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Levels of Tumor Necrosis Factor Alpha and Interleukin-17 in Fertile and Infertile Women with Endometriosis

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Abstract

Background Endometriosis is described by the existence of endometrial tissue outside the uterine cavity. Infertility is one of clinical manifestation of endometriosis shown by the difference of fecundity.

Objective To compare the serum levels of TNF-α and IL-17 in fertile and infertile endometriosis patients.

Methods This study was conducted on 55 women patients with endometriosis (30 infertile women and 25 fertile women) and twenty apparently healthy controls. The technique used to reveal serum level of TNF-α and IL-17 was Enzyme-linked immune sorbent assay (ELISA).

Results This study revealed significant increase ($p<0.05$) in serum levels of TNF-α in infertile patients other than that fertile. On the other hand, there were no significant differences ($p>0.05$) between controls and each group of patients. Moreover, there were significant increase ($p<0.05$) in IL-17 levels in infertile patients than that fertile patients and controls, while there were no significant differences ($p>0.05$) between fertile patients and controls.

Conclusion The current results indicate that TNF-α and IL-17 might play a crucial role in endometriosis-related infertility.

Keywords Endometriosis, TNF-α, IL-17.

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List of abbreviations: ELISA = Enzyme-linked immune sorbent assay, IL-17 = Interleukin-17, TNF-α= Tumor necrosis factor alpha

Introduction

Endometriosis is a prevalent benign chronic inflammatory gynecologic disorder, described as the proliferation and presence of functional endometrial glands (endometrial-like tissue) external of the normal location (uterine cavity) [1,2]. It is affecting about 6-10% of women of reproductive age. These women may be asymptomatic, but the majority will present with pelvic pain, infertility, or an adrenal mass. In fact, endometriosis has been reported to be as high as 35–50% in women presenting with infertility [3]. Endometriosis is a major cause of infertility due to inflammation-associated reductions in oocyte quality and endometrial receptivity to embryonic implantation. However, the connection between infertility and endometriosis is especially obvious for advanced levels of the disease [3]. Though its pathogenesis still unknown, there is evidence showing that environmental factors, immunological, endocrine, and genetic factors play an important role in the development of endometriosis and genesis [4]. Endometriosis is associated with several immunological alterations, which are identified in infertile

patients [5,6]. These alterations participate in the development and progression of endometriosis and infertility [7].

Moreover, endometriosis may be considered an autoimmune disorder due to its immune aberrations, including elevated local production of several proinflammatory cytokines as well as increased autoantibody production and revocation of local and systemic cell-mediated immunity [8]. Strong evidence proposed that endometriosis was correlated with a state of subclinical peritoneal inflammation, noteworthy by raised growth factors and inflammatory cytokines [9].

The role of cytokines in women with endometriosis has been reported by various researches [10]. Tumor necrosis factor-alpha (TNF-α) is known as a pluripotent mediator and angiogenic cytokine that condense the production of other cytokines, including IL-8 in diverse cells as well the production of cytokine in endometriotic tissue. TNFα may be regarded as a key cytokine that actuate many other cytokines in the peritoneal cavity of endometriosis patients [11].

IL-17 is a representative cytokine excreted from Th17 cells. IL-17 deed on a wide range of cell types, including those of the mesenchymal lineages, epithelial, endothelial, and hemopoietic [12]. It has been shown that IL-17 can stimulate the expression of intracellular adhesion molecule (ICAM)-1 and increased the proinflammatory responses induced by IL-1h and TNF-a. In addition, IL-17 has been implicated in several inflammatory disorders [13].

Therefore, the present study aimed to compare between serum of TNF-α and IL-17 levels in fertile and infertile endometriosis patients.

Methods

A total of 55 serum samples were collected from endometriosis patients (30 infertile women and 25 fertile women) aged between 21-43 years who were attended to Kamal Al-Samari Hospital and Baghdad Medical City Teaching Hospital from June 2014 to January 2015. Twenty fertile healthy controls women were enrolled in this study. The diagnosis was done by the gynecologist, which was based on laparoscopy. They were recently diagnosed and all of the patients without treatment and other chronic diseases. 3 ml of venous blood was withdrawn from every subject (patients and control) under aseptic technique and positioning in the plain test tube with no anticoagulant, left to clot at room temperature and then separated the serum by centrifugation at 3000 rpm for 15 minutes, divided into aliquots and kept at -20 °C until used for investigations.

Levels of serum TNF-α and IL-17 have been measured by using commercially available ELISA and accomplished as recommended in leaflet with kit (TNF-α and IL-17 Boster/USA).

Statistical analysis

Comparison of serum levels of TNF-α and IL-17level among groups were counted by student's t-test and ANOVA test. P-values of $P<0.05$ was deemed significant.

Results

The age of the two groups of patients and control was matched. Mean age of infertile patients was 27.5 ± 2.39 year, while a fertile patient was 27.9±1.98 year and for controls was a 26.3±1.35 year as shown in table (1).

This study observed that there were significant differences ($p<0.05$) in mean serum levels of TNF-α between infertile and fertile females' patients (24.54±4.60 vs. 39.77±7.64). Furthermore, there were no significant differences ($p>0.05$) between controls and each group of patients as shown in table (2).

Table (3) revealed the increased ($p<0.05$) in mean serum IL-17 levels in infertile patients (72.95 ± 16.84) than that fertile patients (35.51 ± 5.57) and controls (40.21 ± 6.32), whereas there were no significant differences ($p>0.05$) between fertile patients and controls.

Table 1. Age distribution of the studied groups

Age (years)	Patients		Healthy control n=20	*P* value (ANOVA)
	Infertile n=30	Fertile n=25		
Range	(21-40)	(22-43)	(20-39)	
Mean ± SE	27.5 ± 2.39	27.9 ± 1.98	26.3 ± 1.35	1.39 NS

NS: Non-significant; SE: Standard error

Table 2. Mean serum levels of TNF-α among studied groups

Marker	Infertile patients N=30 Mean ± SE	Fertile patients N=25 Mean ± SE	Healthy control N=20 Mean ± SE	*P (T-test)*
Serum TNF-α (pg/ml)	39.77 ± 7.64	24.54 ± 4.60	31.22 ± 5.12	Infertile vs Fertile 0.041* Infertile vs Control 1.271 *NS* Fertile vs Control 0.881 *NS*

*: Significant; SE: Standard error; NS: Non-significant

Table 3. Mean serum levels of IL-17 among studied groups

Marker	Infertile patients N=30 Mean ± SE	Fertile patients N=25 Mean ± SE	Healthy control N=20 Mean ± SE	*P (T-test)*
Serum IL-17 (pg/ml)	72.95 ± 16.84	35.51 ± 5.57	40.21 ± 6.32	Infertile vs Fertile 0.030* Infertile vs Control 0.049* Fertile vs Control 0.221 *NS*

*: Significant; SE: Standard error; NS: Non-significant

Discussion

It has been reported that about 25-50 % of infertile women possess endometriosis and that 30-50% of endometriosis women are infertile [14]. D'Hooghe and colleagues showed that the prevalence of endometriosis is significantly higher in infertile than fertile women, as well as the infertile women are more probably to have the disease in advance stage [15]. In spite of extensive research, no agreement has been reached and various mechanisms have been suggested to explain

the association between infertility and endometriosis. These mechanisms contain distorted pelvic anatomy, altered peritoneal function, ovulatory abnormalities and endocrine, and altered humeral and cell-mediated functions in the endometrium [16].

Systemic immune modification has also been characterized in endometriosis, with activation of peripheral blood monocytes, which secrete high levels of cytokines [17]. Many studies have involved TNF-α in the progression and pathogenesis of endometriosis as well as in infertility. TNF-α concentration have been shown to exhibit significant value as a qualitative diagnostic measure of women with endometriosis [18].

The current result found significant increase in mean serum levels of TNF-α among infertile patients as compared to fertile patients, and there are no significant differences between controls and each group of patients. Similarly, Malutan and colleagues found that significantly higher serum level of TNF-α in female with endometriosis compared to healthy controls [19].

In addition, Galo and colleagues reported the serum level of TNF-α in endometriosis group was significantly higher than that women without endometriosis group, and suggested that TNF-α serum levels are good marker for diagnosis of endometriosis as noninvasive methods [20].

In concern to the significant increase of TNF-α in infertile female as compared with fertile female, there were no other similar studies to compare with current study results.

Increased levels of cytokines in the serum and peritoneal fluid of endometriosis women may reflect increased synthesis of cytokines by peritoneal lymphocytes, macrophages, ectopic endometrial implants, or mesothelial cells of the peritoneum, all of which can produce cytokines [21,22].

Other important result in this work was significant increase in IL-17 levels in infertile patients than those fertile patients and controls, whereas there are no significant differences between fertile patients and controls, these result was in agreement with other local study conducted by Ali et al. 2016 [23] who showed that mean IL-17 was significantly elevated in patient with endometriosis as compared with healthy and concluded that serum level of IL-17 could be used as marker of susceptibility in endometriosis, and may play a major role in pathogenesis of this disease. Moreover, results reported by Ahn et al. showed the presence of IL-17 in plasma samples and ectopic tissue samples from women with endometriosis [24].

In contrast to others, Malutan et al. 2015, and one year before them, Beste et al. 2014 [19,25] revealed that IL-17 levels were not detected in peritoneal fluid and serum of endometriosis patients.

Furthermore, Zhang and colleagues indicated that the concentration of IL-17 was significantly higher in case of infertility that coexist and endometriosis this result confirms current result [26]. Endometriotic lesions themselves secrete pro-inflammatory cytokines and this inflammatory state, which is thought to reduced fertility by having a toxic effect on embryos, gametes and impairing tubal motility. This finding supports the hypothesis that elevated levels of cytokines may be implicated in the pathogenesis of endometriosis associated infertility [27].

In contrast to the present results, Andreoli and colleagues showed that IL-17 level was similar between infertile and fertile patients with endometriosis [28].

In conclusion, the current results showed that TNF-α and IL-17 might play an important role in endometriosis-associated infertility.

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Conflict of interest

The author reports no conflicts of interest.

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