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THE ROLE OF SOME CYTOKINES IN ECTOPIC PREGNANCY IN IRAQ-THIQAR

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ABSTRACT

This study aimed to investigate the effected of cytokines IL1RA, IL2, IL-6, IL8, TNFα and INFα on the ectopic pregnant women.

A case-controls study conducted in the Department of Obstructive and Gynecology/Bent al Huda Hospital in IRAQ - THI-Qar governorate from April 2017 to October 2017.

Ninety individuals involved in the current study, 50 women with an ectopic pregnancy in the first trimester and 40 health pregnant women in the first trimester as controls were enrolled in this study.

Out of 50 ectopic pregnancy 23 have a risk factor. The highest percentage was showed in those with a pelvic inflammatory disease which was (43.5%), followed by the intrauterine contraceptive device (17.3%) while the smoking was lowest with the percentage of (4.3%).

The most frequency age group for ectopic pregnancy was shown in (25-34) years (42%), followed by (35-45 years) 38%. while (15-24) years showed the lowest percentage (20%).

The mean concentration level of serum interleukinsIL1RA, IL2, IL6, IL8, TNFα, INFα were measured in ectopic pregnancy and controls group.

IL1RA, IL6, IL8, TNFα, INFα serum concentration was higher in ectopic pregnancy than controls group.

Mean±SD Serum concentration levels of IL1RA for ectopic pregnancy and controls group were (9.3375±3.260ng/ml), (2.1151±1.16ng/ml) respectively and this difference was highly significant (P value 0.00), while IL2 concentration Mean±SD in ectopic pregnancy was (2.2582±1.224ng/ml) and control group was (4.9923±1.59ng/ml) (P<0.05) with no statistically significance, Serum concentration level of IL6 for ectopic pregnancy was (23.2508±7.18 Pg/ml) while controls group was (9.0025±2.74 Pg/ml) with statistically significant differences (P value 0.00), IL8 level in ectopic pregnancy and controls show (76.9892±19.8 Pg/ml), (26.5498±11.85 Pg/ml) respectively and this different were statistically significant (P value 0.00).

Serum concentration levels of TNFα for ectopic pregnancy and controls group were (31.5328±10.22 Pg/ml), (9.5346±10.22 Pg/ml) respectively this different was statistic significant (P value 0.00).

Serum concentration levels of INFα for ectopic pregnancy were (24.7813±7.60 Pg/ml) while the control group were (8.2140±2.68 Pg/ml) this different was statistic significant (P value 0.00)

The mean concentration of cytokines for different age groups show IL 1RA

Original Article

Were highly (Mean±SD10.91±3.08 ng/ml) in age group (35 – 45 year), IL 2 were highly (Mean±SD2.26±0.932 ng/ml) in age group (35 – 45 year). IL 6 were highly (Mean±SD27.021±7.628 Pg/ml) in the age group (15 – 24 years). IL 8 were highly (Mean±SD82.912±15.21 Pg/ml) in the age group (25 – 34 year). TNFα were highly (Mean±SD36.09±9.405 Pg/ml) in the age group (25 – 34 year) and INFα were highly (Mean±SD26.147±5.50 Pg/ml) in the age group (35 – 45 years).

KEYWORDS: Gynecology, Serum, Pelvic Inflammatory Disease, IL1RA, IL6, IL8, TNFα & INFα

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INTRODUCTION

Ectopic pregnancy EP is a fertilized egg when implantation outside of the endometrial cavity (Walker *et al.* 2007). Global Incidence of ectopic pregnancy increases between 1970 and 1992 (Chang *et al.* 2003). It was different from region to region with the same geographical conditions and depends on the risk factors concerning the population (Stabile *et al.* 1994). One out of every 100 pregnant women is ectopic with the most common site being within a fallopian tube (Walker *et al.* 2007). Morbidity and Mortality of ectopic pregnancy in the first trimester remains the mainly maternal mortality and morbidity (Grimes.1994, Airede. 2005, Okunlola. 2006). Maternal deaths caused by ruptured account up to 6% and 31.9 cases of deaths per 100,000 has been reported in ectopic pregnancies related to assisted reproductive technology while mortality rate has decreased (Barnhart. 2009, Perkins. 2011). From 1:2000 to 1:18,000 maternal rate of mortality and morbidity occurred in cervical ectopic pregnant women (Yankowitz *et al.* 1990). Because the early diagnosis and treatment before rupture the incidence decreased to 0.5 cases of deaths per 1000 pregnancies (Chang *et al.*, 2003). Cytokines, anti-inflammatory, and Th-2 showed to be predominated in maternal-fetal interface of normal pregnancies (Wegmann. 1993, Mor. 2003). Any disorder between anti-inflammatory and pro-inflammatory cytokines balance effected on implantation(Formby *et al.* 1995) Cytokines important for facilitating of immune modulation in safely continuous of early stages of pregnancy by switching the type 2 responses(Chaouat *et al.* 2004). During the early implantation, the chemokines and Cytokines play important roles and increase trophoblast receptors expression (Hanna. 2003, Hannan.2006, Hannan. 2008). Cytokines can effect changes in cell numbers or structure of placenta and transport function of nutrient through blastocyst gene expression (Kwong. 2000, Sjöblom. 2005). Pre-implantation mechanism in the uterus established by the role of IL-6 family (Van Mourik *et al*. 2009). Pro-inflammatory TNF, IL-6 play important roles in the implantation process and detected in the maternal-fetal interface (Jabbour *et al.* 2009) Th1 produce tumor necrosis factor α, IFN-α and interleukin 2 which promote cellular immune responses while Th2 produce IL-4, IL-5, IL-9, IL-10 and IL-13 which promote humoral responses (Mosmann. 1986, Veenstra. 2003).

The Interleukin-1ra

IL-1 agonistic and antagonistic were implicated in the implantation of embryonic and they evoke target cells to induce signal responses (Dower.1986, Mizel. 1989) Expression IL-1ra during follicular phase in Human endometrium show to be more than in luteal phase (Huang *et al.* 2006). IL-1ra suggested in prevention implantation mechanism of the embryo by the direct effect on microvillus endometrial of the mouse but this mechanism remains unclear (Simón *et al.* 1998).

Interleukin2

IL-2 produced primarily by $CD4^+$ T cells following their activation by antigen (Rubin *et al.* 1985). IL-2 have an abortogenic effect (Tezabwala *et al.* 1989).

Interleukin 6

IL-6 is produced by monocytes, macrophages, fibroblasts, endothelial cells, vascular smooth muscle cells and endometrial epithelial-stromal cells and by several endocrine glands, including the pituitary and the pancreas (Laird *et al.* 1993). Interleukin 6 has an important role in early implantation through a regulation production of ovarian steroid and in folliculogenesis (Machelon. 1994, Orsi. 2008). It also modulates secretion of other cytokines, promotes T-cell activation and B-cell differentiation and inhibits the growth of various human cell lines (Nothnick *et al.* 2001).

Interleukin 8

They produced from different cells type of including monocytes, Endothelial cells and fibroblasts (Yoshimura, 1987, Larsen, 1989, Strieter, 1989) are an inflammatory cytokine found in patients with endometriosis, particularly in the mild but most active stages of the disease (Ryan. 1995, Gazvani. 1998). The molecule mediates neutrophil migration, activation and induces the expression of leukocyte adhesion molecules (Baggiolini, 1989. Koch, 1992). Promotes endometrial stromal cell proliferation (Arici *et al*., 1998).

Tumor Necrosis Factor α

TNF α is pro-inflammatory cytokine which activates signaling pathways for cell survival, apoptosis, inflammatory responses and cellular differentiation, It is mainly secreted by macrophages, lymphoid cells, mast cells, fibroblasts and can induce cell death of certain tumor cell lines (Kawasaki *et al.* 2002). Activated macrophages and Secreted TNF-α play a critical role in the pathogenesis of endometriosis and the systemic manifestations of the disease (Braun *et al.* 2002).

Interferon α

INF α play important role in implantation, stabilize uterine in most mammals and affected the expression of many genes (Spencer. 2006, Spencer. 2008). It can also exert the induction of apoptosis and suppression of cellular growth (Rizza *et al.* 2010).

MATERIALS AND METHODS

The study was involved all reproductive age females were admiration from a Department of Obstetrics and Gynecology in Bent Al Huda Hospital in Iraq- ThiQar governorate during period from all patients data were recorded, Age, gravidity, gestational age, clinical presenting features, and ectopic risk factors (pelvic inflammatory disease, smoking, treatment of infertility, used an intrauterine contraceptive device, previous history of tubal surgery, pelvic surgery and previous ectopic). A Peak level of β-HCG, transvaginal-ultrasonographic In this study 90 cases from controls and patients groups 50 of them with pregnant women in the first trimester and 40 cases of ectopic pregnancy, Their ages were between (15-45) years and referred to the Gynecology Department from April 2016 to August 2016. Women with Ectopic pregnancy were diagnosed by measurement peak level of serum hCG concentrations, pelvic sonography and/or transvaginal ultrasonography. Patients groups were submitted full reports regarding their medical status history physical examination, laboratory tests, signs and symptoms of pelvic pain and/or amenorrhea. However, patients with recurrent

miscarriage, Hydatidi form mole, and ovarian cancer were excluded. Verbal consents of all participants were taken.

Collection of Sera

A total of 5 ml of blood was drawn from controls and patients groups.

Blood samples were centrifuged at 3000 r. p. m and sera were stored at -20 C° was obtained from all the women for (, IL-6, IL-8, IL4, TNF, INF, IL1RA assay).

Evaluation Level of Cytokines

Enzyme-linked technology (kits of Immunosorbent assay) provided by Biobase biotechnology company was used to detect the level of certain cytokines in a blood sample of both groups.

Statistical Analysis

The data were analyzed using description statistic (mean and standard deviation) independent sample t test the level significant was set at $p < 0.05$ SPSS (Statistical Packing for Social Sciences) version 20.

RESULTS

Age Group of Women Included in the Study

Women with ectopic pregnancy included in the current study were fifty. They were classified according to their age group as shown in Table 1.

The highest percentage was (42%) within the age (25-34 years), followed b (35-45 years) which was shown to be 38%. Age group between (15-24 years) showed the lowest percentage (20%).

Table 1: Frequency of the Ectopic Pregnancy Group According to their Age Group

Age Group	Frequency	Percent (%)
15-24	10	20%
25-34	21	42%
35-45	19	38%
Total	50	100%

Classification of ectopic pregnancy according to the risk factor which is noted in23 out of50 patients with ectopic pregnancy. The highest percentage was showed in those with a pelvic inflammatory disease which was 43.5% followed by Intrauterine contraceptive device was17.3% while the smoking was the lowest percentage4.3%.

Table 2: Frequency of Ectopic Pregnancy Groups According to their Risk Factors

Risk Factor	Frequency	Percent
Pelvic inflammatory disease	10	43.5%
Intrauterine contraceptive device	4	17.3%
Previous histories of ectopic pregnancy	3	13%
Tubal surgery	2	8.6%
Assisted conceptively	3	13%
Smoking	1	4.3%
Total	**23**	**100**

Mean Serum Levels of IL6, IL8, IL2, IL1RA, TNFα, INFα in Controls and Patients Group

The mean levels of serum cytokines IL6, IL8, IL1RAb, TNF, INF IL2 were measured in patients groups and control groups. IL6, IL8, IL1RAb, TNF, INF were higher in patients group with serum level (23.2508 pg/ml), (76.9892 pg/ml), (9.3375 ng/ml), (31.5328 pg/ml), (24.7813 pg/ml) respectively than control groups. The differences between patients and control groups were statistically significant (p-value <0.00) serum level of IL2 showed lower than patients (2.2582 ng/ml) the difference between patients and control was statistically not significant.

Table 3: Mean ± SD Serum Levels of IL1RA, IL2, IL6, IL8, TNFα and INFα for Patients and Controls

	No	IL1RA ng/ml	IL2 ng/ml	IL6 Pg/ml	IL8 Pg/ml	TNFα Pg/ml	INFα Pg/ml
Patients	50	9.3375 ±3.260	2.2582 ±1.224	23.2508 ±7.18	76.9892 ±19.8	31.5328 ±10.22	24.7813 ±7.60
Controls	40	2.1151 ±1.16	4.9923 ±1.59	9.0025 ±2.74	26.5498 ±11.85	9.5346 ±10.22	8.2140 ±2.68
P value		*0.00*	>0.05	0.000	0.000	0.000	0.000

Mean Concentration Level of IL1ra for Ectopic Pregnancy and Controls Group

The mean level of **IL1ra** among ectopic pregnancy was 9.3375 ng/ml, while the controls showed a level of 2.1151 ng/ml. The difference was highly significant (p-value 0.00).

Table 4: Mean ± SDIL1ra Concentration Level in Serum for Ectopic Pregnancy and Control Group

IL1ra Concentration	No	Mean Concentration	p. Value
Patient	50	9.3375±3.260 ng/ml	*0.000*
Control	40	2.1151±1.16 ng/ml	

Mean Concentration Level of Serum IL 2 in the Serum of Patients and Controls Group

The mean level of serum IL 2 in patients was 2.2582 ng/ml, while the control group was 4.9923 ng/ml, these differences was insignificant (p-value >0.05).

Table 5: Mean ± SD Concentration Level of IL2 in Serum for Ectopic Pregnancy and Controls Group

IL 2 Concentration.	No	Mean Concentration	p. Value
Patients	50	2.2582±1.224 ng/ml	>0.05
Controls	40	4.9923±1.59 ng/ml	

Mean level of IL6 in the Serum for Ectopic Pregnancy and Controls Group

The IL6 level in patients was 23.2508 pg/ml, while the group of control showed 9.0025 pg/ml. The differences were highly significant (p-value 0.00).

Table 6: Mean ± SD Concentration level of Serum IL6 for Ectopic Pregnancy and Control Group

IL 6 Concentration	No	Mean Concentration	p. Value
Patients	50	23.2508±7.18 Pg/ml	*0.000*
Controls	40	9.0025±2.74 Pg/ml	

The Mean Concentration Level of Serum IL8 for Ectopic Pregnancy and Controls Group

The mean level of IL8 in patients was 76.9892 pg/ml, while the controls were 26.5498 pg/ml. this differences showed highly significant (p-value 0.00).

Table 7: Mean ± SD Concentration IL8 in the Serum of Ectopic Pregnancy and Controls Group

IL8 Concentration	No	Mean Concentration	p. Value
Patients	50	76.9892±19.8 Pg/ml	*0.000*
Controls	40	26.5498±11.85 Pg/ml	

Mean Concentration of Serum TNFα for Ectopic Pregnancy and Controls Group

The mean concentration level of serum TNFα were 31.5328 pg/ml, while the controls were 9.5346 pg/ml. the differences were highly significant (p-value 0.00).

Table 8: Mean ± SD Concentration of TNFα in Serum of Ectopic Pregnancy and Controls Group

TNFα Concentration.	No	Mean Concentration	p. Value
Patients	50	31.5328±10.22 Pg/ml	*0.000*
Controls	40	9.5346±10.22 Pg/ml	

Mean Concentration of the Serum INFα for Ectopic Pregnancy and Controls Group

The mean level of INFα in patients was 31.5328 pg/ml, while the controls showed 9.5346 pg/ml. The differences were highly significant (p-value 0.00).

Table 9: Mean ± SD Concentration Level of INFα in Serum of Ectopic Pregnancy and Controls Group

INFα Concentration	No	Mean Concentration	p. Value
Patients	50	24.7813±7.60 Pg/ml	*0.000*
Controls	40	8.2140±2.68 Pg/ml	

Table 10: Mean Concentration of Cytokines for Different Age Groups

AGE GROUPS	IL1RA ng/ml	IL2 ng/ml	IL6 Pg/ml	IL8 Pg/ml	TNFα Pg/ml	INFα Pg/ml
15 – 24 yrs	8.358±2.72	1.44±1.30	27.021±7.628	80.76±17.33	31.84±7.365	23.65±6.94
25 – 34 yrs	9.162±2.731	2.240±1.212	23.35±6.364	82.912±15.21	36.09±9.405	25.84±7.96
35 – 45 yrs	10.91±3.08	2.26±0.932	22.925±5.225	77.27±19.69	28.71±7.842	26.147±5.50

DISCUSSIONS

Ectopic pregnancy is until a public health problem facing coupling expecting a child. Research has opened the probability of immune response to such pregnancy complications with special emphasis on the effects of pro-inflammatory and anti-inflammatory cytokines on the success or failure of pregnancy

Demographic Property

In the present study serum concentrations of IL-6, IL-8, IL1RA, interferon alpha and TNFα are higher in women with ectopic pregnancy while IL2 was lower than the control group. In comparison with the values of the cytokines with a first-trimester pregnancy.

Case-controls study sought to determination the relative between up and down regulation of the cytokines of ectopic pregnancy as for as age groups are concerned. Women with age of 25-34 years showed a higher rate of ectopic pregnancy, this is in agreement with Opoku *et al*. (2013) which is possibly due to high reproductive activity

Interleukin 1RA

The mean level of serum Interleukin 1RA among ectopic pregnant women was highly elevated and there is statistical significant this finding was in agreement with that reported by Lekovic et al. (2013) in the United States. IL-1 play important role in regulation implantation of the embryo (Simon *et al.* 1995). IL-1RA, on the other hand, acts as a competitive inhibitor by binding to the IL-1 receptor without recruiting the IL-1 receptor-associated protein to the complex, therefore not inducing any intracellular signaling response (Arend *et al.* 1991). The blockade of IL-1 receptors by antagonists during the period before implantation results in inhibition and prevention of embryonic implantation. (Simon *et al.* 1994). A recent study reported failure implantation of the blastocyst and suggested inhibition implantation of the embryo in pregnant mice when injected with IL1ra (Simon *et al.*1994).

Interleukin 2

The mean concentration level of serum Interleukin 2 among ectopic pregnant women showed low than controls groups ,no statistically significant and this finding was in agreement with reported by Soriano *et al*. (2003) in France. In contrast to MacLean *et al* (1991). Who that finding a rise in IL-2 receptor levels in ectopic pregnancy. IL-2 indicates activation of the immune system, and thus monitoring of IL-2 receptor levels may lead to a more accurate prediction of the outcome of the pregnancy (MacLean *et al.* 1991). During pregnancy, inflammatory factors secretion by natural killer cells, and HLA-Class II molecules secretion promote by T-cells that lead to rejection of embryo, natural killer cells and T cells activated by IL-2 (Fu *et al.* 2007).

Interleukin 6

The mean levels of Th2 cytokineIL6 for ectopic pregnancy were high than controls groups and show association significant. This finding was in agreement with findings reported by two studies (Soriano. 2003, Farquhar. 2005). Increased levels of IL-6 in amniotic fluid were associated with membrane rupture of premature and infection of intrauterine (Fukuda *et al*. 2002). IL-6 signaling may be important for transporting embryo, therefore any abnormal levels of IL-6 alter tubal transport (Shao R *et al.* 2009). Interleukin-6 (IL-6) has embryotoxic effected and inhibits implantation of the blastocyst (Jacobs. 1992, Hill JA. 1992) IL-6 has anti-inflammatory effects and leading to induce of HCG from trophoblast, which promotes secretion of progesterone (Hill *et al.* 1992). A recent study reported reduces the tubal ciliary activity of epithelial when treated withIL-6 *in vitro* and restores ciliary activity when used anti-IL-6 (Papathanasiou *et al.* 2008). However, it can be stated that Th2 cytokines might be associated with successful pregnancy.

Interleukin 8

The mean levels of Th2 cytokine IL8 for ectopic pregnancy were higher than the controls groups and show statistically significance and this finding was in agreement with that reported by Soriano *et al*. (2003). Interleukin 8 induced by inflammation in the fallopian tube, may provide signals to the embryo and promote tubal implantation (Shao *et al*. 2007). IL-8 are inflammatory cytokines and its highly elevated in the Fallopian tube infection by *Chlamydia* (Mpiga *et al*. 2006) and also in ectopic implantation (Balasubramaniam *et al.* 2012). Implantation of the embryo in uterus stimulation the local inflammatory response in a similar way as leukocytes accumulates at inflammatory sites of implantation

(Dominguez *et al*., 2005). Another study found the potential utility of IL-8 in diagnosing an ectopic pregnancy, as compared to normal intrauterine pregnancy and miscarriage (Rajendiran *et al.* 2016). In contrast to, Rausch *et al* (2012). Who observed lower values of IL-8 and TNF-α in women with ectopic pregnancy.

Tumor Necrosis Factor Alpha

The result of the current study showed the mean level of tumor necrosis factor alpha was higher in patients with ectopic pregnancy compared to control group the difference were highly statistically significant so that finding was not in agreement with Rausch *et al.* (2012). Therefore the current study were agreement with the role of affected tumor necrosis factor is harmful to pregnancy because they cause embryonic inhibition and in development fet al (Chaouat. 1990, Haimovici. 1991). And in agreement with reported by (Soriano *et al.* 2003). TNF-α has been proposed to act as a mediator of detrimental stimuli inducing embryonic death by activating apoptosis of trophoblast cells, whereas IFN-γ augments TNF-mediated death of trophoblasts (Robaye.1991, Yui J. 1994). Several studies have shown elevated serum TNF-a levels in patients with pregnancies problem compared with normal pregnancies (Vassiliadis. 1998, Gucer. 2001). A recent study has proposed a possible protective role of TNF-α at the fetoplacental unit with an implication in mechanisms preventing the occurrence of malformed offspring (Toder *et al*. 2003). Pathophysiological effects failure in implantation and endometriosis showed to be associated with increased expression of TNFα (Cao WG *et al*. 2005). TNF was shown to increase hCG secretion of JAR cells, whereas the cytokine reduced hCG secretion from term placenta ciliated sloughing of tubal epithelial mucosa cells associated with up-regulation in TNF-α (McGee *et al*. 1999). One study found that higher IL-8, IL-6, and TNF-α were present in women with EP when compared to pregnant with normal pregnancy and, miscarriage though only IL (Rausch *et al*. 2011).

Interferon Alpha

The mean level of serum INFα in patients was 31.5328 pg/ml, while the controls were high level 9.5346 pg/ml. To the best of my knowledge, there are no published studies between the relationship of INFα with ectopic pregnancy

CONCLUSIONS

- The present study revealed a significant indicator between cytokines levels and ectopic pregnancy.
- The cytokines were detected in patients with ectopic pregnancy and control group in first trimester.
- There is a significant associations IL-1ra, IL-6, IL-8 , TNFα and INFα levels of ectopic pregnancy
- There is no significant association between IL-2 level of and ectopic pregnancy.
- The higher concentration of cytokines (IL 1RA, IL 2, INFα)was found among age group (35– 45 years), while TNFα and IL8 highly in age group (25 – 34 year) and IL 6 concentration mean was in age group (15 – 24 years).
- Although a direct causal relationship between cytokines levels of ectopic pregnancy has not been determined, differences, in cytokines activity have been reported for normal pregnant women and those of ectopic pregnancy.
- Mean concentration of interleukin 1ra (9.3375±3.260 ng/ml) were higher in study group than control group (2.1151±1.16 ng/ml).

- Mean concentration of interleukin 2 (2.2582±1.224 ng/ml) were lower in study group than control group (4.9923±1.59 ng/ml).
- Mean concentration of interleukin 6 (23.2508±7.18 Pg/ml) were higher in study group than control group (9.0025±2.74 Pg/ml).
- Mean concentration of interleukin 8 (76.9892±19.8 Pg/ml) were higher in study group than control group (26.5498±11.85 Pg/ml).
- Mean concentration of tumor necrosis factor α (31.5328±10.22 Pg/ml) were higher in study group than control group (9.5346±10.22 Pg/ml).
- Mean concentration of interferon α (24.7813±7.60 Pg/ml) were higher in study group than control group (8.2140±2.68 Pg/ml).

REFERENCES

1. *Abdul IF. Ectopic pregnancy in Ilorin, Nigeria. International Journal of Gynecology & Obstetrics. 1999* Aug 1;*66*(*2*):*179-80*
2. *Airede LR, Ekele BA. Ectopic pregnancy in Sokoto, Northern Nigeria. Malawi Medical Journal. 2005*;*17*(*1*):*14-6*.
3. *Arend WP. Interleukin 1 receptor antagonist. A new member of the interleukin 1 family. The Journal of clinical investigation. 1991* Nov 1;88(*5*):*1445-51*.
4. *Arici A, Seli E, Senturk LM, Gutierrez LS, Oral E, Taylor HS. Interleukin-8 in the human endometrium. The Journal of Clinical Endocrinology & Metabolism. 1998* May 1;*83*(*5*):1783-*7*.
5. *Baggiolini M, Walz A, Kunkel SL. Neutrophil-activating peptide-1/interleukin 8, a novel cytokine that activates neutrophils. Journal of Clinical Investigation. 1989* Oct;*84*(*4*):*1045*.
6. *Balasubramaniam ES, Van Noorden S, El-Bahrawy M. The expression of interleukin (IL)-6, IL-8, and their receptors in fallopian tubes with ectopic tubal gestation. Fertility and Sterility. 2012* Oct 31;*98*(*4*):*898-904*.
7. *Barnhart KT. Ectopic pregnancy. New England Journal of Medicine. 2009* Jul 23;*361*(*4*):*379-87*.
8. *Braun DP, Ding J, Dmowski WP. Peritoneal fluid-mediated enhancement of eutopic and ectopic endometrial cell proliferation is dependent on tumor necrosis factor-α in women with endometriosis. Fertility and Sterility. 2002* Oct 31;*78*(*4*):*727-32*
9. *Cao WG, Morin M, Sengers V, Metz C, Roger T, Maheux R, Akoum A. Tumour necrosis factor-α up-regulates macrophage migration inhibitory factor expression in endometrial stromal cells via the nuclear transcription factor NF-κB. Human Reproduction. 2005* Oct 6;*21*(*2*):*421-8*.
10. *Chang J, Elam-Evans LD, Berg CJ, Herndon J, Flowers L, Seed KA, Syverson CJ. Pregnancy-related mortality surveillance—United States, 1991–1999. MMWR SurveillSumm. 2003* Feb 21;*52*(*2*):*1-8*.
11. *Chaouat G, Ledée-Bataille N, Dubanchet S, Zourbas S, Sandra O, Martal J. TH1/TH2 paradigm in pregnancy: paradigm lost?. International archives of allergy and immunology. 2004*;*134*(*2*):*93-119*.
12. *Chaouat G, Menu E, Clark DA, Dy M, Minkowski M, Wegmann TG. Control of fet al survival in CBA× DBA/2 mice by lymphokine therapy. Journal of Reproduction and Fertility. 1990* Jul *1*;*89*(*2*):*447-58*.
13. *Dominguez F, Yanez-Mo M, Sanchez-Madrid F, Simon C. Embryonic implantation and leukocyte transendothelial migration: different processes with similar players?. The FASEB journal. 2005* Jul 1;*19*(*9*):*1056-60*.

14. *Dower SK, Kronheim SR, Hopp TP, Cantrell M, Deeley M, Gillis S, Henney CS, Urdal DL. The cell surface receptors for interleukin-1α and interleukin-1β are identical. Nature. 1986* Nov;*324*(*6094*):*266*.
15. *Farquhar CM. Ectopic pregnancy. Lancet. 2005*;*366*:*583–591*.
16. *Formby B. Immunologic response in pregnancy. Its role in endocrine disorders of pregnancy and influence on the course of maternal autoimmune diseases. Endocrinology and Metabolism Clinics of North America. 1995* Mar;*24*(*1*):*187-205*.
17. *Fu W, Shi B. Correlation of T lymphocyte subsets and serum IL-2, IL-10 with spontaneous abortion. Jiangsu Medical Journal. 2007*;*33*(*4*):*328*.
18. *Fkuda H, Masuzaki H, Ishimaru T. Interleukin□6 and interleukin□1 receptor antagonist in amniotic fluid and cord blood in patients with pre□term, premature rupture of the membranes. International Journal of Gynecology & Obstetrics. 2002* May 1;*77*(*2*):*123-9*.
19. *Gazvani MR, Christmas S, Quenby S, Kirwan J, Johnson PM, Kingsland CR. Peritoneal fluid concentrations of iterleukin-8 in women with endometriosis: relationship to stage of disease. Human Reproduction* (*Oxford, England*). *1998* Jul 1;*13*(*7*):*1957-61*.
20. *Grimes DA. The morbidity and mortality of pregnancy: still risky business. American Journal of Obstetrics and Gynecology. 1994* May 1;*170*(*5*):*1489-94*.
21. *Gücer F, Balkanli-Kaplan P, Yüksel M, Sayin NC, Yüce MA, Yardim T. Maternal serum levels of tumor necrosis factor-α ad interleukin-2 receptor in threatened abortion: a comparison with normal and pathologic pregnancies. Fertility and Sterility. 2001* Oct 31;*76*(*4*):*707-11*.
22. *Haimovici F, Hill JA, Anderson DJ. The effects of soluble products of activated lymphocytes and macrophages on blastocyst implantation events in vitro. Biology of Reproduction. 1991* Jan 1;*44*(*1*):*69-75*.
23. *Hanna J, Wald O, Goldman-Wohl D, Prus D, Markel G, Gazit R, Katz G, Haimov-Kochman R, Fujii N, Yagel S, Peled A. CXCL12 expression by invasive trophoblasts induces the specific migration of CD16–human natural killer cells. Blood. 2003* Sep 1;*102*(*5*):*1569-77*.
24. *Hannan NJ, Jones RL, White CA, Salamonsen LA. The chemokines, CX3CL1, CCL14, and CCL4, promote human trophoblast migration at the feto-maternal interface. Biology of Reproduction. 2006* May 1;*74*(*5*):*896-904*.
25. *Hannan NJ, Salamonsen LA. CX3CL1 and CCL14 regulate extracellular matrix and adhesion molecules in the trophoblast: potential roles in human embryo implantation. Biology of Reproduction. 2008* Jul 1;*79*(*1*):*58-65*.
26. *HILL JA. Cytokines considered critical in pregnancy. American Journal of Reproductive Immunology. 1992* Oct 12;*28*(*3□4*):*123-6*.
27. *Huang H. The cytokine network during embryo implantation. Chang Gung medical journal. 2006* Jan;*29*(*1*):*25*.
28. *Jabbour HN, Sales KJ, Catalano RD, Norman JE. Inflammatory pathways in female reproductive health and disease. Reproduction. 2009* Dec 1;*138*(*6*):*903-19*.
29. *Jacobs AL, Sehgal PB, Julian JO, Carson DD. Secretion and hormonal regulation of interleukin-6 production by mouse uterine stromal and polarized epithelial cells cultured in vitro. Endocrinology. 1992* Sep 1;*131*(*3*):*1037-46*.
30. *Kawasaki H, Onuki R, Suyama E, Taira K. Identification of genes that function in the TNF-α-mediated apoptotic pathway using randomized hybrid ribozyme libraries. Nature Biotechnology. 2002* Apr 1;*20*(*4*):*376-80*.

31. *Koch AE, Polverini PJ, Kunkel SL, Harlow LA, DiPietro LA, Elner VM, Elner SG, Strieter RM. Interleukin-8 as a macrophage-derived mediator of angiogenesis. Science. 1992* Dec 11;*258*(*5089*):*1798-801*.
32. *Kwong WY, Wild AE, Roberts P, Willis AC, Fleming TP. Maternal undernutrition during the preimplantation period of rat development causes blastocyst abnormalities and programming of postnatal hypertension. Development. 2000* Oct 1;*127*(*19*):*4195-202*.
33. *Laird SM, Li TC, Bolton AE. The production of placental protein 14 and interleukin 6 by human endometrial cells in culture. Human Reproduction. 1993* Jun 1;*8*(*6*):*793-8*.
34. *Larsen CG, Anderson AO, Oppenheim JJ, Matsushima K. Production of interleukin-8 by human dermal fibroblasts and keratinocytes in response to interleukin-1 or tumour necrosis factor. Immunology. 1989* Sep;*68*(*1*):*31*.
35. *Lekovic J, Doulaveris G, Orfanelli T, Shulman B, Witkin SS, Spandorfer SD. Interleukin-1β and Interleukin-1 receptor antagonist serum levels one week after embryo transfer may predict ectopic pregnancy in IVF cycles. Fertility and Sterility. 2013* Sep 1;*100*(*3*):*S138*.
36. *Machelon V, Emilie D, Lefevre A, Nome F, Durand-Gasselin I, Testart J. Interleukin-6 biosynthesis in human preovulatory follicles: some of its potential roles at ovulation. The Journal of Clinical Endocrinology & Metabolism. 1994* Aug;*79*(*2*):*633-42*.
37. *MacLean MA, Wilson R, Thomson JA, Krishnamurthy S, Walker JJ. Changes in immunologic parameters in normal pregnancy and spontaneous abortion. American Journal of Obstetrics and Gynecology. 1991* Oct 1;*165*(*4*):*890-5*.
38. *Mcgee ZA, Jensen RL, Clemens CM, Taylor-Robinson D, Johnson AP, Gregg CR. Gonococcal Infection of Human Fallopian Tube Mucosa in Organ Culture: Relationship of Mucosal Tissue TNF□α Concentration to Sloughing of Ciliated Cells. Sexually Transmitted Diseases. 1999* Mar 1;*26*(*3*):*160-5*.
39. *Mizel SB. The interleukins. FASEB Journal 1989;3:2379-88*.
40. *Mor G, Abrahams VM. Potential role of macrophages as immunoregulators of pregnancy. Reproductive Biology and Endocrinology. 2003* Dec 2;*1*(*1*):*119*.
41. *Mosmann TR, Cherwinski H, Bond MW, Giedlin MA, Coffman RL. Two types of murine helper T cell clone. I. Definition according to profiles of lymphokine activities and secreted proteins. The Journal of Immunology. 1986* Apr 1;*136*(*7*):*2348-57*.
42. *Mpiga P, Mansour S, Morisset R, Beaulieu R, Ravaoarinoro M. Sustained Interleukin□6 and Interleukin□8 Expression Following Infection with Chlamydia trachomatis Serovar L2 in a HeLa/THP□1 Cell Co□culture Model. Scandinavian Journal of Immunology. 2006* Mar 1;*63*(*3*):*199-207*.
43. *Nothnick WB. Treating endometriosis as an autoimmune disease. FertilSteril 2001;76:223– 31*.
44. *Okunlola MA, Adesina OA, Adekunle AO. Repeat ipsilateral ectopic gestation: a series of 3 cases. African Journal of Medicine and Medical Sciences. 2006* Jun;*35*(*2*):*173-5*.
45. *Opoku BK, Nguah SB, Azanu W. Ectopic pregnancy: Are fair-colored women at increased risk?. Gynecology. 2013* Apr 1;*1*(*1*):*1*.
46. *Orsi NM, Tribe RM. Cytokine networks and the regulation of uterine function in pregnancy and parturition. Journal of Neuroendocrinology. 2008* Apr *1*;*20*(*4*):*462-9*.
47. *Papathanasiou A, Djahanbakhch O, Saridogan E, Lyons RA. The effect of interleukin-6 on ciliary beat frequency in the human fallopian tube. Fertility and Sterility. 2008* Aug 31;*90*(*2*):*391-4*.

48. *Rajendiran S, Senthil Kumar GP, Nimesh A, Dhiman P, Shivaraman K, Soundararaghavan S. Diagnostic significance of IL-6 and IL-8 in tubal ectopic pregnancy. Journal of Obstetrics and Gynaecology. 2016* Oct 2;*36*(*7*):*909-11*.

49. *Rausch ME, Barnhart K. Serum biomarkers for detecting ectopic pregnancy. Clinical Obstetrics and Gynecology. 2012* Jun;*55*(*2*):*418*.

50. *Rausch ME, Sammel MD, Takacs P, Chung K, Shaunik A, Barnhart KT. Development of a multiple marker test for ectopic pregnancy. Obstetrics & Gynecology. 2011* Mar 1;*117*(*3*):*573-82*.

51. *Rizza P, Moretti F, Belardelli F. Recent advances on the immunomodulatory effects of IFN-α: implications for cancer immunotherapy and autoimmunity. Autoimmunity. 2010* May 1;*43*(*3*):*204-9*.

52. *Robaye B, Mosselmans R, Fiers W, Dumont JE, Galand P. Tumor necrosis factor induces apoptosis (programmed cell death) in normal endothelial cells in vitro. The American Journal of Pathology. 1991* Feb;*138*(*2*):*447*.

53. *Rubin LA, Kurman CC, Fritz ME, Biddison WE, Boutin BE, Yarchoan RO, Nelson DL. Soluble interleukin 2 receptors are released from activated human lymphoid cells in vitro. The Journal of Immunology. 1985* Nov 1;*135*(*5*):*3172-7*.

54. *Ryan IP, Tseng JF, Schriock ED, Khorram O, Landers DV, Taylor RN. Interleukin-8 concentrations are elevated in peritoneal fluid of women with endometriosis. Fertility and Sterility. 1995* Apr 30;*63*(*4*):*929-32*.

55. *Shao R, Egecioglu E, Weijdegård B, Kopchick JJ, Fernandez-Rodriguez J, Andersson N, Billig H. Dynamic regulation of estrogen receptor-α isoform expression in the mouse fallopian tube: mechanistic insight into estrogen-dependent production and secretion of insulin-like growth factors. American Journal of Physiology-Endocrinology and Metabolism. 2007* Nov 1;*293*(*5*):*E1430-42*.

56. *Shao R, Nutu M, Karlsson-Lindahl L, Benrick A, Weijdegård B, Lager S, Egecioglu E, Fernandez-Rodriguez J, Gemzell-Danielsson K, Ohlsson C, Jansson JO, Billig H. Downregulation of cilia-localized Il-6R alpha by 17beta-estradiol in mouse and human fallopian tubes. Am J Physiol Cell Physiol. 2009*;*297*:*C140–C151*

57. *Simón C, Pellicer A, Polan ML. Interleukin-1 system crosstalk between embryo and endometrium in implantation. Human Reproduction. 1995* Dec 1;*10*(suppl_*2*):*43-54*.

58. *Simón C, Valbuena D, Krüssel J, Bernal A, Murphy CR, Shaw T, Pellicer A, Polan ML. Interleukin-1 receptor antagonist prevents embryonic implantation by a direct effect on the endometrial epithelium. Fertility and Sterility. 1998* Nov 30;*70*(*5*):*896-906*.

59. *Simón CA, Piquette GN, Frances A, el-Danasouri IM, Irwin JC, Polan ML. The effect of interleukin-1 beta (IL-1 beta) on the regulation of IL-1 receptor type I messenger ribonucleic acid and protein levels in cultured human endometrial stromal and glandular cells. The Journal of Clinical Endocrinology & Metabolism. 1994* Mar;*78*(*3*):*675-82*.

60. *Sjöblom C, Roberts CT, Wikland M, Robertson SA. Granulocyte-macrophage colony-stimulating factor alleviates adverse consequences of embryo culture on fet al growth trajectory and placental morphogenesis. Endocrinology. 2005* May 1;*146*(*5*):*2142-53*.

61. *Soriano D, Hugol D, Quang NT, Darai E. Serum concentrations of interleukin-2R (IL-2R), IL-6, IL-8, and tumor necrosis factor alpha in patients with ectopic pregnancy. Fertility and Sterility. 2003* Apr 30;*79*(*4*):*975-80*.

62. *Spencer TE, Johnson GA, Bazer FW, Burghardt RC, Palmarini M. Pregnancy recognition and conceptus implantation in domestic ruminants: roles of progesterone, interferons and endogenous retroviruses. Reproduction, Fertility and Development. 2006* Dec 12;*19*(*1*):*65-78*.

63. *Spencer TE, Sandra O, Wolf E. Genes involved in conceptus–endometrial interactions in ruminants: insights from reductionism and thoughts on holistic approaches. Reproduction. 2008* Feb 1;*135*(*2*):*165-79*.
64. *Stabile I, Grudzinskas JG. Ectopic pregnancy: what's new. Progress in Obstetrics and Gynaecology. 1994*;*11*:*281-305*.
65. *Strieter RM, Kunkel SL, Showell HJ, Remick DG, Phan SH, Ward PA, Marks RM. Endothelial cell gene expression of a neutrophil chemotactic factor by TNF-alpha, LPS, and IL-1 beta. Science. 1989* Mar 17;*243*(*4897*):*1467-9*.
66. *Kadhim, H. N., Hashim, E. A., & Abdulla, S. A. B. Comparative Study Of Gonadotropin-Releasing Hormone Receptor In Fallopian Tube By Immunohistochemistry Among Women With Ectopic Pregnancy, Hysterectomy And Tubal Ligation.*
67. *Tezabwala BU, Johnson PM, Rees RC. Inhibition of pregnancy viability in mice following IL-2 administration. Immunology. 1989* May;*67*(*1*):*115*.
68. *Toder V, Fein A, Carp H, Torchinsky A. TNF-α in pregnancy loss and embryo maldevelopment: a mediator of detrimental stimuli or a protector of the fetoplacental unit?. Journal of assisted Reproduction and Genetics. 2003* Feb 1;*20*(*2*):*73-81*.
69. *Van Mourik MS, Macklon NS, Heijnen CJ. Embryonic implantation: cytokines, adhesion molecules, and immune cells in establishing an implantation environment. Journal of Leukocyte Biology. 2009* Jan 1;*85*(*1*):*4-19*.
70. *Vassiliadis S, Ranella A, Papadimitriou L, Makrygiannakis A, Athanassakis I. Serum levels of pro-and anti-inflammatory cytokines in non-pregnant women, during pregnancy, labour and abortion. Mediators of Inflammation. 1998*;*7*(*2*):*69-72*.
71. *Veenstra van Nieuwenhoven AL, Heineman MJ, Faas MM. The immunology of successful pregnancy. Human Reproduction Update. 2003* Jul 1;*9*(*4*):*347-57*.
72. *Wegmann TG, Lin H, Guilbert L, Mosmann TR. Bidirectional cytokine interactions in the maternal-fet al relationship: is successful pregnancy a TH2 phenomenon?. Immunology today. 1993* Jul 1;*14*(*7*):*353-6*.
73. *Yankowitz J, Leake J, Huggins G, Gazaway P, Gates E. Cervical ectopic pregnancy: review of the literature and report of a case treated by single-dose methotrexate therapy. Obstetrical & Gynecological Survey. 1990* Jul 1;*45*(*7*):*405-14*.
74. *Yoshimura T, Matsushima K, Oppenheim JJ, Leonard EJ. Neutrophil chemotactic factor produced by lipopolysaccharide (LPS)-stimulated human blood mononuclear leukocytes: partial characterization and separation from interleukin 1 (IL 1). The Journal of Immunology. 1987* Aug 1;*139*(3):*788-93*.
75. *Yui J, Garcia-Lloret M, Wegmann TE, Guilbert LJ. Cytotoxicity of tumour necrosis factor-alpha and gamma-interferon against primary human placental trophoblasts. Placenta. 1994 Dec 1;15(8):819-35.*