Original Article

Impact of Intrauterine Administration of Human Chorionic Gonadotropin before Intrauterine Insemination in Infertile Women: A Randomized Controlled Trial

Leena Wadhwa, Anupama Rani

Department of Obstetrics and Gynaecology, IVF and Fertility Research Centre, ESI-PGIMSR, New Delhi, India

Abstract

Background: Implantation is the rate-limiting step in the success of both intrauterine Insemination (IUI) and *in vitro* fertilization cycles. Numerous interventions that target various local signals have been tried to improve the implantation and clinical pregnancy rate (CPR). The most significant of these signals is human chorionic gonadotropin (hCG) which acts as immunomodulator and improves implantation by decidualization of the endometrial stromal cells, trophoblast invasion, proliferation of uterine natural killer cells, stimulation of endometrial angiogenesis, and maintenance of progesterone secretion by the corpus luteum. **Aim:** The aim of the study is to evaluate the effect of intrauterine hCG administration before IUI on CPR. **Settings and Design:** A prospective parallel randomized control study was done from September 2017 to February 2019. **Materials and Methods:** A total of 200 eligible women planned for IUI were randomly divided just before IUI into 2 groups. A computer-generated randomization list with block size of 10 with 1:1 allocation was used to randomize the patients. Experimental group received 0.5 ml containing 500 IU hCG, on the other hand control group received 0.5 ml of normal saline 2–3 min before IUI in single sitting. The main outcomes were CPR, miscarriage rate, and ongoing pregnancy rate. **Statistical Analysis:** It was performed using statistical software version SPSS 17.0. **Results:** Patient's demographic and baseline characteristics were comparable in both the groups. CPR in experimental group was significantly high compared to control group (26% vs. 9%, $P = 0.002$). Ongoing pregnancy rate was also significantly higher in experimental group (23%) compared to control group (7%) ($P = 0.003$). No significant difference in miscarriage rate was seen between the two groups. No cases of ectopic pregnancy, ovarian hyperstimulation syndrome, or multiple pregnancy were reported. **Conclusion:** Intrauterine hCG administration is a simple procedure that can be used to improve pregnancy outcome in IUI cycles.

Clinical Trial Registration Number CTRI/2017/12/010729.

Keywords: *Human chorionic gonadotropin, infertility, intrauterine insemination*

Introduction

Implantation is the rate-limiting step in the success of both intrauterine insemination (IUI) and *in vitro* fertilization (IVF) cycles. Implantation depends on

Address for correspondence: Dr. Anupama Rani, Wz1093/M4, 3rd Floor, Basaidarapur, Ramesh Nagar, New Delhi - 110 015, India.
E-mail: anupamajha09@gmail.com

Received: 01-10-2020 **Revised:** 08-05-2021
Accepted: 09-05-2021 **Published:** 28-06-2021

Access this article online

Quick Response Code:

Website: www.jhrsonline.org

DOI: 10.4103/jhrs.jhrs_196_20

How to cite this article: Wadhwa L, Rani A. Impact of intrauterine administration of human chorionic gonadotropin before intrauterine insemination in infertile women: A randomized controlled trial. J Hum Reprod Sci 2021;14:156-61.

endometrial receptivity, embryo quality, and maternal fetal interface. For the implantation to occur and result in a live birth, blastocyst should hatch, appose, adhere, penetrate, and finally invade a well-synchronized endometrium. It is possible under the influence of ovarian steroid hormones, local autocrine-paracrine signals, and embryo-derived signals.[1-5] All these signals act in a synchronized way during "window of implantation" in which the uterine environment is most receptive to blastocyst implantation.[6,7]

Various interventions have been tried to improve the implantation and thereby clinical pregnancy rate (CPR). These interventions target various local human embryonal signals. The longest known and most significant of these signals is human chorionic gonadotropin (hCG). It is a heterodimeric placental glycoprotein hormone which acts as an early embryonic signal in primates produced by the embryo before its implantation. The direct effect of hCG on human endometrium was first studied by Licht *et al*. in 1998 using an intrauterine microdialysis system that released low concentrations of hCG (500 IU) to the endometrium in the luteal phase.[8-10] They found that intrauterine injection of 500 IU hCG/ml in luteal phase acts as immunomodulator and improves implantation by decidualization of the endometrial stromal cells, trophoblast invasion, proliferation of uterine natural killer (uNK) cells, immunological modulation at the maternal–fetal interface, stimulation of endometrial angiogenesis, and maintenance of progesterone secretion by the corpus luteum.[11-13]

hCG is secreted by embryos before their implantation,[3] beginning from the two-cell stage and also by the endometrial epithelial cells during luteal phase. Embryo with poor morphology is consistent with reduced secretion of hCG which is associated with reduced ability to attract endometrial cells for implantation.[3]

Studies conducted by Mansour *et al*., Santibañez *et al*., and Zarei *et al*. in IVF cycles have shown that intrauterine installation of hCG before cleavage-stage embryo transfer (ET) improves implantation and CPRs.[1,14,15] However, studies conducted by Hong *et al*. and Wirleitner *et al*. showed no beneficial effect of intrauterine administration of hCG when done before blastocyst transfer.[16,17] The reason could be that with the blastocyst transfer, there is not enough time for hCG to have beneficial effect on the endometrium before implantation. To overcome this shortcoming, Navali *et al*. studied the intrauterine administration of hCG after oocyte retrieval and found that it significantly increased the implantation and CPRs in IVF cycles.[18]

IUI is the simplest method of assisted reproductive technology and is one of the most widely used cost-effective treatment options for couples with infertility in developing countries. IUI can be opted in male factor infertility such as ejaculation failure, low sperm count, poor sperm quality, as well as in female factor infertility such as cervical hostility, endometriosis, and unexplained infertility. Since intrauterine hCG has shown some promising results in improving implantation rate in IVF cycles, this study was planned to evaluate the impact of intrauterine hCG before IUI presuming that it will have similar beneficial effect as of intrauterine hCG administration in IVF cycles.

Aims and objective

The aim was to evaluate the impact of intrauterine hCG administration before IUI on CPR, miscarriage rate, and multiple pregnancies rate.

Materials and Methods

This is a parallel, prospective, double-blind randomized controlled trial (RCT) done at a tertiary care center from September 2017 to February 2019. Ethical committee approval was taken before beginning the study, and the study was in compliance with the Declaration of Helsinki. This study has also been registered under Clinical Trial Registration number CTRI/2017/12/010729. Sample size calculation was not performed as there is no previously published work on this intervention in IUI. Hence, all the women planned for IUI were screened regarding eligibility for the study. Informed written consent was taken from all the participants. Infertile women with normal ovarian reserve, <38 years, undergoing IUI cycles with normal thyroid-stimulating hormone, and prolactin were included in the study. Exclusion criteria were tubal blockage, uncontrolled chronic disease, hydrosalpinx, severe endometriosis, and active infection.

A total of 212 subfertile women eligible for the study were selected. Out of 212, two cases declined to participate and 10 cases did not complete the desired investigations so remaining 200 patients underwent natural cycle monitoring or ovulation induction. Trigger was given with 5000 IU of urinary hCG (CHORIOTEC 5000) when the dominant follicle was >18 mm size. IUI was done 36 h after the trigger.

They were randomly divided just before IUI into two groups (Group A and Group B) through block randomization. A computer-generated randomization list with block size of 10 with 1:1 allocation was used to randomize the patients. The treatment allocation was placed in a sealed, opaque envelope and picked up by the staff before IUI. The patient and the doctor were blinded for the type of intervention. Only the staff

giving the prepared samples knew the intervention and the control. Written informed consent was obtained from all the study participants.

Group A (experimental group) received 0.5 ml solution containing 500 IU of hCG, on the other hand, Group B (Control Group) received 0.5 ml normal saline before the IUI cycles. On the day of IUI, 5000 IU of hCG (1 ampule) was dissolved in 2.5 ml sterile water. For each patient in experimental group, 0.25 ml (500 IU) of this solution further diluted with 0.25 ml distilled water (total 0.5 ml) was taken and for the patients in control group, 0.5 ml normal saline was used. IUI catheter (Gynetics) was connected to three-way cannula. One end of which was connected to insulin syringe containing 0.5 ml of hCG 500 IU or normal saline, and the other end was connected to syringe with the semen sample. The doctor injected the solution through an IUI catheter, into the uterine cavity, 1–2 cm above the internal ostium of the cervix and after a gap of 3 min, semen sample was injected, catheter was then slowly withdrawn.

After 2-weeks, urine pregnancy test of the patient was done. Moreover, pregnancy was documented by transvaginal sonography at 6–7 weeks of gestation. The main outcomes were CPR (ultrasound confirmation of a gestational sac), miscarriage rate (follow-up done up to 12 weeks), and ongoing pregnancy rate (calculated by subtracting the miscarriage rate from the pregnancy rate). Results were statistically analyzed.

Results

A flow chart of patient recruitment is shown in Figure 1. Statistical analysis was performed using statistical software version SPSS 17.0. SPSS is a widely used generic statistical tool manufactured by Informer Technologies Inc, 6800 Altamor Drive Los Angeles, CA 90045 United States.

Demographic profile and cycle characteristics were compared; no significant differences were found among the two groups [Table 1]. CPR in experimental Group A receiving 500 IU intrauterine hCG before IUI was significantly high as compared to control Group B receiving 0.5 ml normal saline (26% vs. 9%, $P = 0.002$). Ongoing pregnancy rate was also significantly high in Group A (23%) as compared to Group B (7%) ($P = 0.003$) [Table 2]. No significant difference in miscarriage rate was seen between the two groups. No cases of ectopic pregnancy, ovarian hyperstimulation syndrome (OHSS), or multiple pregnancy were reported in our study.

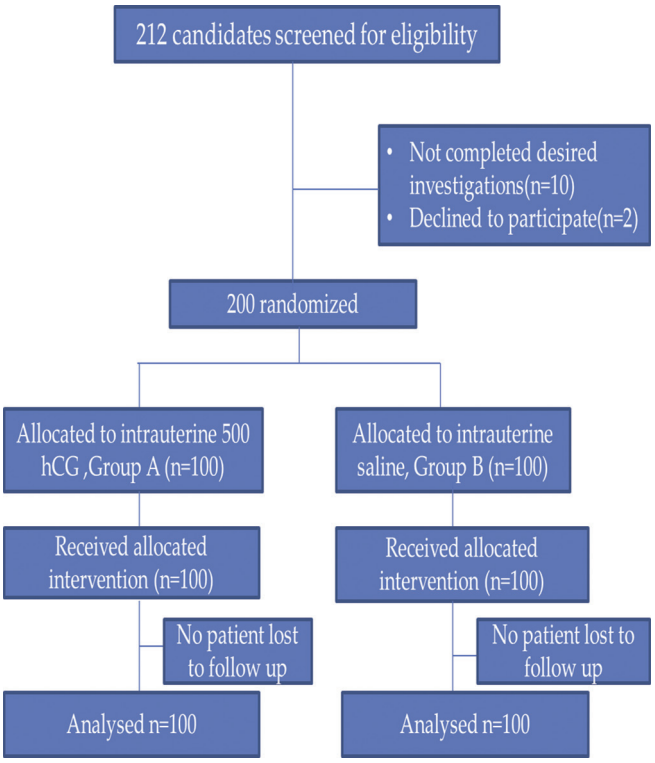

Figure 1: Consort flow diagram

Discussion

The beneficial effect of intrauterine hCG to improve reproductive outcome for women undergoing IVF/intracytoplasm sperm injection (ICSI) cycle is mentioned in multiple studies.[1,9,11,19] This study was done to evaluate the effect of this intervention in non-IVF/ICSI situations like women undergoing IUI presuming that it will have similar beneficial effect as of intrauterine hCG administration in IVF cycles.

In our study, pregnancy rate was higher in first IUI cycles as well as previously failed IUI cycles but the result was significant only in the first cycles ($P = 0.020$). The fecundity rate in the first cycle of IUI is maximum in cases of unexplained infertility and male factor infertility and our majority of patients belong to this group. This might be responsible for significant increase in pregnancy outcome in the first cycle [Table 3].

The ovarian stimulation protocol used in cases was different according to their particular cycle characteristics. The cases were evenly distributed in both the groups according to type of stimulation protocol ($P = 0.223$). CPR was significantly higher in experimental Group A receiving intrauterine hCG (26%) as compared to control Group B (9%) ($P = 0.002$). Ongoing pregnancy rate was also significantly higher in Group A (23%) as compared to Group B (7%) ($P = 0.003$). The reason could be that intrauterine instillation of hCG 500 IU/ml in luteal phase

Table 1: Baseline characteristics of participants in the intrauterine 500 intrauterine insemination human chorionic gonadotropin Group (A) compared with the control Group (B)

	Group A (*n*=100)	Group B (*n*=100)	*P*
Age (years), mean±SD (range)	27.32±4.06 (20-36)	28.04±4.2 (20-38)	0.283
BMI, mean±SD (range)	22.46±2.45	22.65±2.14	0.315
Infertility period (years), mean±SD (range)	6.09±3.95 (1.5-20)	6.54±4.34 (1.5-21)	0.567
Primary infertility (%)	72	75	0.749
Secondary infertility (%)	28	25	0.749
Cause of infertility, *n* (%)			
Male factor	30	24	0.849
Ovulatory dysfunction	11	11	
Tubal	15	13	
Combined	15	17	
Unexplained	29	34	
FSH	6.99±3.31	6.68±2.19	0.969
LH	6.75±3.41	6.42±3.41	0.245
Estriol	48.34±47.32	53.11±49.51	0.493
AMH	4.45±4.19	3.95±3.07	0.336
TSH	2±1.15	2.1±1.07	0.442
AFC on day 2	9.51±3.32	9.02±2.87	0.460
DF>18 mm on day of trigger	1.67±0.78	1.57±0.81	0.230
Endometrial thickness (mm)	9.3±1.73	9.01±1.68	0.276

SD=Standard deviation, BMI=Body mass index, FSH=Follicle-stimulating hormone, LH=Luteinizing hormone, AMH=Anti-müllerian hormone, TSH=Thyroid-stimulating hormone, AFC=Antral follicle count, DF=Dominant follicle

Table 2: Primary and secondary outcomes after intrauterine intervention

	Frequency (%)		OR (95% CI)	*P*
	Group A	Group B		
Primary outcome				
CPR	26 (26)	9 (9)	3.553 (1.568-8.048)	0.002
Secondary outcome				
Miscarriage rate	3 (3)	2 (2)	0.660 (0.108-4.037)	0.653
Ongoing pregnancy rate	23 (23)	7 (7)	3.968 (1.616-9.745)	0.003

CPR=Clinical pregnancy rate, OR=Odds ratio, CI=Confidence interval

Table 3: Effect of cycles of intrauterine insemination on pregnancy outcome after intrauterine intervention

Number of IUI cycles	Pregnancy (%)		*P*
	Group A	Group B	
First cycle	14/63 (22.2)	4/61 (6.5)	0.020
Previous 1 failed cycle	4/22 (18.18)	2/27 (7.4)	0.388
Previous 2 failed cycle	2/7 (28.5)	1/10 (10)	0.537
Previous>2 failed cycles	3/8 (37.5)	0/2 (0.0)	0.533

IUI=Intrauterine insemination

acts as immunomodulator and improves implantation by decidualization of the endometrial stromal cells, trophoblast invasion, proliferation of uNK cells, immunological modulation at the maternal–fetal interface, stimulation of endometrial angiogenesis, and maintenance of progesterone secretion by the corpus luteum.[8,11] There are no documented studies on the use of hCG in IUI cycles.

Most studies compared intrauterine administration of urinary hCG 500 IU versus controls. The rationale use of 500 IU hCG in our study was that previous RCTs with hCG equal to or more than 500 IU has shown improved pregnancy outcome. Furthermore, a recent Cochrane analysis of 17 RCTs with 4751 infertile women by Craciunas *et al*. (2018) investigated the effect of intrauterine administration of hCG in variable doses at different times before the ET for subfertile women undergoing assisted reproduction. The study concluded that there is a moderate quality evidence of increased live birth rate in the subgroup of women undergoing cleavage-stage ET with an intrauterine hCG dose ≥ 500 IU compared to women having cleavage-stage ET without hCG. There was an expected increase of live birth rate ranging from 36% to 51% with previous live birth rate of 27% per cycle before use of intrauterine hCG ≥ 500 IU.[20] The timing of hCG administration in our study was 2–3 days before the physiological hCG secretion by the embryo. The study was designed to mimic the physiological state and to

initiate the cascade of events required for implantation by hCG early so that it is optimized by the time of implantation.

In our study, 500 IU hCG was used in 0.5 ml volume, this volume is greater as compared to studies by Hong *et al*. and Wirleitner *et al*. which used 20–40 µl. A study by Zarei *et al*. and Navali *et al*.[15,18] have used similar volume of intrauterine hCG as used in our study. This volume would reach more endometrial surface area and provoke the pathway of receptivity without effecting IUI outcome.

Intrauterine hCG administration is a simple procedure that can be used to improve pregnancy outcome. It is not expensive and is a cost-effective method, no special equipment is required and can be done with IUI in a single sitting avoiding the stress and discomfort associated with another procedure. This method neither consumes additional time of clinical staff nor does it require complex training. In our study, subfertile women were randomized just before IUI which avoided faulty recruitment of cancelled cycles and increased pregnancy outcome. No patient was lost to follow-up in our study. One of the complications of ovulation induction is ovarian hyperstimulation, but no case of OHSS was reported in our study. No other complications such as ectopic pregnancy or multiple pregnancy were reported in our study. Due to lack of feasibility of long-term follow-up, CPR rather than live birth was used as the outcome measure. This is a limiting factor for this study. Further trials are needed with live birth as the primary outcome, to identify woman who would benefit the most from this intervention. The ovarian stimulation protocol used in cases was different according to their particular cycle characteristics [Table 4]. This is a possible confounding factor, which might affect results. As this is new intervention, long-term follow-up for side effects of the intervention are not available so it should be used with caution. Administration of 0.5 ml of normal saline without hCG into uterine cavity before IUI may also have some effect on implantation. A high pregnancy rate achieved in the study group needs to be replicated in similar studies in future before adapting such intervention into routine clinical practice. Although in our study, it seems to have no effect as pregnancy outcome in control group receiving intrauterine normal saline before IUI is similar to the worldwide reported IUI outcomes.

Table 4: Distribution of cases according to stimulation protocol

Stimulation protocol	Frequency (%)		*P*
	Group A	Group B	
CC	17 (17.0)	15 (15.0)	0.223
CC+gonadotropins	36 (36.0)	30 (30.0)	
Letrozole	19 (19.0)	22 (22.0)	
Letrozole+gonadotropins	18 (18.0)	19 (19.0)	
NCM	10 (10.0)	14 (14.0)	

NCM=Natural cycle monitoring, CC=Clomiphene citrate

Conclusion

Intrauterine hCG administration is a simple procedure that can be used to improve pregnancy outcome in IUI cycles. Further trials are needed with live birth as the primary outcome, to identify woman who would benefit the most from this intervention.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

References

1. Mansour R, Tawab N, Kamal O, El-Faissal Y, Serour A, Aboulghar M, *et al.* Intrauterine injection of human chorionic gonadotropin before embryo transfer significantly improves the implantation and pregnancy rates in *in vitro* fertilization/intracytoplasmic sperm injection: A prospective randomized study. Fertil Steril 2011;96:1370-4.e1.
2. Banerjee P, Fazleabas AT. Extragonadal actions of chorionic gonadotropin. Rev Endocr Metab Disord 2011;12:323-32.
3. Cole LA. Biological functions of hCG and hCG-related molecules. Reprod Biol Endocrinol 2010;8:102.
4. Cole LA. HCG, the wonder of today's science. Reprod Biol Endocrinol 2012;10:24.
5. Diedrich K, Fauser BC, Devroey P, Griesinger G; Evian Annual Reproduction (EVAR) Workshop Group. The role of the endometrium and embryo in human implantation. Hum Reprod Update 2007;13:365-77.
6. Palomino WA, Argandoña F, Azúa R, Kohen P, Devoto L. Complement C3 and decay-accelerating factor expression levels are modulated by human chorionic gonadotropin in endometrial compartments during the implantation window. Reprod Sci 2013;20:1103-10.
7. Racicot KE, Wünsche V, Auerbach B, Aldo P, Silasi M, Mor G. Human chorionic gonadotropin enhances trophoblast-epithelial interaction in an *in vitro* model of human implantation. Reprod Sci 2014;21:1274-80.
8. Licht P, Lösch A, Dittrich R, Neuwinger J, Siebzehnrübl E, Wildt L. Novel insights into human endometrial paracrinology and embryo-maternal communication by intrauterine microdialysis. Hum Reprod Update 1998;4:532-8.
9. Licht P, Fluhr H, Neuwinger J, Wallwiener D, Wildt L. Is human chorionic gonadotropin directly involved in the regulation of human implantation? Mol Cell Endocrinol 2007;269:85-92.
10. Cunningham FG, Leveno KJ, Bloom SL, Hauth JC, Rouse DJ, Spong CY. In: Cunningham FG, Leveno KJ, Bloom SL, Hauth JC, Rouse DJ, Spong CY, editors. Implantation, Embryogenesis, and Placental Development: Secretory or Post Ovulatory Endometrial Phase. USA: McGraw-Hill; 2010. p. 48.
11. Bourdiec A, Shao R, Rao CV, Akoum A. Human chorionic gonadotropin triggers angiogenesis via the modulation of

endometrial stromal cell responsiveness to interleukin 1: A new possible mechanism underlying embryo implantation. Biol Reprod 2012;87:66.

12. Harvey MB, Leco KJ, Arcellana-Panlilio MY, Zhang X, Edwards DR, Schultz GA. Role of growth factors during peri-implantation development. Hum Reprod 1995;10:712-8.
13. Kayisli UA, Selam B, Guzeloglu-Kayisli O, Demir R, Arici A. Human chorionic gonadotropin contributes to maternal immunotolerance and endometrial apoptosis by regulating Fas-Fas ligand system. J Immunol 2003;171:2305-13.
14. Santibañez A, García J, Pashkova O, Colín O, Castellanos G, Sánchez AP, *et al.* Effect of intrauterine injection of human chorionic gonadotropin before embryo transfer on clinical pregnancy rates from *in vitro* fertilisation cycles: A prospective study. Reprod Biol Endocrinol 2014;12:1-5.
15. Zarei A, Parsanezhad ME, Younesi M, Alborzi S, Zolghadri J, Samsami A, *et al.* Intrauterine administration of recombinant human chorionic gonadotropin before embryo transfer on outcome of *in vitro* fertilization/intracytoplasmic sperm injection: A randomized clinical tria. Iran J Reprod Med 2014;12:1-6.
16. Hong KH, Forman EJ, Werner MD, Upham KM, Gumeny CL, Winslow AD, *et al.* Endometrial infusion of human chorionic gonadotropin at the time of blastocyst embryo transfer does not impact clinical outcomes: A randomized, double-blind, placebo-controlled trial. Fertil Steril 2014;102:1591-5.
17. Wirleitner B, Schuff M, Vanderzwalmen P, Stecher A, Okhowat J, Hradecký L, *et al.* Intrauterine administration of human chorionic gonadotropin does not improve pregnancy and life birth rates independently of blastocyst quality: A randomised prospective study. Reprod Biol Endocrinol 2015;13:70.
18. Navali N, Gassemzadeh A, Farzadi L, Abdollahi S, Nouri M, Hamdi K, *et al.* Intrauterine administration of hCG immediately after oocyte retrieval and the outcome of ICSI: A randomized controlled trial. Hum Reprod 2016;31:2520-6.
19. Strug MR, Su R, Young JE, Dodds WG, Shavell VI, Díaz-Gimeno P, *et al.* Intrauterine human chorionic gonadotropin infusion in oocyte donors promotes endometrial synchrony and induction of early decidual markers for stromal survival: A randomized clinical trial. Hum Reprod 2016;31:1552-61.
20. Craciunas L, Tsampras N, Raine-Fenning N, Coomarasamy A. Intrauterine administration of human chorionic gonadotropin (hCG) for subfertile women undergoing assisted reproduction. Cochrane Database Syst Rev 2018;10:CD011537.