DOI: https://doi.org/10.5114/pm.2023.127964

Menopause Rev 2023; 22(2): 83-86

ORIGINAL PAPER

A paradigm of higher success rate of intracytoplasmic sperm injection – preovulatory human chorionic gonadotropin-day serum oestradiol

Ahmed Elsayad[1], Wael Khafagy[2], Walaa ElBassioune[2], Mahmoud Rady[2], Ahmed Abdeltawab[2], Muhammad Altoraky[2], Mahmoud Hegazy[2], Waled Ayad[2], Moatazza Alghazaly[2], Ahmed Elshorbagy[2], Ahmed Shafik Almorsy[2], Ahmed Saeed[2], Elmetwally Farouk [2], Ehab Elhelw[2], Hamada Abuelmatti[2], Mohammed El-Husseny Elkadosi[1]

[1]Department of Obstetrics and Gynaecology, Zagazig University, Egypt
[2]Department of Obstetrics and Gynaecology, Al-Azhar University, Egypt

Abstract

Introduction: The aim of the study is to evaluate the correlation between the level of serum oestradiol (E_2) on the day of human chorionic gonadotropin (hCG-day) administration and successful intracytoplasmic sperm injection (ICSI) outcome.

Material and methods: This prospective study was performed during the period from January 2019 to September 2021, at Zagazig Obstetrics and Gynecology Department, and Al-Azhar Obstetrics and Gynecology Department, and private ART centers. One hundred and fifty women attending the infertility clinic for ICSI cycles. All women were divided into 5 groups according to the serum E_2 level on the day of hCG administration: Group A – serum E_2 < 1000 pg/ml; Group B – serum E_2 1000 to < 2000 pg/ml; Group C – serum E_2 2000 to < 3000 pg/ml;Group D – serum E_2 3000 to < 4000 pg/ml; Group E – serum E_2 ≥ 4000 pg/ml.

Results: The highest fertilization rate (58.1%) was among women with E_2 ≥ 4000 pg/ml, while the lowest (37%) was in women with E_2 1000 to < 2000 pg/ml. Also, the highest pregnancy rate (21.5%) was among women with E_2 > 4000 pg/ml, while the lowest (5.3%) was in women with E_2 < 1000 pg/ml. In the current study the median serum E_2 level on the day of hCG administration was highly significant in women who became pregnant when compared to women who did not. The best cut-off value of serum E_2 at hCG administration was ≥ 3682.3 pg/ml.

Conclusions: this study suggests that the optimal range of E_2 level for achieving a successful pregnancy is > 4000 pg/ml.

Key words: preovulatory hCG, serum oestradiol, intracytoplasmic sperm injection.

Introduction

Intracytoplasmic sperm injection (ICSI), as a substitute to in-vitro fertilization (IVF), is now widely used to overcome male and female infertility problems [1, 2]. Accurate timing of human chorionic gonadotropin (hCG) administration requires careful cycle monitoring because the success of the cycle depends on it [3]. Human chorionic gonadotropin administration is on time when the dominant follicle has reached suitable maturity, which is immediately before the spontaneous luteinizing hormone (LH) peak [4]. The most significant criterion for evaluating follicular maturity is the serum oestradiol (E_2) level, so the relationship between the E_2 level on the day of hCG injection and the cycle outcome is not surprising. Many techniques – namely controlled ovarian hyperstimulation (COH) – were used to attain multiple oocytes; nevertheless, this circumstance results in supraphysiologic E_2. Serum E_2 concentrations during COH are increased 10-fold compared with those of natural cycles [5]. On the day of hCG administration, high E_2 levels might cause variable ICSI outcomes, caused by unsettled endometrial receptivity [6]. Serum E_2 plays a basic role in the reproductive system, being synthesized by dominant follicles, and plasma E_2 levels are used as an index to evaluate follicle maturity [7]. Oestradiol plays a fundamental role in cervical mucus regulation, endometrial propagation for embryo implantation, and initiation of mid-cycle LH surge. During COH, E_2 levels are directly proportional to follicle size, with 200 pg/ml equivalent to a mature ovarian follicle. Nevertheless, high E_2 levels may be incriminated in unwanted adverse outcomes of IVF/ICSI cycles and decreased endometrial receptivity [8]. Several studies have suggested that high serum E_2 concentration on hCG administration day has a positive effect on pregnancy,

Corresponding author:
Ahmed Saeed, lecturer of Obgyn Al-Azhar University, Cairo, Egypt, e-mail: krimabdel2013@gmail.com

Submitted: 15.02.2022
Accepted: 07.09.2022

however, others have reported contrary to this [9]. During COH, supraphysiological E_2 levels are assumed to be interrelated with ICSI outcome, and consequently it was meticulously investigated for many years. Nevertheless, such a relationship could neither be established nor repudiated. Endometrial receptivity, embryo viability, and adhesion capacity are the possible mechanisms through which supraphysiological E_2 levels may affect pregnancy rates in ICSI cycles [10]. The aim of the current work was to assess the relationship between the serum E_2 level on the day of hCG administration and the ICSI outcome.

Material and methods

The current prospective cohort study was conducted during the period from January 2019 to September 2021 at Zigzag Obstetrics and Gynecology Department, Al-Azhar Obstetrics and Gynecology Department and private ART centers after approval of research ethics committee. One hundred and fifty women attended the infertility clinic were recruited. Before participation, written informed consent was obtained from all women. Women with submucous fibroid > 5 cm, ovarian cyst > 2 cm, hydrosalpinx, endometrial polyp, and follicular stimulating hormone (FSH) > 15 IU/l were excluded. All women were subjected to detailed history, physical examination, transvaginal ultrasound (TVUS) examination, and laboratory investigations. Transvaginal ultrasound was done on the 3rd day of the natural cycle for exclusion criteria and for assessment of antral follicular count (AFC). Laboratory investigations included basal serum FSH, basal serum LH, basal serum E_2, serum prolactin, and serum thyroid stimulating hormone (TSH) on the 3rd day of the cycle.

Controlled ovarian hyperstimulation

Controlled ovarian hyperstimulation (COH) was done by the standardized long protocol. The select of it was based on the women's age, body mass index (BMI), basal FSH, and AFC. 500 µg folic acid was received daily by all women before induction. Combined oral contraceptive pills were received on day 2 of the previous cycle. Then standard mid-luteal protocol started on day 21 of the previous cycle by GnRH agonist triptorelin acetate (Decapeptyl®, Ferring, Germany) administered daily at a dose of 0.1 mg subcutaneous injection until ovarian suppression was detected by serum E_2 (≤ 50 pg/ml) on the 2nd day of the menstrual cycle. Then the GnRH agonist was continued daily at a dose of 0.05 mg, and IM daily injections of HP-urinary HMG (Merional® 75 IU LH and 75 IU FSH, IBSA) were given. Follicular development was started in the 6th day of ovarian stimulation, and the dose was adjusted according to ovarian response, which was evaluated by TVUS (Mindray®, 50/60 Hz, Hamburg, Germany). When more than 2 leading follicles reached ≥ 18 mm in diameter, ovulation trigger was done using intramuscular injection of hCG (Choriomon® 10,000 IU, IBSA). Venous samples were obtained from women by venipuncture and processed one hour after withdrawal for E_2 assessment just prior to hCG injection, and serum was stored at 2–8°C for less than 8 hours and assayed for E_2.

Serum oestradiol assay

The ADVIA Centaur Estradiol-6 assay (a competitive immunoassay using direct chemiluminescent technology). ADVIA measured oestradiol concentrations up to 1000 pg/ml (3670 pmol/l) with minimum detectable concentration (sensitivity) of l0 pg/ml (36.7 pmol/l). The conversion formula was 1 pg/ml = 3.67 pmol/l. The included women in our study (n = 150) were divided into 5 groups according to the serum E_2 level on day of hCG administration: Group A – serum E_2 < 1000 pg/ml; Group B – serum E_2 1000 to < 2000 pg/ml; Group C – serum E_2 2000 to < 3000 pg/ml; Group D – serum E_2 3000 to < 4000 pg/ml; and Group E – serum E_2 ≥ 4000 pg/ml. Transvaginal ultrasound-guided oocyte retrieval was done 36 hours after hCG injection under general anaesthesia. Oocyte maturation was evaluated by an embryologist. Oocytes were cultured in modified culture media (global total). Luteal phase support by micronized progesterone was given starting the day after hCG until the occurrence of biochemical pregnancy, which, confirmed by serum B-hCG concentration, was > 50 IU/l on the 14th day after embryo transfer, and clinical pregnancy was defined as the presence of a gestational sac with foetal heart beat on ultrasound performed 7 weeks after embryo transfer. If the patient became pregnant, she continued with progesterone supplementation until 12 weeks of pregnancy.

Statistical analysis

Data were analysed using SPSS® for Windows®, version 15.0 (SPSS, Inc, USA). Description of quantitative (numerical) variables was performed in the form of mean, standard deviation, and range. Description of qualitative (categorical) data was performed in the form of number of cases and percentage. Analysis of numerical variables was performed by using the independent Student's t-test. Analysis of categorical data was performed by using the χ^2 test. The ANOVA test was used to compare multiple variables. The Mann-Whitney U test was used to compare quantitative variables. Diagnostic accuracy was assessed using the following terms: sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and overall ac-

curacy. Receiver operating characteristic curve analysis was constructed for estimation of the association between serum E_2 levels on the day of hCG administration and the clinical pregnancy outcome. A two-sided *p*-value < 0.05 was considered statistically significant. The significance level was set at 0.05.

Results

A total of 150 infertile women were included in the current study; their mean age was 29.4 ±3.8 years and their mean body mass index (BMI) was 26.5 ±4.42 kg/m^2. Most of the infertile women (58%) had primary infertility, while the rest of them (42%) had secondary infertility. The most common cause of infertility was unexplained (42.7%), while the least common cause was male factor (8.7%) (Table 1). The highest number of retrieved oocytes was 11.4 ±4.6 in group D, while the lowest number was seen in group A (7.2 ±3.2). The highest fertilization rate was in group E (58.1%) and the lowest was 37% in group A. The highest number of transferred embryos was in group E (2.9 ±0.5) while the lowest was 1.6 ±0.71 in group A. The highest pregnancy rate was (21.5%) in group E (Table 2). The median serum E_2 level on the day of hCG administration was highly significant in women who became pregnant when compared to women who did not (Table 3). The best cut-off value of serum E_2 on day of hCG administration was ≥ 3682.3 pg/ml. Serum E_2 at hCG administration ≥ 3682.3 pg/ml was associated with a successful clinical pregnancy outcome, with a sensitivity of 81.3%, specificity of 97.1%, PPV of 83.1%, NPV of 89.5%, and an overall accuracy of 83.2% (Table 4).

Table 1. Demographic data of infertile women

Parameters	Date
Patient number	150
Age (years)	29.4 ±3.8
BMI [kg/m^2]	26.5 ±4.42
Type of infertility	
Primary	87
Second	63
Duration of infertility (years)	7.5 ±7.2
Cause of infertility	
Male	13
Female (*N* = 73)	
Tubal	15
Endometr	25
Anovulation	35
Unexplained	64

Discussion

It has been described that 17 b E_2 prompts cytoplasmic maturation of germinal vesicle oocytes through in-

Table 2. Comparison of intracytoplasmic sperm injection outcomes according to serum oestradiol levels on day of human chorionic gonadotropin administration

Parameters	Group A (*n* = 56)	Group B (*n* = 30)	Group C (*n* = 26)	Group D (*n* = 24)	Group E (*n* = 14)
Number of retrieved oocytes	7.2 ±3.2	8.2 ±1.4	8.1 ±3.5[d]	11.4 ±4.6[a,c]	10 ±3.2
Fertilization rate (%)	37	39.3	38.6	42	58.1
Number of transferred embryos	1.6 ±0.71	1.8 ±0.4	1.9 ±0.7	2.4 ±0.3	2.9 ±0.5
Pregnancy rate, *n* (%)	3 (5.3)	3 (10)[a]	5 (19)[a,b]	5 (20.8)[a,b]	3 (21.5)[a,b]

Data presented as mean ± standard deviation, number (percentage). Analysis using one-way ANOVA test and LSD post-hoc test for numeric variables. Analysis using $\chi 2$ test for categorical variables.
[a] Significant ($p < 0.05$) vs. group A
[b] Significant ($p < 0.05$) vs. group B
[c] Significant ($p < 0.05$) vs. group C
[d] Significant ($p < 0.05$) vs. group D

Table 3. Comparison between women who became pregnant and those who did not, regarding serum oestradiol level on day of human chorionic gonadotropin administration

Serum E_2 level at hCG administration [pg/ml]	Women with positive clinical pregnancy (*n* = 21)	Women with negative clinical pregnancy (*n* = 79)	*p*-value*
Median	3899	1406	< 0.001

E_2 – oestradiol, hCG – human chorionic gonadotropin, NS – non-significant
* Analysis using Mann-Whitney *U* test

Table 4. Validity of the association between serum oestradiol levels on day of human chorionic gonadotropin administration and clinical pregnancy outcome

Serum E_2 [pg/ml]	Sensitivity	Specificity	PPV	NPV	Overall accuracy
≥ 3682.3 (%)	81.3	97.1	83.1	89.5	83.2

E_2 – oestradiol, NPV – negative predictive value, PPV – positive predictive value

crease in intra-cytoplasmic calcium level; this has been linked to better fertilization [11]. In the present study the causes of infertility of our patients were male factor (8.7%), anovulation and polycystic ovary syndrome (PCOS) (23.3%), tubal factor (10%), and endometriosis (15.3%). This result disagreed with that of others, who found that the causes of infertility were tubal factor (33.2%), male factor (23%), PCOS (18.4%), and endometriosis (11%) [11]. In the current study the highest number of retrieved oocytes women with E_2 3000 to < 4000 pg/ml (11.4 ±4.6) was statistically significant versus women with E_2 < 1000 pg/ml and women with E_2 2000 to < 3000 pg/ml (7.2 ±3.2) and (8.2 ±1.4), respectively, although Joo *et al*. concluded that the mean number of retrieved oocytes (15.3 ±6.6) was statistically significant in women with E_2 > 4000 pg/ml versus women with E_2 < 1000 pg/ml, women with E_2 1000–2000 pg/ml, women with E_2 2000–3000 pg/ml, and women with E_2 > 4000 pg/ml, while Mittal *et al*. reported that the mean number of retrieved oocytes increased as the serum E_2 value increased from women with E_2 1700 pg/ml to women with E_2 5600 pg/ml [3, 11]. In our study the highest number of transferred embryos was in group E; this result disagreed with Kara *et al*., 2012, who concluded that the mean number of transferred embryos (2.5 ±0.6) was statistically significant in women with E_2 > 4000 pg/ml [6]. In this study the women with $E_2 \geq 4000$ pg/ml had a higher fertilization rate (58.1%) compared with other women with different E_2 levels, but there was no significant difference among these women; this result was analogous with Kong *et al*., who reported a fertilization rate of 60%, which is not significantly different than that seen in women with dissimilar E_2 levels [5]. In the present study the highest pregnancy rate (21.5%) was in women with E_2 > 4000 pg/ml; this result is consistent with Kara *et al*., who showed that the pregnancy rate was high in women with E_2 > 4000 pg/ml, but the difference among women with different E_2 levels was not statistically significant [6]. Joo *et al*. found that the pregnancy rate rises with increasing levels of serum E_2 from women with E_2 < 1000 pg/ml to women with E_2 3000–4000 pg/ml, reaching 50% in women with E_2 3000 to 4000 pg/ml, but falling in women with E_2 > 4000 pg/ml. This may be justified by the fact that oestrogen increases endometrial proliferation and uterine perfusion, increased possibility of pregnancy. In the current study the best cut-off value of serum E_2 at hCG administration was ≥ 3899 pg/ml. A serum E_2 at hCG administration ≥ 3899 pg/ml was associated with a successful clinical pregnancy outcome, with a sensitivity of 81.3%, specificity of 97.1%, PPV of 83.1%, NPV of 89.5%, and an overall accuracy of 83.2%. However, Wu *et al*. stated that the serum E_2 cut-off value is 19,124 pmol/ml; this figure has low sensitivity (58.1%) and low specificity (49.2%), which is not implemented for attaining a successful pregnancy [10].

Conclusions

The serum E_2 level on hCG day may influence a successful ICSI outcome, and this study proposes that the optimal range of E_2 level for attaining a successful pregnancy is > 4000 pg/ml.

References

1. Wu Y, Kang X, Zheng H, Liu H, Huang Q, Liu J. Effect of paternal age on reproductive outcomes of intracytoplasmic sperm injection. PLoS One 2016; 11: e0149867.
2. Kyrou D, Kolibianakis EM, Fatemi HM, Tarlatzi TB, Devroe P, Tarlatzis BC. Increased live birth rates with GnRH agonist addition for luteal support in ICSI/IVF cycles: a systematic review and meta-analysis. Hum Reprod Update 2011; 17: 734-740.
3. Joo BS, Park SH, An BM, Kim KS, Moon SE, Moon HS. Serum estradiol levels during controlled ovarian hyperstimulation influence the pregnancy outcome of in vitro fertilization in a concentration-dependent manner. Fertil Steril 2010; 93: 442-446.
4. Devroey P, Aboulghar M, Garcia-Velasco J, et al. Improving the patient's experience of IVF/ICSI: a proposal for an ovarian stimulation protocol with GnRH antagonist co- treatment. Hum Reprod 2009; 24: 764-774.
5. Kong GWS, Cheung LP, Haines CJ, Lam PM. Comprehensive assessment of serum estradiol impact on selected physiologic markers observed during in vitro fertilization and embryo transfer cycles. J Exp Clin Assist Reprod 2009; 6: 5.
6. Kara M, Kutlu T, Sofuoglu K, Devranoglu B, Cetinkaya T. Association between serum estradiol level on the hCG administration day and IVF-ICSI outcome. Ir J Reprod Med 2012; 10: 53-58.
7. Gahlot A. Predictive value of basal FSH, endometrial thickness, pattern and serum E2 in vitro fertilization embryo transfer cycles success. Int Res J Med Sci 2014; 2: 6-11.
8. Taskin EA, Atabekoglu CS, Musah N, Oztuna D, Sonmezer M. Association of serum estradiol levels on the day of hCG administration with pregnancy rates and embryo scores in fresh ICSI/ET cycles down regulated with either GnRH agonists or GnRH antagonists. Arch Gynecol Obstet2014; 289: 399-405.
9. Hajshafiha M, Oshnouei S, Mostafavi M, Dindarian S, Kiarang N, Mohammadi S. Evaluation of the relationship between serum estradiol levels on human chorionic gonadotropin administration day and intracytoplasmic sperm injection outcomes: a retrospective population-based study. Int J Reprod Biomed 2021; 19: 599-606.
10. Wu Z, Li R, Ma Y, et al. Effect of HCG-day serum progesterone and estradiol concentrations on pregnancy outcomes in GnRH agonist cycles. Reprod Biomed Online 2012; 24: 511-520.
11. Mittal S, Gupta P, Malhotra N, Singh N. Serum estradiol as a predictor of success of in vitro fertilization. Offic J Fed Obstet Gynecol India 2014; 64: 124-129.