Dual Trigger for Final Oocyte Maturation in ICSI Cycles: Does it Make a Difference?

Original Article

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ABSTRACT

Aim: To compare the outcomes of ICSI-fresh embryo transfer cycles by experimenting with various methods of final oocyte maturation triggering.

Methods: A randomized controlled trial consisted of a total of 348 patients who underwent controlled ovarian stimulation using gonadotropin-releasing hormone (GnRH) antagonist, With 174 participants split evenly across the two groups, 174 were triggered by hCG alone (10,000 IU hCG intramuscularly) and 174 were triggered by dual trigger (0.2 mg of triptorelin subcutaneously and 5,000 IU of hCG intramuscularly). Measures of outcome included the clinical pregnancy rate, ongoing pregnancy rate, and mature oocyte yield.

Results: The patient demographics and clinical characteristics of the dual and hCG trigger groups were similar. In terms of total number of oocyte obtained (p=0.312), oocyte quality (p=0.307), and number of embryos of good quality (p=0.567), there were no significant differences between the two groups. Dual trigger group yielded a significantly higher number/ proportion of mature oocytes (p0.001) than the hCG only group. The rates of clinical pregnancy were higher with dual trigger compared to hCG alone (59.2% versus. 47.1%, p=0.032), while the rates of continuing pregnancy were not significantly different (p=0.054).

Conclusions: More mature oocytes were collected from the dual triggered group. Additionally, compared to the hCG trigger group, the dual trigger group achieved better results in clinical pregnancies following fresh embryo transfers**.**

Key Words: Dual trigger, fresh embryo transfer, hCG trigger, ICSI

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INTRODUCTION

Oocyte maturation is established when the first meiotic division of the oocyte is completed, which ultimately leads to metaphase II oocyte. Resumption of the first stage of meiosis is triggered by a surge of luteinizing hormone (LH)[1].

During controlled ovarian stimulation, analogues of gonadotropin-releasing hormone (GnRH), which can be either an agonist or an antagonist, are used to inhibit pituitary function and prevent premature luteinization. Endogenous FSH and LH production is inhibited by these analogues. Both LH and hCG are members of the same glycoprotein family (similar α- and different β-subunits). Human chorionic gonadotropin, due to its structural similarity to LH, enables luteinisation of granulosa cells, oocyte maturation completion, and meiosis resumption[2].

When the pituitary GnRH receptor is activated by GnRH agonist (GnRHa), a phenomenon known as the fare-up effect occurs, which allows for the acute release of endogenous LH and FSH. During fresh embryo transfer cycles, using GnRHa alone for triggering final oocyte maturation causes severe luteal phase deficiency, this has been accompanied with reduced pregnancy rates and increased risks of early miscarriage, calling for significantly adjusted luteal phase support[3,4].

Adding GnRHa to hCG improves oocyte quality, triggers surges in both LH and FSH, and enhances implantation. Research has demonstrated that this dual trigger method can greatly augment the quantity of transferable and cryopreserved embryos. Furthermore, the ratio of oocytes to preovulatory follicles is improved using this method. Nevertheless, it is still unclear how this strategy will affect the pregnancy rate[5- 7].

This study set out to compare the outcomes of ICSI with fresh embryo transfer (in terms of ovarian stimulation parameters, clinical pregnancy rates, and rates of continued pregnancy) when using dual trigger and hCG only trigger during GnRH antagonist protocol.

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MATERIALS AND METHODS

A randomized controlled trial was conducted at a specialized fertility & gynecology center. From January 2019 to January 2024, all GnRH antagonist ICSI cycles involving patients aged 21–41 were reviewed in this study. Excluded from the study were patients with a predicted poor ovarian response, less than four antral follicles, or those who had severe male factor infertility. The Menoufia University Faculty of Medicine Institutional Review Board gave its approval to the study. Before administering any treatments, we made sure to get a written informed consent from each participant.

Sample size calculation

Based on review of past literature[8] who found that Good-quality embryos was 69.8% and 83.7% in hCG and dual groups respectively. The least sample size calculated using statistics and sample size pro is 158 participants per each group and increase up to 174 participants per each group to avoid 10% dropout rate.

Fresh ICSI cycle

The patient's age, BMI, number of antral follicles, and history of responsiveness to ovarian stimulation were among the factors considered while deciding on the starting dosage of gonadotropin. The GnRH antagonist cetrorelix (Cetrotide®, Merck Serono, Germany) was administered daily at a dose of 0.25 mg (when a follicle's diameter reached 14 mm or greater using transvaginal ultrasonography) and continued until the day that oocyte maturation is triggered.

The random allocation into either HCG only trigger group or dual trigger group was based on a list that was generated by a computer in a 1:1 ratio (174 women in each cohort). Intramuscular injections of 10,000 IU hCG (Choriomon®, IBSA) were given to participants in the hCG only trigger group. Those who were in the dual trigger group were given 0.2 mg of triptorelin subcutaneously (Decapeptyl®, Ferring, Germany) and 5,000 IU of hCG intramuscularly (Choriomon®, IBSA).

Between 35 and 36 hours following the trigger injection, the oocyte retrieval was performed with the use of a transvaginal ultrasonography. The embryos' categorisation was established according to the ESHRE Consensus (2011)[9]. All embryo transfers were carried out with the use of transabdominal ultrasonography as a guide. Luteal phase support was provided, starting the day following oocyte retrieval until a negative pregnancy test or 10th week gestational age.

Outcome measures

Clinical pregnancy rate, defined as the frequency of foetal heartbeats identified between week six and week seven of pregnancy, was the principal outcome measure. The proportion of mature oocytes (relative to the total oocytes recovered) and the rate of ongoing pregnancies were included as secondary outcomes. When a clinical pregnancy goes beyond 12 weeks of gestation, it is called an ongoing pregnancy.

Statistical analysis

We conducted the data analysis using the 2017-released Statistical Tool for the Social Sciences, developed by IBM. Armonk, New York's IBM Corp. To ensure the data was normal, we utilized the Kolmogorov-Smirnov test. Concerning the parametric variables, we could statistically compare the two groups using the Student t test. We utilized the Mann Whitney test to determine if the variables were parametric. To determine if there was a correlation between the two subjective factors, we employed a chi-square test. We considered a *p-value* less than 0.05 on a 95% confidence interval to be statistically significant.

RESULTS

Three hundred seventy candidates were assessed for eligibility to participate in the current study. Twenty two patients were excluded (of these, 13 did not meet the inclusion criteria and 9 declined to participate). So, three hundred forty-eight participants were available for random allocation into two equal groups (174 in hCG only triggered group and 174 in dual triggered group) .All participants completed the trial and ready for analysis, as shown in CONSORT flow chart (Figure 1).

Patients' baseline characteristics and demographics are illustrated in (Table 1). Both groups started out with similar traits; no statistically significant differences were found.

ICSI cycle characteristics are presented in (Table 2). While the dual trigger group had a higher total number of oocytes retrieved (15.56 ± 9.99 vs. 14.39 ± 9.09), this difference did not reach statistical significance (p=0.312). Conversely, there was a significant difference in the number of MII oocytes between the two groups, with the dual trigger group having 14.00 ± 9.32 oocytes compared to the hCG trigger group's 10.71 ± 7.07 oocytes (p<0.001). Because of this, the ratio of mature oocytes to total oocytes retrieved was significantly higher in participants who used the dual trigger protocol (88.16 ± 8.82 vs 72.87 ± 10.12, p<0.001). The percentage of good quality oocytes was similar in both groups (86.2% vs. 81.6%; p=0.307). In terms of the percentage of embryos with good quality, there is no statistically significant difference between the two groups (92.5% vs. 90.2%; p=0.567).

(Table 3) displays the ICSI outcomes for each group. The rates of clinical pregnancy were higher with dual trigger compared to hCG alone (59.2% versus. 47.1%, p=0.032), while the rates of continuing pregnancy were not significantly different (p=0.054).

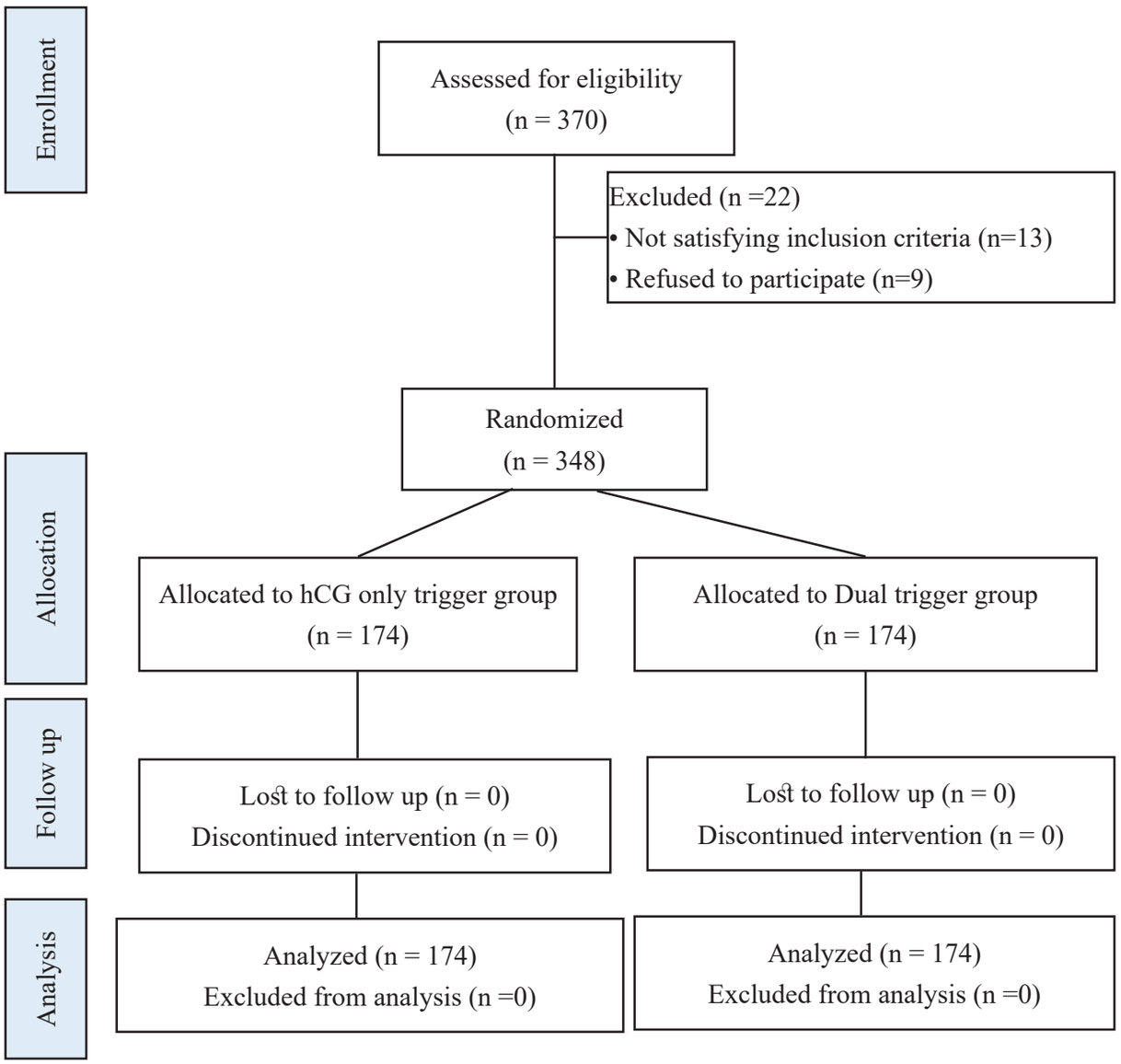

Fig. 1: The CONSORT flow chart

Table 1: Demographic and baseline characteristics of the study groups

	hCG only trigger group (n=174)	Dual trigger group (n=174)	*P value*
Age (years)	29.01 ± 5.69	29.02 ± 5.61	t: 0.028, *p*=0.977
BMI (kg/m^2)	25.01 ± 2.91	25.10 ± 2.93	t: 0.294, *p*=0.769
Mean duration of subfertility (years)	4.33 ± 1.47	4.36 ± 1.45	t: 0.147, *p*=0.883
Causes of subfertility			
Unexplained	71 (40.8%)	70 (40.2%)	
Male factor	26 (14.9%)	27 (15.5%)	
Diminished ovarian reserve	22 (12.6%)	21 (12.1%)	
Mixed	14 (8.0%)	15 (8.6%)	X^2: 0.084, *p*=1.000
PCOS	23 (13.2%)	23 (13.2%)	
Tubal	8 (4.6%)	8 (4.6%)	
Endometriosis	10 (5.7%)	10 (5.7%)	

BMI body mass index; t, Student t-test; χ2, Chi square test

Table 2: ICSI cycle characteristics of the study groups

	hCG only trigger group (n=174)	Dual trigger group (n=174)	*P value*
Duration of ovarian stimulation (Days)	10.63 ± 1.71	10.41 ± 1.62	U=14269.0, *p*=0.347
Total dose of gonadotropins (Iu)	3189.66 ± 513.30	3122.41 ± 484.93	U=14269.0, *p*=0.347
Total number of oocytes retrieved	14.39 ± 9.09	15.56 ± 9.99	U=14191.0, *p*=0.312
Number of MII oocytes	10.71 ± 7.07	14.00 ± 9.32	U=11755, *p*<0.001**
Proportion of mature oocytes	72.87 ± 10.12	88.16 ± 8.82	U=3638.0, *p*<0.001**
Percentage of good quality oocytes	142 (81.6%)	150 (86.2%)	X^2: 1.043, *p*=0.307
Percentage of top quality embryos	157 (90.2%)	161 (92.5%)	X^2: 0.328, *p*=0.567

U, Mann Whitney test; χ2, Chi square test; ** highly significant

Table 3: Outcomes of ICSI cycles

	hCG only trigger group (n=174)	Dual trigger group (n=174)	*P value*
Clinical pregnancy	82 (47.1%)	103 (59.2%)	X^2: 4.616, p=0.032*
Ongoing pregnancy	49 (28.2%)	53 (30.5%)	X^2: 5.820, p=0.054

χ2, Chi square test; *Significant

DISCUSSION

The purpose of this research was to determine whether embryo transfers performed during GnRH-antagonist ICSI cycles resulted in better ovarian stimulation and higher clinical pregnancy rates when GnRHa was added to conventional hCG for triggering the final oocyte maturation.

At the final stage of oocyte maturation, one group of 174 patients received both GnRHa and hCG (a dual trigger), while the other 174 patients received only hCG. The two sets of data did not differ significantly with respect to age, body mass index, or the cause/duration of infertility. The duration of ovarian stimulation and the quantity of gonadotropin required by the two groups were comparable. Contrarily, the data showed that the dual trigger group obtained a significantly higher number of mature oocytes. The percentage of clinical pregnancies was higher with the dual trigger compared to the hCG-only trigger (59.2% vs. 47.1%, *p*=0.032). Regarding oocyte quality (*p*=0.307), total number of oocytes obtained (*p*=0.312), number of good-quality embryos (*p*=0.567), and rates of ongoing pregnancy (*p*=0.054), no significant differences were seen between the groups. Unlike hCG, GnRHa can stimulate the pituitary gland's endogenous production of LH and FSH, which is similar to the menstrual cycle physiology and result in ovulation. One of the main functions of a midcycle FSH surge is to increase the amount of LH receptors in granulosa cells, which improves corpus luteum function. However, the specific implications of this surge are yet unknown[10,11].

According to previous studies, the nuclear maturation process and the expansion of the cumulus-oocyte complex are both facilitated by FSH,[12,13]. Lamb *et al*. observed that patients who underwent hCG in conjunction with bolus FSH had a higher rate of fertilization, suggesting that FSH had a beneficial effect on the IVF outcomes[14].

A lack of enough mature oocytes is the primary factor that leads to in *vitro* fertilization failure. By simulating the body's natural physiology, GnRHa may enhance the percentage of mature oocytes. Grifn *et al*. included 27 patients in total, in their retrospective analysis of stimulated IVF cycles that had previously used the hCG trigger during the antagonist treatment protocol and the dual trigger method was applied in the subsequent cycle. According to the study, over 25% of the oocytes from germinal vesicles and immature oocytes were successfully recovered[15]. The researchers proved that the number and proportion of mature oocytes were significantly increased by the addition of GnRHa to hCG.

Fabris *et al*. evaluated 81 ICSI candidates who had GnRHa added to their HCG in a retrospective study[16]. Metaphase II (MII) oocyte number and ratio were both found to rise when the dual trigger method was employed. Li *et al*. discovered that patients exhibiting a strong ovarian response had a higher success rate in collecting oocytes through the dual trigger technique; however, no details regarding the quantity or ratio of MII oocytes were provided[17].

According to a separate study that included normal responders, dual trigger approach improved the quantity and quality of 2PN embryos, with no observed statistically significant difference regarding the overall oocyte count or the MII oocyte count[18]. Kim *et al*., in a RCT, found no significant differences in the total oocyte yield when comparing the two groups[19]. Additionally, 2 randomized controlled trials (Decleer *et al*. included 120 fresh ICSI cycles and Mahajan *et al*. included 76 fresh ICSI cycles) found no statistically significant difference in the MII oocyte count[20,21]. However, Seval *et al*. found that the dual trigger group had a significantly higher MII oocytes count[5].

We were unable to determine a superiority between the two groups when we compared the number of embryos of high quality. Embryos from the dual trigger group are more likely to be of high quality, according to studies that compared the two methods' effects on embryo quality[5,17,21]. Also, there are reports that suggest this method is not better than hCG alone[18,19,22,23].

GnRH receptors are present in various extra-pituitary organs and tissues, including as the endometrium, myometrium, fallopian tube, ovary, placenta, and fertilised embryos, in addition to the pituitary gland[24–27].

In placental trophoblasts, GnRHa regulates matrix metalloproteinases. The degradation of the extracellular matrix and the invasion of trophoblast cells are both facilitated by matrix metalloproteinases[28]. Compared to patients who have experienced either natural or agonist cycles, those who have undergone antagonist cycles have reduced expression of HOXA-10, a gene that controls endometrial receptivity in endometrial stromal cells[29]. This shift in endometrial receptivity was explained by Devroey *et al*. as a result of the antagonist cycle's low implantation rate[30]. Bukulmez *et al*. compared agonist and antagonist cycles and found that individuals on GnRH antagonist medication had a lower clinical pregnancy rate, regardless of embryo quality[31]. Schachter *et al*. indicated that GnRHa would improve implantation in endometrial cells through the post-receptor impact and thus the dual trigger group had significantly greater rates of clinical and continuing pregnancies[32].

Lin *et al*. had retrospectively investigated 378 fresh embryo transfer cycles and found that dual triggers had a greater pregnancy and live birth rate. However, the rates of miscarriage were not significantly different[23]. Kim *et al*. proved comparable results in a randomized controlled study that analyzed 120 cycles[19]. Another study found that in *vitro* fertilization with dual triggers increased the likelihood of clinical pregnancy and successful implantation[33]. Decleer *et al*.[21] found that the rates of implantation and continuation of pregnancy were similar among the groups.

STRENGTH POINTS

It was a prospective study design with a reasonable sample size. Furthermore, the choice of the trigger for final oocyte maturation was based on the randomization (not by the choice of the treating physician).

LIMITATIONS

It was single center study. Additionally, live birth rates were not investigated in the current trial.

CONCLUSION

After implementing the dual trigger protocol, our investigation showed a statistically significant increase in the number of mature oocytes obtained and their proportion to the total number of oocytes retrieved. In addition, clinical pregnancy outcomes after fresh embryo transfer are positively affected by the dual trigger protocol. But the rates of continued pregnancy seem to be the same as with the hCG-only trigger. To better understand the clinical significance of our improved ICSI cycle outcomes, future larger-scale multicenter trials may be necessary.

CONFLICT OF INTERESTS

There are no conflicts of interest.

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