelvic peritoneum but may also be found on the ovaries, rectovaginal septum, ureter, and rarely in the bladder, pericardium and pleura [4,5]. IL-6 is a protein that in humans is encoded by IL-6 gene [6]. This interleukin acts as both a pro-inflammatory and anti-inflammatory cytokine. It is secreted by T cells and macrophages to stimulate immune response [7]. IL-6 response was dysregulated in the peritoneal macrophages, endometrial stromal cells and peripheral macrophages in patient with endometriosis.[8,9,10] IL-6 levels were raised in peritoneal fluid in endometriosis and the presence of this interleukin association with red glandular lesions could be related to infertility [11]. The increased IL-6 levels in patient with endometriosis will dropped after surgery or treatment with Gonadotrophin releasing hormone (GnRH) analogues.[12].

Materials and method:-

A study was conducted on the following groups in the period between November 2011 and January 2012. 51 female patients attend «Kamal Al-Samari» hospital and Baghdad medical city teaching hospital as (20) newly diagnosed cases of endometriosis and (31) treated cases of endometriosis for follow up. 33 apparently healthy fertile women without endometriosis their age was matched with the patients group ranging from (18-45) years. Blood samples were obtained from each individual by venous puncture, then was left to clot at room temperature, centrifuged and serum was collected for the detection of human Interleukin 6 (IL-6) by using Enzyme linked immunosorbent assay (ELISA) technique. IL-6 EILSA kit: This assay employs an antibody specific for human IL-6 coated on a 96 well plate. Standards and samples are pipetted into the wells and IL-6 present in a sample is bound to the wells by the immobilized antibody . The wells are washed and biotinylated anti-human IL-6 antibody is added. After washing away unbound biotinylated antibody, horseradishperoxidase (HRP) conjugated streptavidin is pipetted to the wells. The wells are again washed. A 3'3,5'5-tetramethyl-benzidine (TMB) substrate solution is added to the wells and color develops in proportion to the amount of IL-6 bound . The stop solution changes the color from blue to yellow, and the intensity of the color is measured at 450nm by spectrophotometer. The amount of color is directly proportional to the concentration of IL-6 antibody present in the original sample.

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Statistical analysis:-Data expressed as mean \pm SD.statistical differences between the groups were determined according to ANOVA test and student t-test was considered to be significant < 0.05.

were shown in table (1), endometriosis patients were mainly presented at the age between 28 and 37 year, (54%), but this percentage was not significant. Table (2) shows the mean changes of IL-6 between endometriotic and healthy women.

Results:-

The distribution of all study sample according to their age

Table (1): Frequency distribution of the study sample by age

		Type		Total	
		Patient	Control		
Age Gp**	18-27	Count	9	12	21
		% within Age Gp	42.9%	57.1%*	100.0%
		% within Type	17.6%	36.4%	25.0%
	28-37	Count	28	16	44
		% within Age Gp	63.6%	36.4%	100.0%
		% within Type	54.9%	48.5%	52.4%
	38-47	Count	14	5	19
		% within Age Gp	73.7%	26.3%	100.0%
		% within Type	27.5%	15.2%	22.6%
Total	Count	51	33	84	
	% within Age Gp	60.7%	39.3%	100.0%	
	% within Type	100.0%	100.0%	100.0%	

* % = percentage ** Gp= group

Table (2): Distribution of the serum IL-6

	Type	N*	Mean \pm SD	p.value
IL-6	Patient	51	47.568 \pm 20.834	0.851[N.S**]
	Control	33	100.087 \pm 39.199	

* N= number, **N.S=non significant

Endometriosis was classified into 4 stages according to the revised American Fertility society (rAFS), but in our study there was no patient at stage I. Figure (1) shows that serum levels of IL-6 were significantly decrease in those with stage

IV endometriosis. IL-6 remain low even in treated patients as shown in figure (2) which compare the mean change of the IL-6 between the treated and non treated groups of patients.

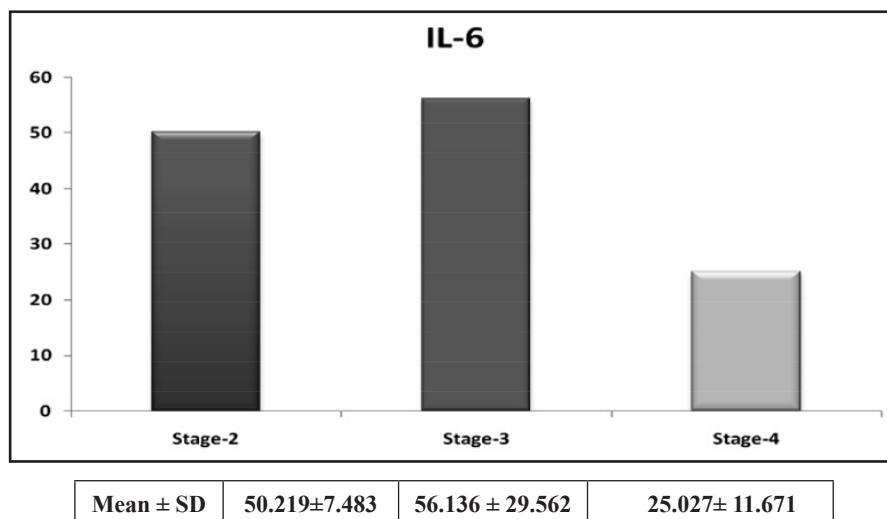


Figure (1): Serum IL-6 level in different stages of endometriosis

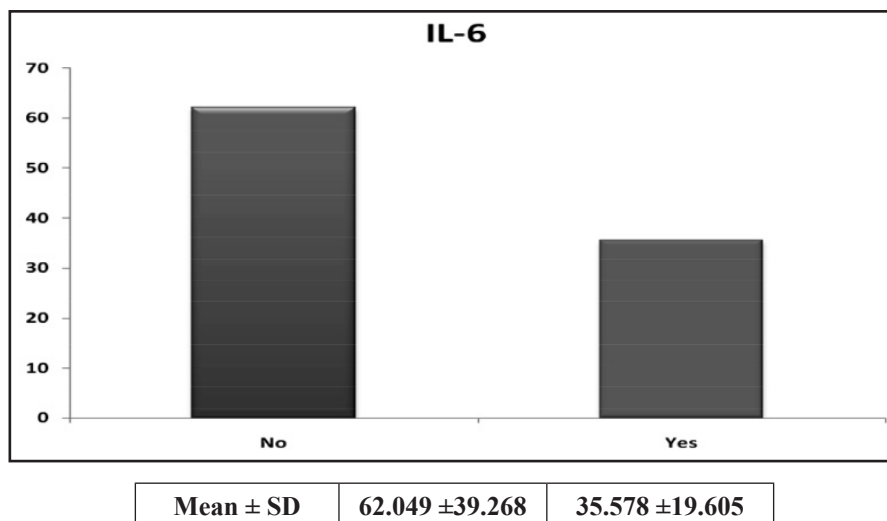


Figure (2): The mean changes IL-6 in treated and non treated patients

Table(3) shows the high specificity of IL-6 level when used in differentiating endometriosis patients from healthy controls but with a very low sensitivity (8%).

Table (3): Validity parameter for the studied endometriosis in differentiating it from healthy control

Overall	Sensitivity	Specificity	Accuracy	PPV	NPV
IL-6	8%	100%	60.7%	70%	100%

Discussion:-

The present study, shows that the mostly affected age with endometriosis is mainly those between 28-37 years old, this result agree with many other studies [13,14,15]. Eskenazi and Warner in 1997 published that endometriosis may effects (5-15)% of women within childbearing age, and at 2010 Gylfason et al. concluded that the incidence of endometriosis is higher in women of reproductive age [16,17]. Although multifunctional pro inflammatory cytokine may play a role in the pathogenesis of endometriosis [18,19]. Our thesis reveals a non significant decrease in the level of IL-6 in endometriosis patients when compared to healthy women. This result disagrees with many previous studies were their authors had been found that serum IL-6 were significantly higher in women with endometriosis than healthy women and may reaches its highest level in women with untreated chocolate cysts [20]. Our study showed that serum IL-6 levels were significantly decreasing in those with stage IV endometriosis. Figure (1) shows that serum IL-6 level was higher among those with stage II and III, this agrees with some previous studies [21]. Figure (2) shows that the mean change in IL-6 in treated patients much lower than that in non treated patients, this result agrees with many previous results showing the inhibitory effect of danazol and zoladex on IL-6 secretion by endometrotic cells [22]. The present study, shows 100% specificity of IL-6 but with only 8% sensitivity, this result agrees with many studies [23].

Conclusion:-

Endometriosis patients mainly presented at age (28-37) years old. Serum IL-6 levels were higher among those with stage II and III and significantly decrease in those with stage IV. The serum concentration of IL-6 appear to be significantly affected by the use of danazol or zoladex treatment of endometriosis.

Author Contributions:-

Tharwa Hadi Hassan AL-Tai / acquisition of data analysis, interpretation of data and drafting of manuscript.
 Hayfaa S. AL-Hadithi / study conception, design, interpretation of data and critical revision
 Hind S. Abdulsalam / acquisition of data analysis and critical revision

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