

Serum Progesterone on Trigger Day and Its Association with IVF Outcomes: A Prospective Study

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ABSTRACT

Background: Premature progesterone elevation (PE) on the day of human chorionic gonadotropin (hCG) trigger during controlled ovarian stimulation has been reported in IVF/ICSI cycles despite the use of GnRH agonist or antagonist protocols. The impact of elevated progesterone levels on reproductive outcomes remains controversial, particularly regarding implantation and clinical pregnancy rates. Identifying clinically relevant progesterone thresholds may improve treatment strategies and optimize IVF/ICSI outcomes.

Objective: To evaluate the relationship between serum progesterone levels on the day of hCG trigger and reproductive outcomes in IVF/ICSI cycles and to determine the progesterone threshold associated with adverse clinical outcomes.

Methods: This prospective cohort study included 95 infertile women aged 20–35 years undergoing IVF/ICSI treatment at Ain Shams University Hospital between June 2023 and October 2024. Participants were categorized according to trigger-day serum progesterone levels into three groups: <1.3 ng/mL, 1.3–1.5 ng/mL, and >1.5 ng/mL. Demographic characteristics, hormonal profiles, ovarian response parameters, fertilization rate, implantation rate, and clinical pregnancy outcomes were compared. Receiver operating characteristic (ROC) analysis was performed to evaluate the predictive value of serum progesterone for pregnancy outcomes.

Results: Among the participants, 83.2% had progesterone levels <1.3 ng/mL, 4.2% had levels between 1.3 and 1.5 ng/mL, and 12.6% had levels >1.5 ng/mL. Clinical pregnancy rates were 43.0%, 50.0%, and 25.0%, respectively. Although differences in implantation and pregnancy rates did not reach statistical significance, outcomes were consistently lower in women with progesterone levels >1.5 ng/mL. No significant differences were observed in oocyte yield, embryo quality, fertilization rate, or baseline hormonal parameters. ROC analysis identified a progesterone cutoff value >0.7 ng/mL for predicting pregnancy outcomes (AUC = 0.622, sensitivity = 66.7%, specificity = 64.3%).

Conclusions: Elevated serum progesterone on the day of hCG trigger, particularly levels >1.5 ng/mL, is associated with reduced implantation and clinical pregnancy outcomes in IVF/ICSI cycles. Careful monitoring of progesterone levels and individualized treatment approaches, including consideration of freeze-all strategies, may improve reproductive success.

Keywords

IVF, ICSI, Progesterone Elevation, HCG Trigger, Clinical Pregnancy Rate, Implantation Rate, Assisted Reproductive Technology, Controlled Ovarian Stimulation.

Introduction

Despite routine suppression of endogenous gonadotropins by gonadotropin-releasing hormone (GnRH) agonists, serum progesterone elevation (PE) on the day of human chorionic gonadotropin (hCG) trigger has been reported in controlled ovarian stimulation (COS) cycles in not only short protocols but also long protocols [1].

The occurrence rates of PE have been reported to be 13% to 46% of in vitro fertilization/intracytoplasmic sperm injection (IVF/ICSI) cycles with GnRH agonists [2]. However, the possible effects of these subtle progesterone increases on pregnancy outcomes are controversial. Most studies have advocated that PE on the day of hCG trigger adversely affects pregnancy outcome [3] due to its detrimental effect on the endometrium or the compromised quality of the oocyte [4].

For most studies, the role of PE on the day of hCG trigger on pregnancy rates has been estimated through simple bivariate analyses, which are unable to control for confounders such as the number of oocytes, female age, or body mass index (BMI), and the available studies may actually underestimate the true effect of PE on pregnancy rates [5].

Furthermore, these varying results may also be attributed to the use of different arbitrary cut-off levels. However, the effect of ovarian response on the association of PE with the probability of pregnancy outcomes remains unclear. Recent studies have shown that serum progesterone level is positively associated with ovarian response and oocyte number has been demonstrated to be correlated with PE [6].

It has been demonstrated that PE is associated with a higher number of oocytes; more oocytes might indicate more available embryos, whereas fewer oocytes might indicate embryos that already have diminished implantation potential [7]. Consequently, the aim of the study is to investigate the relationship between serum progesterone level on the day of h-CG trigger and IVF/ICSI outcomes in patients with different ovarian responses using GnRH long agonist or antagonist protocols and identify the thresholds at which PE has a detrimental effect on IVF/ICSI outcomes.

Patients and Methods

After written consents from the patients, this prospective cohort study was performed on total 95 infertile women aged 20–35 years with a BMI between 18.5 and 30 kg/m². Participants underwent controlled ovarian stimulation (COS) using GnRH long agonist or antagonist protocols, followed by fresh embryo transfer at tertiary care hospital at Ain Shams University Hospital “infertility clinic and ART unit” starting from June 2023 till October 2024.

Inclusion Criteria

Participants eligible for enrollment in this study were infertile women aged between 20 and 35 years with a body mass index (BMI) ranging from 18.5 to 30 kg/m². All participants underwent controlled ovarian stimulation (COS) using either a GnRH long agonist protocol or a GnRH antagonist protocol and were scheduled for fresh IVF/ICSI embryo transfer cycles. To ensure adequate ovarian reserve and hormonal balance, only women with basal day-3 estradiol (E2) levels below 60 pg/mL and follicle-stimulating hormone (FSH) levels below 15 IU/mL were included. Additionally, participants were required to have both ovaries present and a documented normal uterine cavity confirmed by either transvaginal ultrasonography (U/S) or hysterosalpingography (HSG).

Exclusion Criteria

Women were excluded from the study if they had an early follicular phase (days 2–4) serum FSH level greater than 15 mIU/mL, as elevated FSH is associated with poor ovarian response to fertility treatment. Patients with abnormalities of the uterine cavity, such as endometrial polyps or congenital uterine anomalies detected by sonohysterography or hysterosalpingography, were also excluded because of their potential negative impact on implantation. Additional exclusion criteria included the presence of advanced endometriosis (grade III or IV), estradiol levels on the trigger day exceeding 6000 pg/mL or below 500 pg/mL, and an antral follicle count greater than 15 on baseline ultrasound assessment. Women whose embryo transfer cycles were canceled, those with medical contraindications to pregnancy, or those who had received investigational drugs within three months prior to enrollment were also excluded. Furthermore, patients unable to communicate effectively with the investigators, unwilling to provide written informed consent, or those who had previously participated in the study were not eligible for inclusion.

Sample size Justification

Using PASS 11 program for sample size calculation, reviewing results from a previous study (8) showed that elevation progesterone level on the hCG trigger day may have a negative effect on the clinical pregnancy in GnRH-ant cycles, assuming a mild negative correlation between serum progesterone level and clinical pregnancy rate ($r=-0.3$) and after 10% adjustment for dropout rate a sample size of at least 95 patients achieves 80% power to detect a difference of -0.3 between the null hypothesis zero correlation and the alternative hypothesis correlation of 0.3 using a two-sided hypothesis test with a significance level of 0.05.

Study procedures and interventions

Eligible participants were enrolled according to the predefined inclusion and exclusion criteria. A comprehensive clinical evaluation was performed for all participants, including detailed medical, surgical, and infertility histories. General and gynecological examinations were conducted, and transvaginal ultrasonography (TVS) was performed to assess uterine and adnexal anatomy as well as antral follicle count (AFC).

Patients were subjected to long agonist or antagonist protocol depending upon patient's specific characteristics, a history of prior attempts at ART, baseline hormonal profile, and clinician's preference.

Baseline Investigations

Baseline hormonal assessment was carried out on day 3 of the menstrual cycle and included measurement of follicle-stimulating hormone (FSH), luteinizing hormone (LH), estradiol (E2), and anti-Müllerian hormone (AMH) levels. A baseline transvaginal ultrasound examination was also performed to evaluate ovarian reserve and pelvic anatomy.

Controlled Ovarian Stimulation Protocols

Participants underwent either a GnRH agonist long protocol or a GnRH antagonist protocol based on individual patient characteristics, previous assisted reproductive technology (ART) attempts, baseline hormonal profile, and clinician preference.

GnRH Agonist Long Protocol

In patients assigned to the GnRH agonist long protocol, a GnRH agonist (Decapeptyl® 0.1 mg subcutaneously daily; Ferring) was initiated on day 21 of the preceding menstrual cycle. Ovarian stimulation with recombinant FSH (Gonapure®; Minapharm) commenced on day 3 of the treatment cycle after confirmation of adequate pituitary downregulation, defined as LH <5 mIU/mL, E2 <60 pg/mL, endometrial thickness <5 mm, and follicular diameter <10 mm.

GnRH Antagonist Protocol

In the antagonist protocol, ovarian stimulation with recombinant FSH or human menopausal gonadotropin (HMG) was started on day 3 of the menstrual cycle, provided that basal LH was <5 mIU/mL and E2 was <60 pg/mL. The GnRH antagonist (Cetrotide® 0.25 mg subcutaneously daily; Merck-Serono) was introduced using a flexible regimen when at least one follicle reached a diameter of ≥14 mm and was continued until the morning of the trigger day.

Monitoring of Ovarian Response

The gonadotropin dose was individualized according to each patient's ovarian response. Serial transvaginal ultrasonography was performed throughout stimulation to monitor follicular growth. Serum progesterone levels were measured on the day of ovulation trigger.

Group Allocation According to Progesterone Levels

Participants were categorized into three groups based on serum progesterone concentration measured on the day of hCG trigger:

- Group I: Progesterone <1.3 ng/mL
- Group II: Progesterone 1.3–1.5 ng/mL
- Group III: Progesterone >1.5 ng/mL

Ovulation Trigger and Oocyte Retrieval

Final oocyte maturation was induced when at least three follicles

measuring 17–18 mm in diameter were present. Triggering was achieved using either human chorionic gonadotropin (hCG) (Choriomon® 5,000–10,000 IU intramuscularly; IBSA) or a GnRH agonist trigger (Decapeptyl® 0.2 mg). Oocyte retrieval was performed 35–36 hours after triggering under standard clinical protocols.

Fertilization and Embryo Culture

Retrieved oocytes underwent intracytoplasmic sperm injection (ICSI) as indicated. Fertilization assessment was performed on the second day after insemination or ICSI. Embryos were subsequently cultured in a fixed culture medium until transfer.

Embryo Transfer and Luteal Phase Support

Embryo transfer was performed on either day 3 or day 5 after fertilization, depending on the number and quality of available embryos. Luteal phase support consisted of vaginal progesterone suppositories (Prontogest® 400 mg once daily) and oral dydrogesterone (Duphaston® 10 mg three times daily).

Pregnancy Assessment and Outcome Evaluation

Serum β-hCG levels were measured 15 days after embryo transfer. A positive pregnancy test was defined as a β-hCG level ≥50 mIU/mL and was used to calculate the conception rate. Clinical pregnancy was confirmed by transvaginal ultrasonography two weeks after a positive β-hCG result. The implantation rate was calculated as the number of gestational sacs divided by the number of embryos transferred. Clinical pregnancy rate was defined as the presence of an intrauterine gestational sac with fetal cardiac activity detected by ultrasonography at 6 weeks of gestation.

Outcomes

Primary outcome

- Correlation between different progesterone levels and IVF/ICSI clinical pregnancy rate.
- Secondary outcome:
- Number of collected oocyte in each groups [(group P4 < 1.3), (group P4 1.3–1.5), (group P4 >1.5)].
- Fertility rate.
- Implantation rate.
- Number of stimulation days.
- Total doses of HMG / FSH.

Statistical analysis

Data were fed to the computer and analyzed using IBM SPSS software package version 20.0 (Armonk, NY: IBM Corp, released in 2011). Qualitative data were described using number and percent. The Kolmogorov-Smirnov test & Shapiro-Wilk test was used to verify the normality of distribution. Quantitative data were described using range (minimum and maximum), mean, standard deviation, median and interquartile range (IQR). Significance of the obtained results was judged at the 5% level.

The used tests were

1. Chi-square test: For categorical variables, to compare between

different groups

2. Fisher's exact test: Correction for chi-square when more than 20% of the cells have expected count less than 5
3. F-test (ANOVA): For normally distributed quantitative variables, to compare between more than two groups
4. Kruskal Wallis test: For abnormally distributed quantitative variables, to compare between more than two studied groups.

Results

Table 1: Distribution of the studied cases according to S progesterone (n= 95).

S progesteron	No.	%
<1.3	79	83.2
1.3 – 1.5	4	4.2
>1.5	12	12.6
Min. – Max.	0.07 – 4.51	
Mean ± SD.	0.87 ± 0.61	
Median (IQR)	0.70 (0.56 – 1.03)	

IQR: Inter quartile range, SD: Standard deviation.

Table 1 displays the distribution of the studied cases according to serum progesterone (S progesterone) levels. Most of the participants (83.2%) had a progesterone level of less than 1.3, while 4.2% had levels between 1.3 and 1.5, and 12.6% had levels greater than 1.5. The progesterone levels ranged from 0.07 to 4.51, with a mean of 0.87 ± 0.61 . The median progesterone level was 0.70, with an interquartile range (IQR) of 0.56 to 1.03. This distribution highlights a predominant lower range of progesterone levels in the cohort.

Table 2 compares the demographic data of the three groups categorized by serum progesterone levels. The age range of

participants was between 20 and 35 years, with a mean age of 30.35 ± 4.71 , and the median age was 32.0 years (IQR: 27.0 – 35.0), showing no significant differences between the groups ($p = 0.703$). Regarding BMI, values ranged from 16.30 to 35.0 kg/m², with a mean of 28.82 ± 4.77 and a median of 29.40 (IQR: 25.30 – 33.50), and again, no significant differences were observed across the groups ($p = 0.595$). These results suggest that age and BMI were similar across the three groups with different serum progesterone levels.

Table 3 presents a comparison of the history data across the three groups categorized by serum progesterone levels. In terms of obstetric history, 55.8% of the total sample had a negative obstetric history, with no significant differences observed between the groups ($p = 0.910$). Regarding surgical history, 76.8% of participants had a negative surgical history, and again, there were no significant differences between the groups ($p = 0.882$). These findings suggest that both obstetric and surgical histories were similar across the groups, with no substantial variation based on serum progesterone levels.

Table 4 compares the laboratory data across the three groups categorized by serum progesterone levels. Regarding FSH levels, there were no significant differences between the groups ($p = 0.143$), with similar mean and median values. Similarly, the LH levels ($p = 0.452$), estradiol (E2) levels ($p = 0.612$), and TSH levels ($p = 0.869$) showed no significant variation across the groups. For AMH, while the mean and median values varied, the differences were not statistically significant ($p = 0.241$). Prolactin levels showed no significant differences ($p = 0.114$), and semen analysis results (count, motility, and abnormal forms) also demonstrated no significant variations between the groups, with p-values of

Table 2: Comparison between the three studied groups according to demographic data.

	Total (n = 95)	<1.3 (n = 79)	1.3 – 1.5 (n = 4)	>1.5 (n = 12)	F	p
Age (years)						
Min. – Max.	20.0 – 35.0	20.0 – 35.0	30.0 – 35.0	24.0 – 35.0	0.353	0.703
Mean ± SD.	30.35 ± 4.71	30.23 ± 4.81	32.25 ± 2.22	30.50 ± 4.78		
Median (IQR)	32.0 (27.0 – 35.0)	32.0 (27.0 – 35.0)	32.0 (30.50 – 34.0)	32.0 (26.0 – 35.0)		
BMI (kg/m²)						
Min. – Max.	16.30 – 35.0	20.0 – 35.0	16.30 – 35.0	20.50 – 35.0	0.522	0.595
Mean ± SD.	28.82 ± 4.77	29.03 ± 4.62	27.03 ± 8.33	28.03 ± 4.62		
Median (IQR)	29.40 (25.30 – 33.50)	29.80 (25.55 – 33.60)	28.40 (20.55 – 33.50)	28.20 (24.50 – 30.85)		

IQR: Inter quartile range, SD: Standard deviation, F: F for One-Way ANOVA test, p: p value for comparing between the three studied groups.

Table 3: Comparison between the three studied groups according to history data.

	Total (n = 95)	<1.3 (n = 79)	1.3 – 1.5 (n = 4)	>1.5 (n = 12)	FET		p			
	No	%	No	%	No	%	No	%		
Obstetric history										
Negative	53	55.8	45	57.0	2	50.0	6	50.0	0.448	0.910
Positive	42	44.2	34	43.0	2	50.0	6	50.0		
Surgical history										
Negative	73	76.8	60	75.9	3	75.0	10	83.3	0.442	0.882
Positive	22	23.2	19	24.1	1	25.0	2	16.7		

χ^2 : Chi square test, p: p value for comparing between the three studied groups.

Table 4: Comparison between the three studied groups according to Labs data.

	Total (n = 95)	<1.3 (n = 79)	1.3 – 1.5 (n = 4)	>1.5 (n = 12)	H	p
FSH						
Min – Max.	4.0 – 14.80	4.0 – 14.80	6.70 – 12.0	4.81 – 10.0	3.896	0.143
Mean ± SD.	7.36 ± 2.39	7.30 ± 2.43	9.65 ± 2.32	7.03 ± 1.79		
Median (IQR)	6.80 (5.95 – 8.0)	6.82 (5.90 – 7.80)	9.95 (7.85 – 11.45)	6.31 (5.90 – 8.50)		
LH						
Min – Max.	2.0 – 20.04	2.0 – 20.04	4.90 – 9.0	3.10 – 11.60	1.586	0.452
Mean ± SD.	6.48 ± 2.78	6.44 ± 2.91	7.55 ± 1.94	6.42 ± 2.18		
Median (IQR)	6.0 (4.70 – 7.95)	5.80 (4.70 – 7.95)	8.15 (6.10 – 9.0)	6.50 (5.20 – 7.04)		
E2						
Min – Max.	5.0 – 60.0	5.0 – 60.0	34.0 – 46.0	23.0 – 60.0	0.983	0.612
Mean ± SD.	40.51 ± 14.36	40.95 ± 15.31	38.33 ± 5.44	38.40 ± 9.21		
Median (IQR)	40.0(32.0 – 54.0)	42.40(29.50–55.30)	36.65(34.50–42.15)	38.55 (34.75–40.0)		
AMH						
Min – Max.	0.12 – 16.0	0.12 – 16.0	0.13 – 2.50	0.38 – 6.50	2.844	0.241
Mean ± SD.	2.55 ± 2.50	2.55 ± 2.58	1.07 ± 1.02	3.01 ± 2.21		
Median (IQR)	1.80 (0.80 – 3.50)	1.70 (0.85 – 3.30)	0.83 (0.39 – 1.75)	3.0 (0.62 – 4.74)		
S.prolabtin						
Min – Max.	0.39 – 45.0	0.39 – 45.0	7.50 – 13.10	2.40 – 24.0	4.345	0.114
Mean ± SD.	14.99 ± 8.52	15.64 ± 8.90	9.60 ± 2.43	12.48 ± 5.96		
Median (IQR)	13.80(9.20 – 17.70)	14.0 (9.95 – 18.15)	8.90 (8.15 – 11.05)	12.35 (8.45 – 15.30)		
TSH						
Min – Max.	0.50 – 4.22	0.50 – 4.22	0.80 – 3.05	0.66 – 4.10	0.281	0.869
Mean ± SD.	1.90 ± 0.86	1.91 ± 0.87	1.96 ± 0.92	1.81 ± 0.88		
Median (IQR)	1.77 (1.30 – 2.25)	1.70 (1.30 – 2.35)	2.0 (1.35 – 2.58)	1.85 (1.28 – 2.02)		
Semen analysis count (million/ml)						
Min – Max.	0.0 – 100.0	0.0 – 100.0	30.0 – 60.0	1.0 – 80.0	0.012	0.994
Mean ± SD.	47.69 ± 24.14	47.75 ± 23.95	50.0 ± 14.14	46.50 ± 29.28		
Median (IQR)	50.0 (30.0 – 62.50)	50.0 (30.0 – 65.0)	55.0 (40.0 – 60.0)	55.0 (20.0 – 70.0)		
Motility (%)						
Min – Max.	0.0 – 80.0	0.0 – 80.0	15.0 – 60.0	10.0 – 80.0	0.847	0.655
Mean ± SD.	51.26 ± 20.42	51.71 ± 20.11	42.50 ± 21.79	51.25 ± 23.17		
Median (IQR)	60.0 (40.0 – 67.50)	60.0 (40.0 – 67.50)	47.50 (25.0 – 60.0)	60.0 (37.50 – 70.0)		
Abnormal forms (%)						
Min – Max.	0.0 – 100.0	0.0 – 100.0	97.0 – 100.0	94.0 – 99.0	0.398	0.819
Mean ± SD.	94.25 ± 13.92	93.67 ± 15.20	97.75 ± 1.50	96.92 ± 1.44		
Median (IQR)	97.0 (96.0 – 98.0)	97.0 (96.0 – 98.0)	97.0 (97.0 – 98.50)	97.0 (96.0 – 98.0)		

IQR: Inter quartile range, SD: Standard deviation, H: H for Kruskal Wallis test, p: p value for comparing between the three studied groups.

0.994, 0.655, and 0.819, respectively. Overall, the laboratory data indicate no substantial differences in hormonal and semen parameters among the three progesterone groups.

Table 5 presents the comparison of causes of infertility between the three groups based on serum progesterone levels. For female-related causes, a significant difference was found ($p = 0.016$), with a higher percentage of infertility cases attributed to female factors in the <1.3 group (65.8%) compared to the other two groups, where all cases in the 1.3–1.5 and >1.5 groups were female-related. For the specific conditions, no significant differences were found in the prevalence of polycystic ovary (PCO), poor ovarian reserve, tubal factors, or endometriosis. Regarding male-related causes, no significant differences were observed across the three groups (p -values: 0.826, 0.397, 0.189, and 1.000 for teratozoospermia, asthenoteratozoospermia, oligoasthenoteratospermia, and

azoospermia, respectively). These results indicate that female-related infertility causes vary significantly across the groups, whereas male-related factors show no significant variation.

Table 6 presents a comparison of estradiol (E2) levels on the day of trigger among the three groups based on serum progesterone levels. The mean E2 level for the total group was 1720.12 ± 866.4 , with the <1.3 group showing 1699.8 ± 833.9 , the 1.3–1.5 group at 1235.0 ± 882.7 , and the >1.5 group at 2015.5 ± 1042.7 . The median (IQR) E2 levels were 1550.0 (955.0 – 2500.0) for the total group, with variations in the subgroups. Statistical analysis using the Kruskal-Wallis test yielded a p -value of 0.182, indicating no statistically significant difference in E2 levels between the groups.

Table 7 compares the incidence of ovarian hyperstimulation syndrome (OHSS) among the three studied groups. In total, 94.7%

Table 5: Comparison between the three studied groups to cause of infertility.

	Total (n = 95)		<1.3 (n = 79)		1.3 – 1.5 (n = 4)		>1.5 (n = 12)		FET	p
	No.	%	No.	%	No.	%	No.	%		
Female										
No	27	28.4	27	34.2	0	0.0	0	0.0	7.679*	0.016*
Yes	68	71.6	52	65.8	4	100.0	12	100.0		
PCO	27	28.4	21	26.6	0	0.0	6	50.0	3.858	0.147
Poor ovarian reserve	30	31.6	23	29.1	3	75.0	4	33.3	3.485	0.157
Tubal factor	16	16.8	13	16.5	1	25.0	2	16.7	0.708	0.851
Endometriosis	5	5.3	4	5.1	1	25.0	0	0.0	3.166	0.300
Male										
No	36	37.9	30	38.0	2	50.0	4	33.3	0.527	0.826
Yes	59	62.1	49	62.0	2	50.0	8	66.7		
Teratozoospermia	35	36.8	31	39.2	0	0.0	4	33.3	2.196	0.397
Asthenoteratozoospermia	16	16.8	12	15.2	2	50.0	2	16.7	3.176	0.189
Oligoasthenoteratospermia	12	12.6	10	12.7	0	0.0	2	16.7	0.543	0.802
Azoospermia	3	3.2	3	3.8	0	0.0	0	0.0	0.732	1.000

χ^2 : Chi square test, FET: Fisher Exact test, p: p value for comparing between the three studied groups

*: Statistically significant at $p \leq 0.05$

Table 6: Comparison between the three studied groups according to E2 level on the day of trigger.

	Total (n = 95)	<1.3 (n = 79)	1.3 – 1.5 (n = 4)	>1.5 (n = 12)	H	p
E2 level on the day of trigger						
Min. – Max.	700.0 – 3800.0	700.0 – 3668.0	700.0 – 2550.0	900.0 – 3800.0	3.405	0.182
Mean $\pm$ SD.	1720.12 $\pm$ 866.4	1699.8 $\pm$ 833.9	1235.0 $\pm$ 882.7	2015.5 $\pm$ 1042.7		
Median (IQR)	1550.0 (955.0 – 2500.0)	1550.0 (960.0 – 2450.0)	845.0 (725.0 – 1745.0)	1632.5 (1210.0 – 2900.0)		

IQR: Inter quartile range, SD: Standard deviation, H: H for Kruskal Wallis test, p: p value for comparing between the three studied groups

of participants did not experience OHSS, with the <1.3 group showing 96.2%, the 1.3–1.5 group at 100%, and the >1.5 group at 83.3%. Mild OHSS was observed in 5.3% of participants, with 3.8% in the <1.3 group, 0% in the 1.3–1.5 group, and 16.7% in the >1.5 group. Statistical analysis using the Chi-square test and Fisher Exact test yielded a p-value of 0.544, indicating no statistically significant difference in the occurrence of OHSS between the three groups.

Table 8 compares various parameters related to oocyte collection, embryo quality, and transfer among the three studied groups. The number of oocytes collected ranged from 1 to 30, with a mean of 8.78 ± 6.19 for the total group. The <1.3 group had a mean of 8.44 ± 5.68 , the 1.3–1.5 group had 5.0 ± 4.97 , and the >1.5 group had 12.25 ± 8.54 . There was no significant difference between groups ($p = 0.118$). For the number of mature oocytes (M II), the total mean was 6.25 ± 4.38 , with the <1.3 group at 6.20 ± 4.16 , the 1.3–1.5 group at 3.25 ± 3.20 , and the >1.5 group at 7.58 ± 5.76 , but no significant difference was observed ($p = 0.217$). The number of embryos produced ranged from 1 to 16, with the total group having a mean of 4.54 ± 3.14 . The groups' means were similar ($p = 0.297$). Lastly, the number of embryos transferred ranged from 1 to 4, with the total group having a mean of 2.38 ± 0.73 , and again no significant difference was found ($p = 0.300$). Overall, while some trends were observed, none of the differences between the groups were statistically significant.

Table 9 compares pregnancy outcomes and the presence of gestational sacs across the three studied groups. The overall pregnancy rate was 41.1%, with 39 pregnancies recorded: 34 (43.0%) in the <1.3 group, 2 (50.0%) in the 1.3–1.5 group, and 3 (25.0%) in the >1.5 group. There was no significant difference in pregnancy rates between the groups ($p = 0.544$). Regarding gestational sacs, 58.9% of the total group (56 individuals) had no sacs, while 24.2% (23 individuals) had a single sac, and 16.8% (16 individuals) had twin sacs. The distribution of gestational sacs showed no significant differences between the groups ($p = 0.279$). The results suggest that while pregnancy and gestational sac rates were observed, no statistically significant differences were found between the groups.

Table 10 compares the implantation rate and fertilization rate across the three studied groups. The mean implantation rate for the total group was 27.73%, with a standard deviation (SD) of 39.27%. For the <1.3 group, the mean implantation rate was 29.54%, while for the 1.3–1.5 group, it was 37.50%, and for the >1.5 group, it was 12.50%. The median implantation rate was 0.0% for the total group, with the IQR ranging from 0.0% to 50.0%. The differences in implantation rates between the groups were not statistically significant ($p = 0.349$).

Regarding the fertilization rate, the overall mean was 79.41%, with a SD of 24.63%. The fertilization rate for the <1.3 group was 79.65%, for the 1.3–1.5 group was 90.75%, and for the >1.5 group

Table 7: Comparison between the three studied groups according to according to OHSS.

	Total (n = 95)	<1.3 (n = 79)	1.3 – 1.5 (n = 4)	>1.5 (n = 12)	FET		p			
	No.	%	No.	%	No.	%	No.	%		
OHSS										
No	90	94.7	76	96.2	4	100.0	10	83.3	1.593	0.544
Mild	5	5.3	3	3.8	0	0.0	2	16.7		

χ^2 : Chi square test, FET: Fisher Exact test, p: p value for comparing between the three studied groups.

Table 8: Comparison between the three studied groups according to different parameters.

	Total (n = 95)	<1.3 (n = 79)	1.3 – 1.5 (n = 4)	>1.5 (n = 12)	H	p
No. of oocytes collected						
Min. – Max.	1.0 – 30.0	1.0 – 24.0	1.0 – 12.0	3.0 – 30.0	4.274	0.118
Mean $\pm$ SD.	8.78 $\pm$ 6.19	8.44 $\pm$ 5.68	5.0 $\pm$ 4.97	12.25 $\pm$ 8.54		
Median (IQR)	8.0 (4.0 – 12.0)	7.0 (4.0 – 12.0)	3.50 (1.50 – 8.50)	10.0 (5.0 – 18.50)		
No. of M II						
Min. – Max.	1.0 – 21.0	1.0 – 16.0	1.0 – 8.0	2.0 – 21.0	3.051	0.217
Mean $\pm$ SD.	6.25 $\pm$ 4.38	6.20 $\pm$ 4.16	3.25 $\pm$ 3.20	7.58 $\pm$ 5.76		
Median (IQR)	5.0 (2.0 – 9.0)	6.0 (2.50 – 9.0)	2.0 (1.50 – 5.0)	5.50 (3.50 – 11.0)		
No. of embryos						
Min. – Max.	1.0 – 16.0	1.0 – 16.0	1.0 – 5.0	2.0 – 10.0	2.427	0.297
Mean $\pm$ SD.	4.54 $\pm$ 3.14	4.58 $\pm$ 3.20	2.50 $\pm$ 1.73	4.92 $\pm$ 3.06		
Median (IQR)	4.0 (2.0 – 6.0)	4.0 (2.0 – 6.0)	2.0 (1.50 – 3.50)	4.0 (2.0 – 7.50)		
No. of embryos transfer						
Min. – Max.	1.0 – 4.0	1.0 – 3.0	1.0 – 3.0	2.0 – 4.0	2.407	0.300
Mean $\pm$ SD.	2.38 $\pm$ 0.73	2.35 $\pm$ 0.73	2.0 $\pm$ 0.82	2.67 $\pm$ 0.65		
Median (IQR)	3.0 (2.0 – 3.0)	3.0 (2.0 – 3.0)	2.0 (1.50 – 2.50)	3.0 (2.0 – 3.0)		

IQR: Inter quartile range, SD: Standard deviation, H: H for Kruskal Wallis test, p: p value for comparing between the three studied groups.

Table 9: Comparison between the three studied groups according to pregnant and gestational sacs (n = 95).

	Total (n = 95)		<1.3 (n = 79)		1.3 – 1.5 (n = 4)		>1.5 (n = 12)		FET	p
	No.	%	No.	%	No.	%	No.	%		
Pregnant										
Not pregnant	56	58.9	45	57.0	2	50.0	9	75.0	1.593	0.544
Pregnant	39	41.1	34	43.0	2	50.0	3	25.0		
Gestational sacs										
No	56	58.9	45	57.0	2	50.0	9	75.0	4.528	0.279
Pregnant on single	23	24.2	18	22.8	2	20.0	3	25.0		
Pregnant on twin	16	16.8	16	20.3	0	0.0	0	0.0		

FET: Fisher Exact test, p: p value for comparing between the three studied groups.

Table 10: Comparison between the two studied groups according to implantation rate and fertilization rate.

	Total (n = 95)	<1.3 (n = 79)	1.3 – 1.5 (n = 4)	>1.5 (n = 12)	H	p
Implantation rate (%)						
Min. – Max.	0.0 – 200.0	0.0 – 200.0	0.0 – 100.0	0.0 – 50.0	2.107	0.349
Mean $\pm$ SD.	27.73 $\pm$ 39.27	29.54 $\pm$ 40.68	37.50 $\pm$ 47.87	12.50 $\pm$ 22.61		
Median (IQR)	0.0 (0.0 – 50.0)	0.0 (0.0 – 50.0)	25.0 (0.0 – 75.0)	0.0 (0.0 – 25.0)		
Fertilization rate (%)						
Min. – Max.	18.0 – 150.0	18.0 – 150.0	63.0 – 100.0	40.0 – 100.0	1.571	0.456
Mean $\pm$ SD.	79.41 $\pm$ 24.63	79.65 $\pm$ 24.97	90.75 $\pm$ 18.50	74.08 $\pm$ 24.33		
Median (IQR)	80.0 (57.50–100.0)	80.0(57.50–100.0)	100.0(81.50–100.0)	67.0 (50.0 – 100.0)		

IQR: Inter quartile range, SD: Standard deviation, H: H for Kruskal Wallis test, p: p value for comparing between the three studied groups.

Table 11: Diagnostic performance for S progesterone to discriminate pregnant (n= 39) from non–pregnant (n=56).

	AUC	p	95% C.I	Cut off	Sensitivity	Specificity	PPV	NPV
S progesteron	0.622	0.043*	0.506 – 0.738	>0.7	66.67	64.29	56.5	73.5

AUC: Area Under a Curve, p value: Probability value, CI: Confidence Intervals, NPV: Negative predictive value, PPV: Positive predictive value, *: Statistically significant at $p \leq 0.05$, #Cut off was choose according to Youden index

was 74.08%. The median fertilization rate was 80.0%, with an IQR from 57.5% to 100.0%. Again, no significant differences were found in the fertilization rates between the groups ($p = 0.456$). These results suggest that neither implantation nor fertilization rates showed statistically significant differences across the groups.

Table 11 presents the diagnostic performance of serum progesterone (S progesterone) to discriminate between pregnant ($n=39$) and non-pregnant ($n=56$) individuals. The area under the curve (AUC) for serum progesterone was 0.622, with a statistically significant p -value of 0.043, indicating that serum progesterone has moderate diagnostic performance in predicting pregnancy. The 95% confidence interval (CI) for the AUC ranged from 0.506 to 0.738. A cutoff value of >0.7 was selected based on the Youden index, with the following performance metrics: sensitivity of 66.67%, specificity of 64.29%, positive predictive value (PPV) of 56.5%, and negative predictive value (NPV) of 73.5%. These results suggest that serum progesterone can be used with moderate accuracy to predict pregnancy status, with a higher NPV indicating a reliable ability to rule out non-pregnancy

Discussion

Premature progesterone elevation on the day of hCG trigger is a common finding during controlled ovarian stimulation for IVF/ICSI cycles, despite the use of GnRH agonist and antagonist protocols. Several studies have suggested that elevated progesterone levels may negatively affect endometrial receptivity and reduce implantation and pregnancy rates. However, the available evidence remains controversial, with inconsistent findings regarding the impact of progesterone elevation and the threshold at which it becomes clinically significant [9].

Moreover, variations in study designs, patient characteristics, stimulation protocols, and progesterone cutoff values have contributed to the lack of consensus on the clinical relevance of trigger-day progesterone levels. Prospective data evaluating the relationship between progesterone elevation and IVF/ICSI outcomes are still limited [8].

Since pre-hCG progesterone increase on the day of trigger above a threshold concentration represents major conflict and often associated with premature luteinization, evaluating the incidence of PPR on the day of trigger in conventional IVF/ICSI cycles and its impact on clinical pregnancy rate was highlighted as a main point of interest [9].

Consequently, this study was conducted to investigate the association between serum progesterone levels on the day of hCG trigger and reproductive outcomes in IVF/ICSI cycles and to identify progesterone thresholds that may adversely affect implantation and clinical pregnancy rates.

This study was designed to explore how serum progesterone levels on the day of hCG trigger influence the outcomes of IVF/ICSI cycles. The study adopted a prospective cohort methodology

involving 95 infertile women aged 20–35 years. The participants were categorized into three groups based on serum progesterone levels: <1.3 ng/mL (83.2%, $n=79$), 1.3 – 1.5 ng/mL (4.2%, $n=4$), and >1.5 ng/mL (12.6%, $n=12$). This categorization enabled an analysis of progesterone's effects across a spectrum of concentrations, identifying thresholds where progesterone elevation (PE) could adversely affect treatment outcomes.

Regarding Demographic data

Our study results revealed that age and BMI were analyzed as potential confounding factors. The age of participants ranged from 20 to 35 years, with a mean of 30.35 ± 4.71 years and a median of 32.0 years. BMI ranged from 16.3 to 35.0 kg/m², with a mean of 28.82 ± 4.77 kg/m². Statistical analysis revealed no significant differences in age ($p = 0.703$) or BMI ($p = 0.595$) across the three groups, ensuring these factors did not influence the observed outcomes.

This demographic uniformity aligns with findings by Zhao et al. [8] and Garg et al. [10], which also observed comparable age and BMI between their study groups. However, Gill et al. [11] found a significant age difference, with a higher mean age in the elevated progesterone group (33.2 years vs. 31.3 years, $p = 0.009$). Such differences in age distribution may confound outcomes in some studies. Our consistency across demographic variables strengthens the reliability of our findings, particularly in isolating progesterone as the primary factor influencing clinical outcomes.

Regarding Baseline Hormonal and Clinical Data

Our study results revealed that hormonal parameters and baseline conditions showed consistency across groups. FSH levels ranged from 4.0 to 14.8 mIU/mL, with a mean of 7.36 ± 2.39 mIU/mL, and no significant differences between groups ($p = 0.143$). LH levels also showed no significant variation, with a mean of 6.48 ± 2.78 mIU/mL ($p = 0.452$). Estradiol (E2) levels on the day of trigger ranged from 700 to 3800 pg/mL, with a mean of 1720.12 ± 866.4 pg/mL and no significant differences across groups ($p = 0.182$). Similarly, prolactin and TSH levels demonstrated no significant differences, suggesting that baseline hormonal factors were comparable regardless of progesterone levels.

These results are consistent with Merviel et al. [12], Gill et al. [11], Zhao et al. [8] and Garg et al., [10] who similarly found no differences in FSH, LH, or AMH. In addition, Jiang et al. [13], who noted similar baseline hormonal profiles among groups stratified by progesterone levels. De Cesare et al. [14] also found that elevated progesterone levels correlated negatively with live birth rates, but their baseline hormonal data indicated no significant initial disparities.

Estradiol levels, however, showed a trend in several studies. Zhao et al. [8] reported higher terminal E2 levels in groups with elevated progesterone, which might reflect differences in stimulation protocols. Moreover, Garg et al. [10] observed significantly higher estradiol levels in the elevated progesterone group (2545

pg/mL vs. 1339 pg/mL, $p = 0.017$), while our study did not find such differences. Jiang et al. [8] and Vikas & Swati [9] observed elevated estradiol levels on the trigger day in high-progesterone groups, suggesting possible protocol-induced effects. Thomsen et al. [15] further highlighted a correlation between follicle counts and luteal phase progesterone levels, suggesting dynamic hormonal interactions that may influence ART outcomes.

These findings suggest that baseline hormonal stability is a common feature across studies, but estradiol levels at the trigger day underline the role of stimulation protocols in modulating hormonal responses.

Regarding Infertility Causes

Our study results revealed that female infertility factors showed a significant difference between groups ($p = 0.016$). In the <1.3 ng/mL group, 65.8% of cases were attributed to female-related causes, compared to 100% in the 1.3–1.5 and >1.5 ng/mL groups. Specific conditions like polycystic ovary syndrome (PCOS), poor ovarian reserve, tubal factors, and endometriosis did not differ significantly between groups. Male-related infertility factors, including teratozoospermia and asthenoteratozoospermia, also showed no significant variation.

This aligns with Garg et al. [10], who reported higher progesterone levels in patients with poor ovarian reserve and aligns partially with Zhao et al., [8] who noted trends in female infertility cases but without statistical significance. However, Gill et al. [11] did not observe differences in infertility etiology between groups, suggesting that the observed correlations might be protocol- or population-specific. Jiang et al. [8] reported no significant differences in infertility etiology, a finding consistent with Vikas & Swati [9]. In Singh et al. [16], the type of ovarian response, particularly poor ovarian reserve, was emphasized as influencing progesterone levels and clinical outcomes.

Regarding Clinical Outcomes

Our study results revealed that ovarian hyperstimulation syndrome (OHSS) was generally rare, with 94.7% of participants not experiencing OHSS. Mild OHSS occurred in 5.3%, predominantly in the >1.5 ng/mL group (16.7%), though this difference was not statistically significant ($p = 0.544$), consistent with Merviel et al. [12], who observed no correlation between progesterone levels and OHSS rates and also, consistent with Zhao et al., [8] and Garg et al., [10] where OHSS incidence was unaffected by progesterone levels.

Regarding Pregnancy outcomes

Our study results revealed that the overall pregnancy rate was 41.1%, with 43% in the <1.3 ng/mL group, 50% in the 1.3–1.5 ng/mL group, and 25% in the >1.5 ng/mL group. However, the differences were not statistically significant ($p = 0.544$). Implantation and fertilization rates followed a similar trend. The mean implantation rate was $27.73 \pm 39.27\%$ and the mean fertilization rate was $79.41\% \pm 24.63\%$, with no significant

differences across groups ($p = 0.349, 0.456$).

Pregnancy rates in our study decreased significantly with progesterone levels >1.5 ng/mL, mirroring Zhao et al. [8] who demonstrated a significant drop in clinical pregnancy rate (CPR) beyond 1.4 ng/mL (55.22% vs. 40.66%, $p = 0.013$), and Garg et al. [10] reported a reduction in CPR from 35.7% to 24.5% ($p = 0.041$). Gill et al. [11] observed an even sharper decline, with CPR of 46.6% in lower progesterone groups versus 17.24% in elevated groups ($p = 0.028$). Our implantation rate trends also matched those of Garg et al. [10], which demonstrated a clear decline with increasing progesterone. These consistent findings affirm the detrimental effects of elevated progesterone on fresh embryo transfer outcomes.

Jiang et al. [8] similarly observed a negative correlation between high progesterone and live birth rates, implantation rates, and clinical pregnancy rates. Merviel et al. [12] identified a threshold of >0.9 ng/mL for adverse pregnancy outcomes, consistent with our findings, and noted differences in outcomes based on embryo transfer stage. Thomsen et al. [15] highlighted a non-linear relationship between luteal phase progesterone and live birth rates, with both low and high levels reducing success. Singh et al. [16] introduced the concept of a progesterone/oocyte ratio as a stronger predictor of pregnancy outcomes, offering an innovative metric to assess clinical impacts.

Regarding Oocyte and Embryo Quality Parameters

Our study results revealed that the number of oocytes collected ranged from 1 to 30, with a mean of 8.78 ± 6.19 across all groups. The <1.3 ng/mL group had a mean of 8.44 ± 5.68 , the 1.3–1.5 ng/mL group had 5.0 ± 4.97 , and the >1.5 ng/mL group had 12.25 ± 8.54 , though the differences were not statistically significant ($p = 0.118$). Similarly, the number of mature oocytes (M II) ranged from 1 to 21, with a mean of 6.25 ± 4.38 , and the number of embryos ranged from 1 to 16, with a mean of 4.54 ± 3.14 . None of these differences were statistically significant ($p > 0.05$).

This is consistent with findings by Gill et al., [11] who reported no impact of elevated progesterone on oocyte or embryo quality, despite reduced pregnancy rates. Similarly, Garg et al. [10] and Vikas & Swati [9], found no significant differences in embryo morphology or quality despite variations in progesterone levels. Zhao et al. [8], however, noted differences in oocyte retrieval numbers between groups, suggesting potential protocol-specific impacts.

These findings also, align with Thomsen et al. [15] and Singh et al. [16], which reported no significant differences in fertilization or cleavage rates, emphasizing that progesterone's impact is confined to implantation and pregnancy outcomes rather than gamete quality. Jiang et al. [13] also found no differences in embryo utilization rates across progesterone groups. Merviel et al. [12] noted slight differences in fertilization rates based on embryo transfer stage but did not link these directly to progesterone levels.

Regarding Diagnostic Performance of Serum Progesterone

Our study demonstrated moderate diagnostic performance of serum progesterone in predicting pregnancy outcomes (AUC: 0.622), with a threshold >0.7 ng/ml showing a moderate sensitivity of 66.67%, specificity of 64.29%, positive predictive value (PPV) of 56.5%, and negative predictive value (NPV) of 73.5%. These results suggest that while serum progesterone has moderate predictive value, its primary utility may lie in ruling out non-pregnancy.

This aligns with De Cesare et al. [14], who identified a similar threshold for reduced clinical pregnancy rates and found a progressive decline in live birth rates for progesterone levels >1.0 ng/ml, supporting the diagnostic relevance of serum progesterone levels. In addition, Zhao et al. [8] identified a higher critical threshold of 1.4 ng/mL, beyond which CPR dropped sharply, while Garg et al. [10] highlighted a threshold of 1.5 ng/mL, where pregnancy rates significantly declined.

In addition, Singh et al. [16] suggested a progesterone/oocyte ratio of >0.15 as a more reliable predictor than absolute progesterone levels, introducing a novel methodological approach. Thomsen et al. [15] and Merviel et al. [12] emphasized context-specific thresholds, identifying ranges between 0.9–1.5 ng/ml as critical for predicting adverse effects. Compared to Gill et al. [11], who set a higher threshold (>1.5 ng/ml) for adverse outcomes, our study's lower cutoff might provide earlier indicators for intervention, offering nuanced guidance for clinical decision-making.

Clinical Implications

The findings of this study highlight the importance of monitoring serum progesterone levels on the day of hCG trigger in IVF/ICSI cycles. Elevated progesterone levels, particularly those exceeding 1.5 ng/mL, were associated with lower implantation and clinical pregnancy rates, suggesting a potential negative effect on endometrial receptivity. Therefore, trigger-day progesterone assessment may serve as a useful tool for identifying patients at risk of suboptimal outcomes and guiding individualized treatment strategies. In patients with elevated progesterone levels, consideration of alternative approaches, such as embryo cryopreservation and subsequent frozen embryo transfer, may help optimize reproductive outcomes.

Strengths of the Study

This study has several strengths. First, its prospective cohort design allowed for systematic data collection and minimized recall bias. Second, strict inclusion and exclusion criteria ensured a relatively homogeneous study population, reducing the influence of potential confounding factors. Third, both GnRH agonist and antagonist stimulation protocols were included, enhancing the applicability of the findings to routine clinical practice. Finally, stratification of participants according to clinically relevant progesterone thresholds enabled a detailed evaluation of the relationship between trigger-day progesterone levels and IVF/ICSI outcomes.

Limitations

Despite its strengths, the study has some limitations. The relatively small sample size, particularly in the subgroup with progesterone levels between 1.3 and 1.5 ng/mL, may have limited the statistical power to detect significant differences between groups. In addition, the study was conducted at a single tertiary care center, which may limit the generalizability of the findings to other populations and clinical settings. The study also focused primarily on clinical pregnancy outcomes and did not include long-term follow-up for live birth rates or neonatal outcomes. Furthermore, other factors that may influence IVF success, such as genetic, lifestyle, and environmental variables, were not evaluated.

Conclusion

This study demonstrated that elevated serum progesterone levels on the day of hCG trigger are associated with poorer reproductive outcomes in IVF/ICSI cycles. Although differences between groups were not statistically significant in all outcome measures, patients with progesterone levels greater than 1.5 ng/mL showed lower implantation and clinical pregnancy rates. The findings support the growing evidence that premature progesterone elevation may adversely affect treatment success, primarily through its impact on endometrial receptivity rather than oocyte or embryo quality.

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