Assessment of ITGB3 gene expression and hormonal status in infertile female undergoing an in vitro fertilization protocol

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Abstract
Infertility is defined as the failure to achieve a clinical pregnancy after 12 months of regular unprotected sexual intercourse. Causes of Female Infertility, according to the Center of Disease Control (CDC, 2013), can be divided into three broad categories including defective ovulation, transport and implantation. Embryo implantation is a complex process involving multiple biological factors,one of these factors is a genetic factor. One of infertility solution is invitro fertilization program. Integrins are a family of transmembrane heterodimeric glycoproteins, their major roles are focused on differentiation, apoptosis, motility and facilitate cell-extracellular matrix adhesion. Integrin and its ligands are not only involved in physiological processes and pathological processes but also play an essential role in human implantation. According to the previous, this study aimed to study the role of ITGB3 expression in implantation of embryo. In the present study, whole blood was isolated from 120 women and distributed into three groups as follows: Group 1 included: 29 Infertile women who underwent successful IVF surgery; Group 2: 42 Infertile female who underwent failed IVF surgery; Group 3: 50 fertile women. The messenger RNA (mRNA) expression levels of ITGB3 in the peripheral blood were analyzed using reverse transcription-polymerase chain reaction (RT-PCR). The expression of ITGB3 mRNA in Groups 1, 2and 3 were 0.0013, 0.012, and 1.0 folds of the gene expression, respectively, Using GAPDH as Housekeeping Gene. In conclusion, according to our results, β3 integrin expression reduces in women with unexplained infertility. The low expression of β3 integrin in female with unexplained infertility supports the possible role of these molecules in the endometrial receptivity during implantation. Therefore, these genes could account as the potential molecular markers of infertility.
Keywords: infertility, ITGB3 Gene, implantation, gene expression, RT-PCR

1. Introduction

Infertility is a reproductive health problem around the world, The prevalence rate for females increased by 0.37% annually, and the global disease burden of infertility increased from 1990 to 2017 [1]. With environment and lifestyle changes, the incidence of infertility might be associated with the delay of marriage and giving birth to the first child [2], environmental pollution [3], and unhealthy lifestyles [4]. Although not life-threatening, infertility has a negative impact on patients, their families, and society as a whole. Patients with an infertility diagnosis have increased psychological stress and deteriorating family dynamics [5], which may significantly lower their quality of life. Additionally, the aging issue may become worse due to the dropping birth rate.
Infertility is defined as the failure to achieve a clinical pregnancy after 12 months of regular unprotected sexual intercourse [6]. The absence of identified reasons of infertility is referred to as unexplained infertility (UEI) [6]. Male factor, ovulatory dysfunction, or tubal-peritoneal disease are the three main reasons of infertility in the majority of couples [7]. According to the Center of Disease Control (CDC, 2013) [8], the causes of female infertility can be categorized into three general groups, including defective ovulation, transport and implantation [9].
The phrase "implantation failure" really refers to a number of circumstances in which, the embryo does not implant in the maternal endometrium after both spontaneous and in vitro fertilization (IVF) [10]. Recurrent implantation failure (RIF) is an undefined, quite often, clinical condition that can result from the repeated failure of embryo transfers to obtain implantation in Iraqi females under IVF program [11].

Mediators of Implantation

In both humans and primates, In order to start a pregnancy, a series of carefully orchestrated events known as implantation must take place, For endometrial receptivity, a variety of mediators that ovarian hormones regulate are crucial. One of these mediators, along with cytokines, growth factors, and others, is cellular adhesion molecules (CAM) [12]. Cellular Adhesion Molecules are a family that includes members such as integrins, cadherins, and selectins.
In this study, we will address the ITGB3 gene and reveal whether there is a relationship between

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integrin expression, hormonal status and pregnancy in women.

Integrins

Integrins are a family of transmembrane heterodimeric glycoproteins that facilitate cell-extracellular matrix adhesion [13]. The integrin family includes 18 alpha (α) and 8 beta (β) subunits that form 24 distinct αβ heterodimers. Each integrin heterodimer consists of a large extracellular domain region, two single-pass transmembrane helices (one in each subunit), and short cytoplasmic tails [14]. They experience dynamic spatial and temporal changes in the endometrium during the menstrual cycle [15].

They are also expressed in the endometrium during the time of implantation [16]. Their major roles are focused on differentiation, apoptosis, motility and attachment [17]. During implantation, integrins play a role during the attachment of the cells to the ECM and initiating a signaling transduction from the embryo to the ECM to initiate the translation of genes involved in the implantation process [18]. There are different isoforms in mammals, but the α1β1, α4β1 and αVβ3 are the most relevant during implantation. In humans, these three isoforms are expressed in the endometrium during the window of implantation, when the endometrium is structurally and physiologically responsive to blastocyst implantation [19]. integrin activates FAK, which in turn can switch on the VAV-RAC1 signaling axis to bring the endometrial epithelial cell receptivity for blastocyst attachment. Furthermore, integrin directly interact with the implanting-blastocyst[20]. Reduced in endometrial expression of integrin may contribute to unexplained infertility and this genes could account as the potential molecular markers of infertility [21].in this study, we selected ITGB3 to investigate its expression role in embryonic implantation.

Aliases for ITGB3 Gene

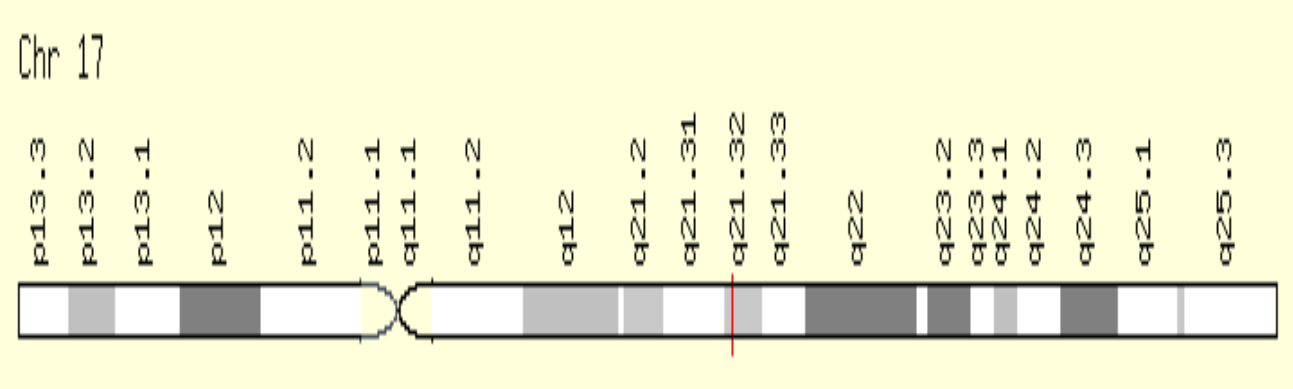
GeneCards Symbol: *ITGB3* (Integrin Subunit Beta 3)

GPIIIa, CD61, GP3A, Integrin, Beta 3 (Platelet Glycoprotein IIIa, Antigen CD61), Platelet Membrane Glycoprotein IIIa, Integrin Beta-3, Antigen CD61, Integrin Beta Chain, Beta 3, Platelet Glycoprotein IIIa Integrin Beta 3, CD61 Antigen, BDPLT16, BDPLT24, BDPLT2, GT2, GT.

genome assembly for ITGB3 Gene (GRCh38/hg38)

Genomic localization for ITGB3 Gene

Chromosome 17 Cytogenetic band: 17q21.32



Start 47.253.827 bp End 47.313.743bp

Size: 59,917 bases Orientation: Plus strand Exon count: 15 [22]

2. Materials and Methods

This study is carried out between March 2021 to April 2022 to determine the various hormones, some biomarkers and ITGB3 gene expression in females with infertility attending General Hospitals in Baghdad, Iraq and to correlate them with infertility conditions.

Subjects

All females included in this study were within the reproductive age range (20- 43 years) without any endocrine diseases were included; while those with endocrine diseases or polycystic ovary syndrome or male infertility were excluded; 71 infertile females under IVF protocol (divided into two subgroups , 29 success implantation and 42 failure implantation after follow up all these cases to determine the results of implantation after blastocytes implantation process within IVF program)and 50 fertile females as control were involved in the study, each fertile female have, at least , one previous birth .

Blood sampling

Venous blood samples (6ml) were collected from each female. Each blood sample was distributed into three aliquots; the first in Eppendorf tubes with TRIzol for gene expression study, while the second aliquot was drawn in EDTA tube and stored at -18°C until DNA extraction for ITGB3 gene variations determination. the third, put in Gel tubes, for hormonal profile study. then the tubes were centrifuged at 4000rpm for five minutes the resulted serum was collected and kept in -18 °C until used.

Anthropometric measurements

These measurements included: age,age of menarch by years , Weight (kg), height (m), and body mass index (BMI) was calculated by dividing the body weight by the square of the height according to the following equation: BMI = Weight (Kg) / [height

(cm)]2. information has been set for each female in a special questionnaire list.

Hormonal profile

The hormones Estradiol (E2), Prolactine (PRL) , Progesterone (PRG) , Luteinizing Hormone (LH) and human chorionic gonadotropin (HCG) were measured by Enzyme-Linked fluorescent Assay (ELFA)by using mini-VIDAS (mini VIDAS® by bioMérieux).

Gene expression

Total RNA of each sample were extracted using the Trans Zol Up Plus RNA Kit (TransGen, biotech. ER501) following the protocol provided by the manufacturer.

Total RNA was reversely transcribed to complementary DNA (cDNA) using EasyScript® One-Step gDNA Removal and cDNA Synthesis SuperMix Kit (TransGen, biotech. AE311). The procedure was carried out in a reaction volume of 20 µl according to the manufacturer's instructions.

To confirm the expression of target genes, TransStart®TopGreen qPCR SuperMix (TransGen,biotech.AQ131 01) was used. The PCR program started with an initial melting cycle for 30 sec at 94C°to activate the polymerase, followed by 40 cycles of melting (5 sec at 94C°), annealing (30 sec at 60C°for GABDH and ITGB3) and extension (22 sec at 72C°). The quality of the PCR reactions was confirmed by melting curve analyses. All the gene expression levels were normalized to that of the housekeeping gene glyceraldehyde-3-phosphate dehydrogenase (GAPDH).

Primers for ITGB3 were forward, 5′-CAGGGCAGGGAACAACTT -3′, and reverse, 5′-GGATTGGTCCTTATACTCAAAA -3′ [23]. Primers for GAPDH were forward, 5′-GAAATCCCATCACCATCTTCCAGG -3′, and reverse, 5′-GAGCCCCAGCCTTCTCCATG -3′.

3. Statistical Analysis

Real-time PCR depend on relative gene expression levels normalized by GAPDH, which were calculated by the formula $2-\Delta Ct$, where ΔCt (critical threshold) = Ct of genes of interest - Ct of GAPDH. fold changes of gene expression levels in patients and control were calculated by the $2-\Delta\Delta Ct$ method as described in [24], where ΔΔ CT=ΔCT (test)-ΔCT (calibrator) . ΔCT (calibrator) = CT gene of interest (target, calibrator) – CT internal control. The calibrator was chosen from control samples. A 2-fold change threshold was set for identifying significant changes in gene expression. Comparison done by using t-test and ANOVA to evaluate group-level differences. We considered $P \leq 0.05$ to be significant (*) and $P < 0.01$ (**) to be high significant.

4. Results and Discussion

This study examined the hormonal profile, some stress biomarkers, and ITGA2, ITGB3 expression in infertile females who are under IVF protocol divided between the success of implantation and failure it.

The demographic and gynecological characteristics are presented in Table 1.

Table 1: Comparison between different groups in Age, BMI and Age at menarche

Study groups		Mean ± SE		
		Age (year)	BMI (kg/m^2)	Age at menarche (year)
Infertile	Success	30.57 ±0.77 a	28.45 ±0.78	12.63 ±0.19 ab
	Failure	30.30 ±0.92 a	26.98 ±0.59	13.17 ±0.21 a
Fertile		25.56 ±0.89 b	27.68 ±0.54	12.20 ±0.16 b
LSD value		2.581 **	1.790 NS	0.546 **
P-value		0.0001	0.306	0.0011

Means having the different letters in the same column differed significantly. ** (P≤0.01).

The infertile female in the success group had a mean age of 30.30 ±0.92 years, female in the failure group had a mean age of 30.30±0.92 years, while the fertile female had a mean age of 25.56 ±0.89 years. These results, show that age was significantly higher ($p<0.01$) in infertile female in the success and failure groups (30.57, 30.30 years) than in the fertile female in the control group (25.56 years). A previous study showed similar results to what was obtained in the current study[25]. On the other hand, other Iraqi studies, showed that there are no significant differences in age between infertile and fertile female [26;27].

In previous studies, the mean age of menarche (AOM) amounted to 12.86 years [28, 29], Thus the age at menarche in the present sample lies within the normal age range (12 -14 years). For 8 females (11.4%) the first menstrual bleeding occurred relatively early i.e., before the 12th year, 55 females (78.6%) experienced the menarche between the 12th and 14th year, with 19 females (10%) the menarche occurred after the age of 14.

The table above shows that there are highly significant differences ($p<0.01$) between the failure and control groups (13.17 years versus 12.20 years). So maybe there is a correlation between the age of menarche and infertility.

Menarche that happens too early, too late, or not at all is particularly concerning because these situations can have negative effects in the future. Menarche is regarded as early if it happens before or at the age of 10, and late if it happens at or beyond the age of fifteen [30]. An earlier menarche history has been associated with a higher risk of preterm birth, according to a prior study [31]. Another study looks into how nulliparous ladies' menarche ages correlate

with their obstetric outcomes [32].The onset of menarche on fecundity is not well known since the few existing results are conflicting.

The infertile women in the success and in the failure groups had a mean BMI (of 28.45, and 26.98) kg/m2 respectively, while the fertile women in the control group had a mean BMI of 27.68 kg/m2. Thus, there are no significant differences between the groups. These results were consistent with what Simon and Rana discovered in their studies [33,34].

Table 2 shows a comparison of the groups in terms of the duration of the menstrual cycle and Duration of menstrual flow.

Table 2: Comparison between different groups in Duration of menstrual cycle and Duration of menstrual flow

Study groups		Mean ± SE	
		Duration of the menstrual cycle (days)	Duration of the menstrual flow (days)
Infertile	Success	32.96 ± 0.96 a	5.03 ± 0.29
	Failure	30.10 ± 0.81 b	5.10 ± 0.19
Fertile		29.52 ± 0.13 b	5.14 ± 0. 16
LSD		1.812 **	0.595 NS
P-value		0.0010	0.940
Mean having the different letter in the same column differed significantly. ** (P ≤ 0.01).			

However, the duration of menstrual cycles was significantly higher ($p<0.01$) in infertile female ,however the success group has longer cycle than the infertile females in the failure group , at the same time , both infertile groups recorded longer cycle days than the fertile females group (32.96 days versus 30.10 and 29.52 days respectively).

While the mean duration of menstrual flow of the infertile females was not statistically different from those of the fertile females ($p>0.05$).

Table 3: Comparison between Successful and Failure groups in hormone level .

Groups		Mean ± SE (ng/ml)				
		E2	PRL	PRG	LH	HCG
infertile	success	1212.78 ±155.33	27.18 ±1.27	49.57 ±1.73	1.012 ±0.12	23.86 ±1.32
	failure	1087.03 ±97.48	28.61 ±1.34	47.73 ±1.72	1.114 ±0.10	19.21 ±1.30
References		93-575	5-35	0.4 – 2.50	9.6-80.0	0.5 - 2.24
t-test		349.78 NS	3.790 NS	4.977 NS	0.330 NS	3.769 *
P-value		0.475	0.455	0.463	0.540	0.0164
Significant * (P≤0.05), NS: Non-Significant.						

Table 3 illustrates that the concentration of E2 is high in infertile female which compared to what Gruber approved [35], Looking at these results, we find that there are no significant differences between the success and faiure groups, and this contradicts what Zainab found in her study [27].

Previously, Xiaoyuan studied the relationship of a high level of estradiol in pregnancy and proved that there was a positive association between serum E2 and live birth rates [36].

The mean HCG concentration of the Success group (23.86 ng/ml) was statistically significant ($p<0.05$) when compared with the failure group (19.21 ng/ml). HCG effectively modulates several metabolic pathways within the decidua contributing to endometrial receptivity [37].

Human chorionic gonadotropin (hCG) is a multi-effect hormone which has an incredible impact on humans and more especially on the acceptation and the success of the gestation. The cytokine–hCG interaction is well known and cytokines play a key role in the female immune response during conception, implantation, maintenance of pregnancy, embryo development, etc. HCG acts through different pathways and on multiple cell types. It promotes the acceptation of the embryo implantation, angiogenesis and vasculogenesis, and the control of the trophoblast differentiation, as well as the immune regulation of the maternal embryonic or fetal interface during the entire pregnancy [38].

Therefore, the effects of HCG on human endometrium constitute the theoretical basis to develop clinical research protocols aiming to investigate HCG efficacy in improving clinical parameters of assisted reproduction protocols. In that view, since 2011, two reports have been published upon this issue with conflicting results [39].

On the other hand, the mean of E2, PRL, PRG, and LH of the success group (1212.78, 27.18, 49.57, and 1.012 ng/ml) show no significant difference ($p>0.05$) when compared with the failure Group (1087.03, 28.61, 47.73, 1.114 ng/ml).

Our results showed that serum prolactin had no significant impact on implantation, as did the results from previous work by Zhong et al. [40]. This was contradictory to the work presented by Molitch [41].

As well as the results showed that progesterone level has no significant impact on pregnancy, this disagreed with another study has proven that the serum progesterone is significantly associated with a number of oocytes retrieved which in turn is associated with successful IVF outcome and therefore seems to be an indirect parameter for predicting successful IVF outcome. This is in agreement with another large retrospective cohort

study [42].
The present data showed that LH and E2 had no significant effect on implantation rate, A previous study showed that the level of LH can affect the IVF/ICSI outcome in the follicular-phase long protocol [43]. E2 level can also predict the outcomes of IVF [44]

Table 4: Estimate of the correlation coefficient between variables in infertile groups

Parameters	Age	BMI	Age at menarche	Trial period	E2	PRL	PRG	LH	HCG
Age	-	0.21	0.24	0.01	-0.01	0.02	-0.06	-0.23	0.08
BMI	0.06	-	-0.11	0.20	0.23	0.07	0.02	-0.14	0.16
Ag at menarche	0.10	-0.02	-	0.26	-0.19	-0.14	-0.28	0.09	-0.08
Trial period	0.21	0.24	-0.23	-	-0.15	0.11	-0.06	0.11	0.10
E2	-0.15	-0.04	0.14	0.01	-	-0.01	0.21	-0.16	0.01
PRL	-0.02	-0.06	-0.10	0.15	-0.02	-	0.16	0.09	0.09
PRG	-0.07	-0.14	-0.18	-0.13	0.16	-0.31	-	0.02	-0.26
LH	0.15	0.02	-0.02	0.06	-0.10	0.19	-0.20	-	-0.03
HCG	0.41 *	0.08	-0.04	0.22	-0.17	0.34	-0.35*	0.20	-

* (P≤0.05), ** (P≤0.01), NS: Non-Significant.
Under diagonal: Successful group , Above diagonal : Failure group

From the above table, we notice that there is a significant relationship between age and the level of HCG in women in the Success group, as the concentration of HCG increases with increasing age. In a previous study [45], it is consistent with our findings, whereby the concentrations of HCG in the blood increased with age in women. HCG results were higher and significantly different (P < 0.0001) for nonpregnant women >55 years compared with women 18-40 years and 41-55 years, and HCG in women 41-55 years, was significantly higher than in women 18-40 years.
premenopausal women's HCG and LH levels rise during ovulation.as women get older, HCG levels, like those of FSH and LH, arise due to the loss of negative feedback inhibition from estrogen and progesterone [46].
In addition to this relationship, current study illustrated, in the success group, that there is a significant relationship between progesterone and HCG, and this relationship is inverse, meaning that high progesterone leads to a decrease in HCG levels. In fact, there is no study that discussed this relationship, but indirectly, during the controlled ovarian hyperstimulation (COS) cycles, elevated serum progesterone level is observed in some cases, particularly at the end of the stimulation cycle [47]. The incidence of elevated P levels was reported as 12.8%–38% in GnRH antagonist cycles [48]. Most studies believe that clinical pregnancy rate CPR decreases dramatically once the progesterone level is higher than 1.4 ng/ml [49].
On the other hand, there is no significant relationship between the biomarkers in the failure group.

Results of gene expression

In present study, the Ct values that were obtained by RT-qPCR indicate the number of cycles that are required for the signal to cross the detection threshold, exceeding the indicated background. thus, the lower Ct value, the more abundant the gene target is in a given sample. The variation in Ct values of all the samples indicated that the expression levels of the tested gene differed among the different samples.
GAPDH expression quantification by Real-time PCR
Table 5 displays the Ct value of GAPDH (glyceraldehyde-3-phosphate dehydrogenase), the internal reference gene (RG) (housekeeping gene) used for this work. The Ct values for GAPDH among all study groups ranged from 19 -20, with a means of Ct values (20.43), (20.45), and (20.45), in the success, failure, and control groups. There was no significant difference between these groups in terms of the Ct value means of GAPDH (p = 0.895), as appeared in Table 5.

Table 5: Threshold value of GAPDH in the different studied group

Study groups		Ct value of GAPDH Mean ± SE	Range
infertile	success	20.43 ± 0.12	19-21
	failure	20.45 ± 0.11	19-21
Fertile		20.45 ± 0.10	19-21
LSD		1.036 NS	---
p- value		0.895	---

NS: Non-Significant.

The ideal RG should be stably expressed across different tissues and experimental treatments [50].
The fact that the Ct value of GAPDH gene varies slightly in different sample types indicates that this gene is suitable as standard RG for that GAPDH used as reference gene for expression in present study.

Quantification of ITGB3 expression by Real-time PCR.

Infertile Patients were categorized into two subgroups: success and failure. Table 6 shows the

mean of Ct values of ITGB3 cDNA amplification in success , and failure (28.77, and 25.59) respectively, compared to the fertile group's corresponding Ct values (19.27). The mean CT results differed significantly across research groups. The mean Ct values in the success group were greater than those in the failure group, which were higher than those in the fertile control group. This outcome reflects its lower expression in the study groups (success and failure) compared to the control group. These findings demonstrate that ITGB3 gene expression is reduced in all infertile, implying that the ITGB3 gene could be used as a biomarker for early diagnosis of infertility.

Table 6: Comparison of ITGB3gene in Ct, Δ Ct, and $2^{-\Delta Ct}$ values between the studied group.

Study group		Mean ± SE			
		ITGB3 Ct value	GAPDH Ct value	ITGB3 Δ Ct value	ITGB3 2^{-} Δ Ct values
infertile	success	28.77 ±0.25	20.43 ± 0.12	8.34± 0.29	0.0030 ±0.001 b
	failure	25.59 ±0.57 0.57	20.45 ± 0.11	5.14 ± 0.61	0.028 ±0.004 b
Fertile		19.27	20.45 ± 0.10	-1.18± 0.37	2.26 ±0.33 a
P value		0.0001 **	0.895 NS	0.0001 **	0.0001 **

Significant ** (P≤0.01), NS: Non-Significant.

The relative quantification equation was used to determine gene expression fold change [24]. This is based on the normalizing of Ct values of the ITGB3 cDNA to the GAPDH (which is the ΔCt). The mean of ΔCt (normalization Ct values) for every group of studies is obvious in Table 6. The ΔCt means were (8.34, 5.15 and -1.18) in the success, failure, and control groups, respectively. The study groups differed significantly (p= 0.0001). The 2-ΔΔCt values have been used by each research group to determine the ITGB3 gene's expression. Table 7 displays the outcomes. Every group's 2-ΔΔ Ct results have been compared to the control group. The 2-ΔΔ Ct mean values were 0.450, and 4.14 respectively, in the success and failure groups, while the control group's 2-ΔΔ Ct mean value was (329.69), depending on 2-ΔΔ Ct method, Fold of ITGB3 expression was calculated, and the results were as follows (0.0013 for success, 0.012 for failure and 1 for control). these findings imply that the fold of expression differed significantly among these groups, and the control group (p=0.0395).

Table 7: Fold of ITGB3 expression depending on 2^{-} Δ Δ Ct method

Study group		Mean of ITGB3 Ct	Mean of GAPDH Ct	Mean of Δ Ct	Mean of ΔCt calibrator	Δ Δ Ct	2^{-} Δ Δ Ct	Experimental group/control group	Fold of gene expression
infertile	success	28.77	20.43	8.34	7.19	1.15	0.450	0.45/329.69	0.0013 ±0.0005 b
	failure	25.59	20.45	5.14	7.19	-2.05	4.14	4.14/329.69	0.012 ±0.003 b
Fertile		19.27	20.45	-1.18	7.19	-8.36	329.69	329.69/329.69	1 ±0.00 a
LSD		-	-	-	-	-	-	-	0.537 *
P- value		-	-	-	-	-	-	-	0.0395*

Significant * (P≤0.05), NS: Non-Significant.

Integrins are a family of transmembrane heterodimeric glycoproteins that mediate cellular differentiation and adhesion, which is a crucial step in embryonic implantation [51].

Studies have been providing variable outcomes regarding αvβ3 integrin expression and its relation to failed implantation with some trials concluding that the significantly decreased expression of endometrial αvβ3 integrin in unexplained infertility could be a reflection of its crucial role in enhancing endometrial receptivity at the implantation window [52].

Endometrial preparation is a critical step in both the natural and artificial frozen embryo transfer (FET) cycle because the development of the endometrium must be synchronized with the embryo transfer for successful implantation [53]. Undoubtedly, estradiol (E2) coordinately interacts with progesterone and plays an important role in endometrial development [54]. Moreover, excess E2 levels have been found to increase the incidence of abnormal pregnancy conditions, such as intrauterine growth restriction and abnormal implantation of the placenta [55].

Although estrogen levels in normal natural cycles reach 300–400 pg/ml before ovulation, a study on donor cycles revealed that the E2 requirement for embryo implantation is low (< 100 pg/ml) [56].Fritz with his colleagues demonstrated that elevated E2 levels in artificial frozen embryo transfer (FET) cycles were related to a low ongoing pregnancy, live birth rate [57].

5. Conclusion

ITGB3 is significantly lower in cases of infertility, which may suggest that underexpression of ITGB3 gene linked to defective uterine receptivity, and play a role as an unrecognized cause of infertility in this population of women. We need larger studies of adequate statistical power, ideally investigating more than one menstrual cycle in the same woman, to investigate the usefulness of using these molecular molecules in clinical practice. This is to overcome the cyclic changes between different menstrual cycles.

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