A Significant Correlation of Glutamine and Interlukin-6 with Pregnancy Rate in Women Undergoing ICSI

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KEYWORDS

Follicular fluid, ICSI, Pregnancy rate.

ABSTRACT

Background : The composition of follicular fluid (FF) is significantly influenced by metabolic factors, and the analyses of metabolites in FF could reveal the correlation between metabolites and oocyte quality, embryo development, and pregnancy outcome. FF provides nutrients and growth factors that promote oocyte growth, development, and maturation.

Objective: To investigate the relationship between serum and FF glutamine and FF IL-6 levels and their influence on the pregnancy rate in women who are undergoing an ICSI treatment cycle.

Materials and Methods: A prospective cross-sectional study was performed on 90 women aged 20- 44 years who underwent ICSI treatment from July 2023 to February 2024. Serum and follicular fluid collected on the day of oocyte pick-up were used as biological material. The embryological laboratory examined the retrieved follicular fluid and collected oocytes using stereomicroscopy. ELISA analysed serum, FF glutamine and FF IL-6. The ICSI method was utilized for fertilization. The pregnancy test is done by measuring serum BHCG. Statistical data processing was conducted using the software package SPSS 23.

Results: There was a highly significant correlation between FF glutamine and FF IL-6 ($p < 0.001$) and a positive correlation between FF glutamine and FF IL-6 with pregnancy rate ($p < 0.001$) ($p < 0.001$).

Conclusion The correlation of FF glutamine and FF IL-6 affects the pregnancy rate in women undergoing ICSI. FF glutamine and FF IL-6 can be considered predictors for pregnancy while serum glutamine has no significance.

1. Introduction

Glutamine plays a crucial role in the physiology of reproduction, particularly in the development and maturation of oocytes and follicles (Vardhana, S. A., et al 2019 [1]). Glutamine is essential for oocyte maturation, as it provides nutrients for the oocyte and helps preserve the granulosa cells (GC) wall(Zhang, KH., et al. 2024[2]).

Ovarian glutamine and ovarian interleukin-6 (IL-6) are interconnected in the area of ovarian function and disease.(**Ma, G., et al. 2022**[3]) Glutamine is a vital amino acid that acts as a precursor for multiple metabolic processes, such as generating energy and synthesizing proteins. It also has a vital function in cell growth and multiplication. (**Escobar-Morreale,. et al.2011[4])**

Interleukin-6 (IL-6) is a cytokine that controls the body's immunological responses and inflammation. It performs various roles in different tissues, including the ovaries. Within the ovarian setting, IL-6 can be generated by multiple cell types, such as granulosa cells, theca cells, and immune cells that infiltrate the ovarian tissue.

Metabolic regulation of glutamine can impact the generation of cytokines, specifically IL-6. Alterations in glutamine availability or metabolism can affect the synthesis or function of IL-6 in ovarian cells. .(**Ma, G., et al 2022[3]) (Escobar-Morreale,2011[4])**The presence of IL-6 can contribute to the occurrence of inflammation in the ovary. Glutamine metabolism could impact the inflammatory response by regulating the synthesis of IL-6 or the signaling pathways related to IL-6.(**Zhang, Y., 2021[5]).**

Regarding ovarian function, glutamine and IL-6 can both affect ovarian function. Glutamine is necessary for the growth of follicles, the maturation of oocytes, and the creation of hormones in the

ovary. IL-6 can impact the production of ovarian hormones, the growth of follicles, and the process of ovulation.(**Browning, L., 2018[**6]) Imbalances in the regulation of glutamine metabolism or IL-6 signalling can play a role in ovarian disorders like polycystic ovary syndrome (PCOS), ovarian cancer, or ovarian inflammation. Studying the relationship between glutamine and IL-6 in these conditions could offer valuable information about the causes of these diseases and potential targets for treatment.(**Rogeri, P. S., 2020[7]).**

This study examined the levels of glutamine in both the serum and follicular fluid, as well as the levels of IL-6 in the follicular fluid acquired from stimulated cycles. The main objective was to examine the potential influence of glutamine levels in the serum and follicular fluid, as well as follicular fluid IL-6, on the quality of oocytes and embryos, and ultimately, the outcome of ICSI.

Patient, Material, and Method

2-1 Study population

A prospective cross-sectional study was conducted at the High Institute for Infertility Diagnosis and Assisted Reproductive Technologies/ Al-Nahrain University, from July 2023 to February 2024. The study received approval from the Local Medical Ethical Committee. The research involved 90 infertile women who completed an ICSI protocol, ensuring they were fully informed about the study's aims and completed a consent form. The age range was between 20 and 44 years, and their time of infertility lasted from one to twelve years. The study involved women with infertility, both primary and secondary cases, some with no prior experience with IVF treatment, and others with prior experience.

Baseline laboratory evaluation includes testing for hepatitis B virus, hepatitis C virus, human immunodeficiency virus, On the second or third day of the menstrual cycle measuring estradiol, progesterone, LH, FSH, testosterone, prolactin, thyroid-stimulating hormone (TSH), triiodothyronine (FT3), thyroxine (FT4), antiMullerian hormone (AMH), and transvaginal ultrasound. The fallopian tube patency can also be assessed through hysterosalpingography or saline infusion sonohysterography.

Inclusion criteria involve all women who had undergone ICSI protocol and reached the day of embryo transfer. The study employed the following criteria for excluding participants: Women who have a body mass index (BMI) greater than 32 kg/m2, endometriosis, abnormalities, and benign tumors in the uterus, fallopian tubes, and ovaries that interfere with the process of ICSI, any medical conditions (such as cardiovascular, immunological, infectious, neurological, or psychiatric diseases.

2-2 Material and Methods

2-2-1 Stimulation of ovulation

This study used the flexible GnRH antagonist strategy, administering recombinant FSH-Gonal F (75IU) on the second day of the menstrual cycle. The first dosage of FSH ranged from 150 to 300 IU per day, based on the patient's clinical state, including factors such as age, antral follicle count, AMH levels, BMI, and prior cycle results, if applicable. The GnRH antagonist, Cetrorelix acetate (Cetrotide) vial 0.25 mg Merck-Serono/Germany, was given at a daily dose of 0.25 mg until the day of hCG injection, which occurred when the leading follicle reached a diameter of 12-14 mm (Patrizio P,.2018 [8])
Ovulation was induced by delivering Human chorionic gonadotrophin (HCG) Ovitrelle 250 microgram/0.5 ml prefilled syringe Merck-Serono/Germany through a subcutaneous injection.

Oocyte retrieval was performed under general or spinal anesthesia using transvaginal ultrasound-guided aspiration 34-36 hours after a trigger shot and before physiological ovulation. A single-lumen needle(Gynetics®, Belgium) is attached to a suction device used for retrieval. The FF was taken to an

embryologist to collect COC, and FF samples were collected, centrifuged at 3000 rpm for 10 minutes to remove debris, and stored at -20℃ for analysis. These samples were then used to measure follicular FF glutamine. On the day of oocyte pick-up, 5ml of venous blood was collected from women, clotted at room temperature for 15 minutes, and centrifuged at 3000 rpm for 10 minutes. Serum samples were stored at -20℃ in Eppendorf tubes to measure serum glutamine by using Glutamine ELISA KIT (CES124Ge)/ Cloud- Clone corp/china.and IL-6 using IL 6 ELISA KIT (SEA079Hu)/Cloud-Clone Corp/China.

Semen evaluation and preparation: Samples were collected in a dedicated room adjacent to the laboratory, abstaining from sexual activity for 2 to 5 days. The samples were masturbated and ejaculated into a clean container, maintained at a constant temperature of 20°C to 37°C, and marked with the husband's information. The container was placed on a bench or in an incubator, set at a temperature of 37 °C until the semen reached a liquid state.

Oocyte preparation and maturity evaluation: The COCs were collected and rinsed with flushing media, then placed in Ferticult flushing media/ FertiPro NV/Belgium and washed in Gain medium. The COCs were covered with FerticultTM Mineral oil /FertiPro NV/Belgium and transported to an incubator at 37°C with a CO2 concentration of 5%. The cumulus cells were removed 38 to 42 hours after the trigger injection at the Laminar Flow Cabinet. The cumulus was then immersed in a solution of Hyaluronidase medium 80IU/ml / FertiPro NV/Belgium, present in Ferticult flushing media. This media also contained 4g/L of human serum albumin (HSA), 4-(2-hydroxyethyl)-1piperazineethanesulfonic acid (HEPES), phenol red, and 10 mg/L of gentamycin.
The cumulus was mechanically separated from the oocyte using an EZ-Grip micro pipettor/ Cooper Surgical Fertility Companies, Malov/Denmark, with a size of 170 and 135 μm. The oocytes were categorized based on their state of development, with nuclear maturity evaluated by the presence of a first polar body outside the oocyte and the absence of a germinal vesicle.

ICSI procedure is performed on MII stage oocytes without any signs of degeneration. The sperm is observed and rendered immobile using a 10% solution of polyvinylpyrrolidone (PVP). The oocyte is held, the injection pipette RI/ ICSI-P us™ Spiked Injection Pipette is inserted into the zona pellucida and the outer surface of the oolemma. The oolemma is penetrated using ICSI micromanipulators. The cytoplasm is gradually reintroduced into the oocyte until the sperm enters the cytoplasm. The injected oocytes are then cultivated in an IVF medium (global® total® /Cooper surgical/US Single-step medium for uninterrupted embryo culture) and placed under mineral oil at 37 °C in a 5% CO2 incubator.

Evaluation of fertilization, cleavage, and embryo grading: Examination was performed approximately 16-18 hours after the ICSI to confirm fertilization by the existence of two pronuclei and two polar bodies.

The morphological characteristics of the embryos were used to determine their quality. (Nasiri and Eftekhari-Yazdi 2015[9]) According to the Istanbul consensus workshop published in 2011 by the Alpha Scientist in Reproductive Medicine and the ESHRE Special Interest Group of Embryology, embryos were ranked from I to III based on their scoring
Grade I: stage-specific cell size, no multinucleation, and less than 10% fragmentation.
Class II: There is 10-25% fragmentation, most cells have a size peculiar to their stage, and there is no sign of multinucleation.
Cell size is not stage-specific; there is evidence of multinucleation, and severe fragmentation (>25%) is grade III.
The formula was used to determine the percentage of distinct embryo quality grades (I, II, and III), According to (Nasiri and Eftekhari-Yazdi 2015[9]) the result is equal to the sum of all embryos divided

by the total number of embryos in each grade (GI, GII, GIII) and multiplied by 100.

Embryo transfer and Luteal support: The transfer occurs on day 2 (four-cell embryo), day 3 (six-cell embryo), or day 5 (blastocyst stage) following ICSI. A flexible catheter is inserted into the uterine cavity using trans-abdominal ultrasound. Then the catheter is examined under a microscope to confirm successful embryo transfer. On the day of oocytes pick up progesterone injection depote 250mg twice weekly and vaginal progesterone (Cyclogest®400mg; Actavis, UK), or (Crinone, ® 8% progesterone gel, MERK, Switzerland), and daily continued. A serum B-hCG assay is conducted fourteen days after embryo transfer. A pregnancy test is conducted fourteen days after embryo transfer.

2-2-2 Statical analysis

The data were analyzed using Statistical Package for Social Sciences (SPSS) version 23.0 and Microsoft Office 2010. The descriptive statistics, including frequency, range, mean, and standard error, were measured to describe the data. The groups were compared by applying an independent sample t-test (Unpaired t-test between two groups), and the degree of association between continuous variables was calculated by Pearson's correlation coefficient (r). The cut-off value, sensitivity, and specificity were calculated using the Receiver operative characteristics (ROC) curve, and the results were considered statistically significant when the p-value was equal to or less than 0.05.

1. Result

3.1.Baseline characteristics of patients who participated in the current study.

The current cross-sectional study included 90 female infertile subjects, and the results were presented as mean plus minus standard error of the mean (SEM). Table 1 revealed baseline demographic features. The age of the patients ranges from 20-44 with a mean of 31.16 ± 0.61, and BMI ranges from 19.50 - 32.00 with a mean of 26.59 ± 0.35; women and the duration of infertility in years range from 1-12, and the mean is 5.14 ± 0.39. primary infertility is 62(69%), secondary infertility is 28 (31%), women with BMI < 25 kg/m² Normal weight are 30 (33.4 %), and ≥ 25 kg/m² (Overweight & obese) are 60 (66.6 %).

Table (3-1) Baseline characteristics of patients who participated in the current study.

Parameters		Range	Mean ± SE
Age (years)		20 - 44	31.16 ± 0.61
BMI (Kg/m²)		19.50 – 32.00	26.59 ± 0.35
Duration of infertility (years)		1 - 12	5.14 ± 0.39
Parameters		N. (%)	
Type of infertility	Primary	62 (69.0 %)	
	Secondary	28 (31.0 %)	

BMI ranking	< 25 kg/m² (Normal weight)	30 (33.4 %)
	≥ 25 kg/m² (Overweight & above)	60 66.6 %)

SD: Standard error; BMI: Body mass index; n: Number of patients

3-2 Pregnancy rate

Thirty four out of 90 women became pregnant (Pregnancy rate=37.8 %). As shown in the pie chart figure (1).

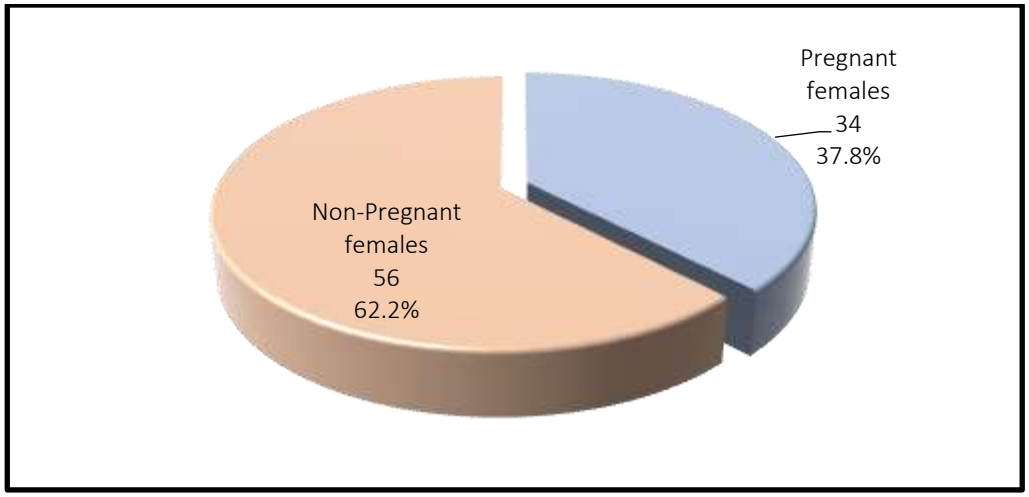

Figure (3-1) demonstrates the pregnancy rate.

3-3 Comparison of serum glutamine, FF glutamine and FFIL-6 between pregnant and non-pregnant females

The pregnant females had significantly higher levels of FF glutamine (4254 ± 125.2 vs. 2985 ± 79.15; p <0.001) in addition to FF IL-6 (151.34 ± 5.42 vs. 89.79 ± 3.83; p <0.001), on the other hand there was no significant difference of serum glutamine between pregnant and non-pregnant females (6087 ± 108.8 vs. 6042 ± 85.99; p=0.751) as shown in Table (3-2).

Table (3-2) Comparison of serum glutamine, FF glutamine and FF IL-6 levels between pregnant and non-pregnant females

Parameters	Pregnant females	Non-pregnant females	p value
Serum glutamine (ng/ml)	6087 ± 108.8	6042 ± 85.99	0.751 Ŧ NS
Follicular fluid glutamine (ng/ml)	4254 ± 125.2	2985 ± 79.15	< 0.001 Ŧ S
Follicular fluid IL-6 (pg/ml)	151.34 ± 5.42	89.79 ± 3.83	< 0.001 Ŧ S

NS: Not significant (p > 0.05); S: Significant (p ≤ 0.05); Ŧ: Independent sample t test

4 Correlations of serum glutamine, FF glutamine and FF IL-6 with patient's age and body mass indices

There was a significant negative correlation between patient's age with both FF glutamine (r=-0.212 & p=0.044) and FF IL-6 (r=-0.247 & p=0.019), as shown in Table 3-3.

Table (3-3) Correlations of serum glutamine, FF glutamine and FF IL-6 with patient's age and body mass index

Parameters	*Statistics*	*Serum glutamine*	*Follicular fluids glutamine*	*Follicular fluids IL-6*
Age	*r*	*- 0.090*	***- 0.212***	***- 0.247***
	p value	*0.401 NS*	***0.044 S***	***0.019 S***
BMI	*r*	*- 0.026*	*- 0.054*	*- 0.178*
	p value	*0.807 NS*	*0.610 NS*	*0.094 NS*

r: Pearson's correlation coefficient; NS: Not significant (p > 0.05); S: Significant (p ≤ 0.05)

3-4 Correlation between serum glutamine, FF glutamine and FF IL-6.

There was a positive significant correlation of FF glutamine with FF IL-6 (r=0.606 & p <0.001); however, there were no significant correlations between serum glutamine with FF glutamine (r=0.147 & p=0.165) and between serum glutamine with FF IL-6 (r=0.002 & p=0.986) as illustrated in Table (4).

Table (3-4) Correlation of serum glutamine, FF glutamine and FF IL-6

r:

ICSI parameters	*Statistics*	*Serum glutamine*	*Follicular fluids glutamine*	*Follicular fluids IL-6*
Serum glutamine	*r*	*1*	*0.147*	*0.002*
	p value		*0.165 NS*	*0.986 NS*
Follicular fluids glutamine	*r*	*0.147*	***1***	***0.606***
	p value	*0.165 NS*		***< 0.001 S***
Follicular fluids IL-6	*r*	*0.002*	***0.606***	*1*
	p value	*0.986 NS*	***< 0.001 S***	

Pearson's correlation coefficient; NS: Not significant (p > 0.05); S: Significant (p ≤ 0.05)

3-5 FF glutamine and IL-6 levels as a predictor of positive pregnancy.

Receiver Operative Characteristic curve (ROC curve) was applied to calculate both follicular fluids glutamine and IL-6 cut off values as a predictor of positive pregnancy with accepted sensitivity, specificity and area under the curve. According to the results, follicular fluids glutamine cut-off value was ≥ 3977 ng/ml with sensitivity= 70.7 %, specificity = 96.4 % and excellent area under the curve (AUC= 0.910), on the other side; follicular fluids IL-6 cut off value to predict positive pregnancy was ≥ 121.3 pg/ml with sensitivity =79.4%, specificity = 80.4% and also excellent area under curve (AUC=0.913) Table (3-5) Figure (3-2).

Table (3-5) ROC of follicular fluids glutamine and IL-6 levels to predict positive pregnancy

ROC curve characteristics	*Follicular fluids glutamine*	*Follicular fluids IL-6*
Cut off value	*≥ 3977 ng/ml*	*≥ 121.3 pg/ml*
Area under curve (AUC)	*0.910*	*0.913*
Sensitivity %	*70.6 %*	*79.4 %*
Specificity %	*96.4 %*	*80.4 %*
p value	*< 0.001 S*	*< 0.001 S*

S: Significant (p ≤ 0.05)

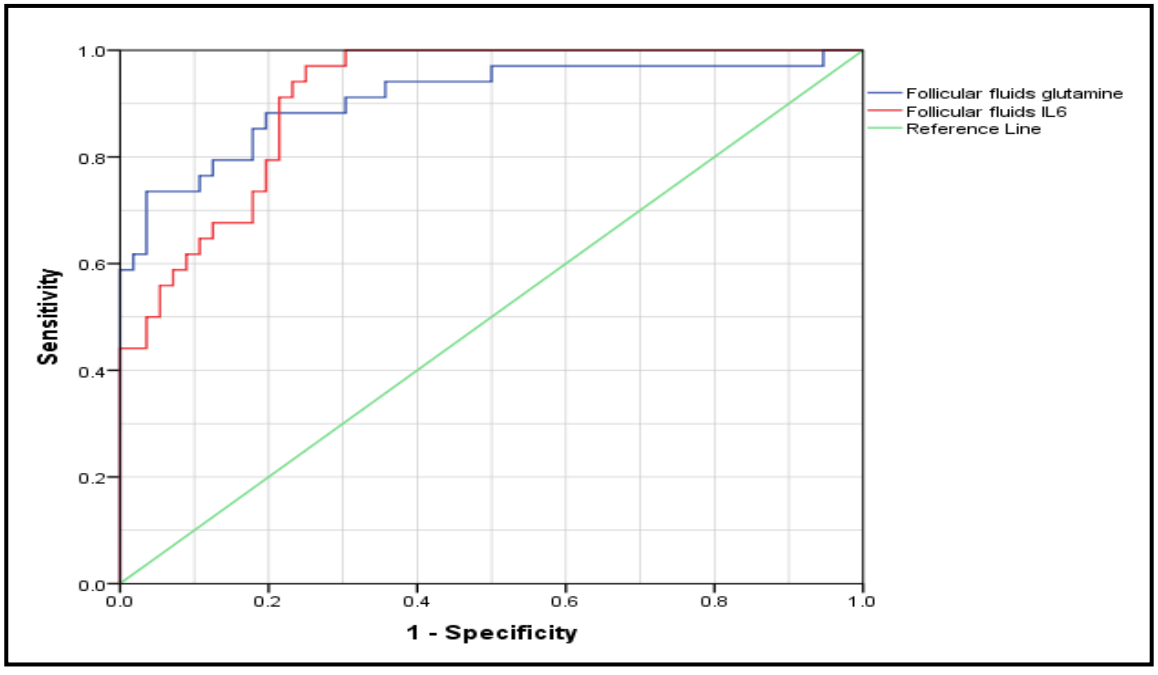

Figure (2) ROC curve of follicular fluids glutamine and IL-6 to predict positive pregnancy

4. Discussion

4-1 Comparison of serum and FFglutamine levels, and FF IL-6 between pregnant and non-pregnant women

The pregnant women had highly significant differences in levels of glutamine and IL-6 in follicular fluid than non-pregnant. Interestingly, in the current study, the pregnancy rate is 37.8 %, which is consistence with the success rate of ART applications, which remains at ~35% (pregnancy rate per embryo transfer based on the ESHRE annual report 2016[10]). Several previous studies found there was a positive correlation between the number of retrieved oocytes and good-quality embryos with pregnancy rate (A.Michèle Arnot, et al.,1995[11], Michael Fanton, 2023[12], Ming-I Hsu, 2016[13]). It is important to highlight the fact that the likelihood of pregnancy is significantly influenced by the number of embryos transferred, which is determined by transfer efficiency, embryo quality, and endometrium responsiveness. (Coughlan *et al.*, 2014[14]) In a study performed by Winkle, Lon J. 2021 to understand the role of glutamine on implantation, glutamine was believed to serve as an osmolyte within the cells of early embryos when exposed to high osmotic circumstances. Cleavage-stage embryos were expected to develop in the highly concentrated oviductal fluid. The osmolarities of oviductal and uterine secretions remain consistently hyperosmotic throughout the various stages of preimplantation development, but they do change by glutamine transport into the embryos as osmolytes (Winkle, Lon. J. et al., 2021[15]).

Notably, IL-6 levels exhibit a fluctuating pattern throughout the menstrual cycle. IL-6 is expressed at a modest level during the proliferative phase. However, its expression level gradually rises after ovulation and reaches its peak during the mid-to-late-secretory period. The cyclical synchronization of this expression pattern matches with the window of implantation, suggesting that IL-6 may have a role in the receptivity of the endometrium(Ochoa-Bernal et l., 2020[16], Vilotić Aleksandra, et al., 2022[17]). It is generally accepted that IL-6, a proinflammatory cytokine, is primarily secreted by endometrial epithelium and stromal cells during the process of implantation. Moreover, this cytokine has dual functions in embryo implantation and placental development and is essential for pregnancy maintenance (Ochoa-Bernal. Et al., 2020[16]) This may be because IL-6 contributes to angiogenesis and vasculogenesis in the endometrium. Furthermore, it enhances the placenta's synthesis of hormones and acts as an essential controller of the placenta's inflammatory reaction and immune equilibrium (Matić, M. et al., 2022[18]). Another promising finding was a study by Yang, J.,2022, who stated that "High levels of IL-6 in the follicular fluid can increase the clinical pregnancy rate and reduce embryo fragmentation."(Yang, J.et al., 2022[19]). This agrees with the present study. Also, this result ties well with a previous study by Bedaiwy M et al. 2007 which stated that FF IL6 is higher in pregnant women than non pregnant. On this basis, the present study concluded that FF IL-6 and glutamine can be considered predictors of pregnancy.(Bedaiwy M. et al. 2007[20])

However, overall, these findings disagreed with the findings reported by Altun, T., 2011, which may be due to the small sample size and use of agonist and antagonist protocol.(Altun T.et al., 2011[21])

4-2 Correlation of serum and follicular fluid glutamine levels and follicular fluid IL-6 levels with patient`s age.

women age is the single most important predictor of pregnancy in women undergoing assisted reproduction (Al-Obaidi et al., 2018[22]). Based on the human circadian rhythm, female reproductive capacity experiences a notable fall in the early 30s, followed by a sharp decrease after age 35. In other words, individuals' ovarian reserves decrease with age (Moridi A et al., 2019[23]).

In the current study, There was a significant negative correlation between the patient's age and FF glutamine as showen in Table (3-3).

The mean age of women involved in this study was 31.16 ± 0.61. As women get older the follicular fluid glutamine decreases, while serum glutamine levels show no significant correlation with age.

Previous studies showen that the impact of maternal aging on follicular amino acid profiles and their

metabolism remain largely unknown, with only a few published reports available (Babayev and Duncan, 2022[24]; Krisher, 2019b[25]).

The result of this study agreed with a study done by Smits et al., 2023 who found that human ovarian aging which was caused by oxidative stress and dysfunctional mitochondria in granulosa cells, can impact the quality and functionality of oocytes and disturbance of amino acid metabolism, mainly glutamine and glutamate that in turn leads to disturbed glutathione levels (Smits et al., 2023[26]). Since FF glutamine level decreased in association with impaired mitochondrial tricarboxylic acid cycle efficiency and reduced glutathione synthesis, promoting mitochondrial dysfunction and oxidative stress in oocytes from advanced maternal age women (Smits et al., 2023[26])

Regarding IL-6, There was a significant negative correlation between patient age and FF IL-6 as illustrated in Tables (3-3). This result agrees with the result obtained by Stojanovic Gavrilovic et al.in 2022[27], who found the IL-6 level was higher in the young age group, and that group has had positive IVF outcomes. Also, two other studies supported this study (Yang J.et al 2020[19]) (Tugba Altun. et al., 2011[21]). This may be explained by Sadraie, S. H., 2000 who demonstrated that advanced maternal age which was associated with a reduction of oocyte number and more apoptosis of granulosa cells resulting in a drop in the secretion of IL-6 by these cells (Sadraie, S. H. et al.,2000[28]).

4-3 The Correlation of serum and FF glutamine levels and FF IL-6 levels.

There was a statistically significant strong positive correlation between FF glutamine and FF IL-6, whereas there were no significant correlations between serum and FF glutamine levels and between serum glutamine and follicular fluid IL-6, as demonstrated in Table (3-4).

As already mentioned above, Glutamine is an important energy source for ovarian macrophages and lymphocytes; it provides essential nitrogen and carbon for producing multiple bioactive materials. Optimal quantities of glutamine enhance the synthesis of cytokines, such as IL-6 (Ma, G. et al., 2022[3]) (Sartori, Talita. Et al., 2017[29]) these findings seem to be supported by Rogeri P. S et al. 2020[7], who found an important relationship between IL-6 and glutamine; any dysregulation of glutamine metabolism or IL-6 signaling can participate in polycystic ovarian disease, inflammation in the ovary and even ovarian cancer. these important findings may be supported by the findings of Sartori, Talita; 2017 who explained the immunological roles of glutamine because that study confirmed that the glutamine possesses immunomodulatory properties, and the reduced availability of glutamine to immunological-competent cells can impact their functionality and IL-6 production. (Sartori, Talita. et al.,2017[29]). So based on the above findings , the current study can conclude that the follicular fluid glutamine level can affect IL-6 level and function and, as a consequence, ovarian function.

4-4 FF glutamine and IL-6 levels as a predictor of positive pregnancy.

According to the results, the follicular fluid glutamine cut-off value was ≥ 3977 ng/ml with sensitivity = 70.7 %, specificity = 96.4 %, and excellent area under the curve (AUC=0.910). On the other hand, the follicular fluid IL-6 cut-off value to predict positive pregnancy was ≥ 121.3 pg/ml with sensitivity =79.4%, specificity = 80.4%, and also excellent area under curve (AUC=0.913). No previous study about the cut- off value of follicular fluid glutamine and ICSI outcome existed, the current study showed that serum glutamine levels cannot predict pregnancy after ICSI. However, follicular fluid glutamine and IL6 levels may be beneficial as a screening test for predicting pregnancy following ICSI.

while a study about IL6 was performed by Stojanovic Gavrilovic et al. 2022, It has been determined that when the IL-6 concentrations fall between 3.67 ng/mL and 10 ng/mL, this can anticipate a favorable outcome in IVF with a viable pregnancy, considering the differences in measuring units present study used (pg/ml) (Stojanovic Gavrilovic et al. 2022 [27]).

5. Conclusions

Infertility is a serious issue in the modern world. Even with advances in medicine, it is still difficult to

completely comprehend the intricate connections between the various cells involved in the processes of fertilization and implantation. The analysis of follicular fluid composition is becoming more and more used as a means of determining the underlying cause of infertility. The aim of this study was to examine the effects of glutamine and IL-6 on pregnancy rate, in women undergoing ICSI. The present study concluded that there was a highly significant correlation between FF glutamine and FF IL-6, and both FF glutamine and FF IL-6 can be considered predictors for pregnancy.

Funding

This work received no funding.

Conflict of Interest

The author declares no conflict of interest.

Ethical Clearance

The study was approved by the local Ethical Approval Committee.

Reference

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