Hakimi *et al.*
European Journal of Medical Research (2024) 29:639
https://doi.org/10.1186/s40001-024-02242-3

European Journal of Medical Research

RESEARCH

Open Access

Efficacy of uterine flushing with human chorionic gonadotropin (hCG) on pregnancy rates in primary unexplained infertility: a randomized controlled trial

Parvin Hakimi[1], Mahshid Alborzi[2], Ehsan Nikkhou[1] and Hosein Azizi[1*]

Abstract

Background There are limited and controversial findings concerning ovulation induction using intrauterine and intramuscular human chorionic gonadotropin (hCG) injection compared to intramuscular hCG alone. The study aimed to examine the impact of intrauterine hCG injection, which is used to induce ovulation, on the efficacy of the intrauterine insemination (IUI) technique in patients with unexplained infertility.

Methods A randomized controlled clinical trial was conducted involving 80 subjects with unexplained primary infertility at the infertility clinic of Al-Zahra Hospital in northwest Iran. Patients were randomly allocated into two groups: control and intervention. Both groups received initial treatment with letrozole and Recombinant follicle-stimulating hormone (r-FSH). After confirmation of at least one follicle measuring 18 mm or larger through ultrasonography, in the control group, two ampoules of 5000 units of hCG were administered intramuscularly. The intervention group received 500 units of hCG diluted in 0.5 cc of normal saline and was injected into the uterine cavity along with the two intramuscular ampoules. Primary outcomes were clinical and chemical pregnancy rates and the secondary outcome was any adverse pregnancy outcomes. Multiple logistic regression analysis was used to estimate crude and adjusted odds ratios (AORs) of the pregnancy rates with 95% confidence intervals (CIs).

Results No significant differences were found between the two groups regarding baseline characteristics ($p > 0.05$). Chemical and clinical pregnancy rates in the control and intervention groups were (32.5 vs. 40%) (32.5% vs. 35%), respectively. In the final analysis after adjusting the potential confounders, intrauterine and intramuscular hCG injection increased the likelihood of chemical pregnancy by 1.39 times AOR = 1.42 (1.31–4.12; $p = 0.036$), and clinical pregnancy by AOR = 1.25 (1.03–3.74; $p = 0.048$) compared to intramuscular hCG alone. There were no statistical differences regarding adverse pregnancy outcomes between the study groups (p value > 0.05).

Conclusions It seems that ovulation induction through intrauterine and intramuscular hCG injection increased the odds of both chemical and clinical pregnancy rates compared with intramuscular hCG alone. Multicenter clinical trials and meta-analysis studies are needed for decision making in clinical settings.

Keywords Chorionic gonadotropin, Intrauterine insemination, Pregnancy, Reproductive techniques

*Correspondence:
Hosein Azizi
aziziepid@gmail.com; h.azizi@tbzmed.ac.ir
Full list of author information is available at the end of the article

Background

Unexplained (idiopathic) infertility, defined as the inability of women to conceive after a minimum of 12 cycles of unprotected sexual activity (or six cycles in women over the age of 35) and without apparent cause for the infertility of couples, accounts for 30% of infertile couples globally [1–3]. About 85% of infertility causes can be identified. The most common causes of infertility include ovulation disorders, male factor infertility, and uterine tube diseases. The remaining 15% of infertile couples have "unexplained infertility" [4]. Female infertility is a complex issue that requires attention and practical solutions from governments and organizations globally, especially those handling population issues [5, 6].

One of the costly yet successful treatments for unexplained infertility encompasses a range of assisted reproductive technologies, including in vitro fertilization (IVF) with or without intracytoplasmic sperm injection [7]. In cases of unexplained infertility, IVF is the preferred treatment option when more affordable alternatives, such as medicinal therapy and intrauterine insemination (IUI), have been ineffective. The success rate in IUI is about 15% per treatment cycle, and if fertility treatments do not succeed after 3 to 6 IUI cycles, other treatment options, including IVF, should be considered [8].

IUI can be performed in a natural cycle or in combination with ovarian stimulation using clomiphene citrate, letrozole, or gonadotropins. The purpose of ovarian stimulation in IUI is to enhance the number of dominant follicles in each cycle, thereby boosting pregnancy rates [9, 10]. In IUI cycles, human chorionic gonadotropin (hCG) and gonadotropin-releasing hormone agonist (GnRHa) are often used for follicular maturation and ovulation induction, with hCG being used as a substitute for naturally increasing luteinizing hormone (LH) levels [11, 12]. The direct effects of hCG on the human endometrium were first studied by Mr. Licht in 1998. He discovered that intrauterine injection of low-concentration hCG during the luteal phase is an immunomodulation. It increases embryo implantation by decasualizing the endometrial stromal cells, invading trophoblasts, and proliferation of uterine natural killer (u-NK) cells. In addition, this gonadotropin induces immunological modulation at the embryo–maternal interface by stimulating the angiogenesis of endometrial cells and maintaining progesterone secretion from the corpus luteum, thus enhancing the success rate of implantation [13].

IVF remains an expensive assisted reproductive technology (ART) in certain economically disadvantaged settings. There is limited and contentious evidence concerning ovulation induction with intrauterine and intramuscular hCG injections compared to intramuscular hCG alone [14, 15]. Considering the significant impact of hCG on the physiological ovulation cycle, the aim of this study is to investigate the effect of intrauterine hCG injection on ovulation induction success rates in couples diagnosed with unexplained infertility undergoing IUI.

Methods

Study design and setting

This study is a randomized controlled trial with parallel arms. The study population consisted of couples diagnosed with unexplained primary infertility at the infertility referral clinic of Al-Zahra Hospital at Tabriz University of Medical Sciences, between August 2022 and August 2023.

Participants and study groups

The study had two groups. Patients were randomly assigned into two groups (intervention and control). Both groups were first treated with letrozole and recombinant follicle stimulating hormone (r-FSH). After ultrasonography confirmed the presence of at least one follicle measuring 18 mm or larger, the control group received two ampoules containing 5000 units of hCG intramuscularly. The intervention group received 500 units of hCG diluted in 0.5 cc of normal saline, which was then injected into the uterine cavity with the two intramuscular ampoules. Both groups experienced normal IUIs 34–36 h later.

Eligibility

Inclusion criteria were confirmed primary unexplained infertility [4, 16], women aged between 20 and 38 years, healthy uterine structure and appendages, pelvic examination, ovulation, normal pap smear test results, normal blood test results, normal hormonal profile, and having informed consent. The exclusion criteria were also lack of informed consent to participate, diagnosis of any identifiable cause of infertility, structural abnormalities in the uterus and uterine appendages, uterine infection, moderate to severe endometriosis.

Sample size

According to the previous trial [14], the clinical pregnancy rate was 34% and 26% in the intervention and control groups, respectively. Therefore, considering 5% type I error, 80.0% power, and a 5% increase to account for potential loss to follow-up, 40 cases were selected for each group. In total, 80 primary infertile women were selected.

Randomization and blinding

Participants who met the criteria were allocated to intervention and control groups through balanced block randomization using Stata software version 14. Balanced block randomization prevents an imbalance in

the baseline characteristics and clinical features between study groups. There were 20 blocks, each containing four individuals. An experienced methodologist performed random allocation, and *allocation concealment* was ensured. Figure 1 illustrates the participant flow diagram, depicting the random allocation of participants to study groups, follow-up, and outcome assessment.

The open-ended nature of the intervention made it impossible to blind the clinical staff (infertility fellowship) who administered injections and carried out the IUI process. However, participants and the statistical analyst were blinded to the intervention groups (double-blind).

Measurements

The diagnosis of female infertility was conducted through clinical examinations, laboratory tests, and imaging by an infertility specialist [14, 15]. Women who were unable to conceive after 1 year of unprotected sexual intercourse (or 6 months for women over 35 years) were referred to the infertility clinic for evaluation. A comprehensive medical history was obtained, including information about regular menstrual cycles, medications, and routine examinations such as pelvic examination, pap smear, blood tests, vaginal ultrasonography, hysterosalpingography, spouse's semen analysis, and if necessary,

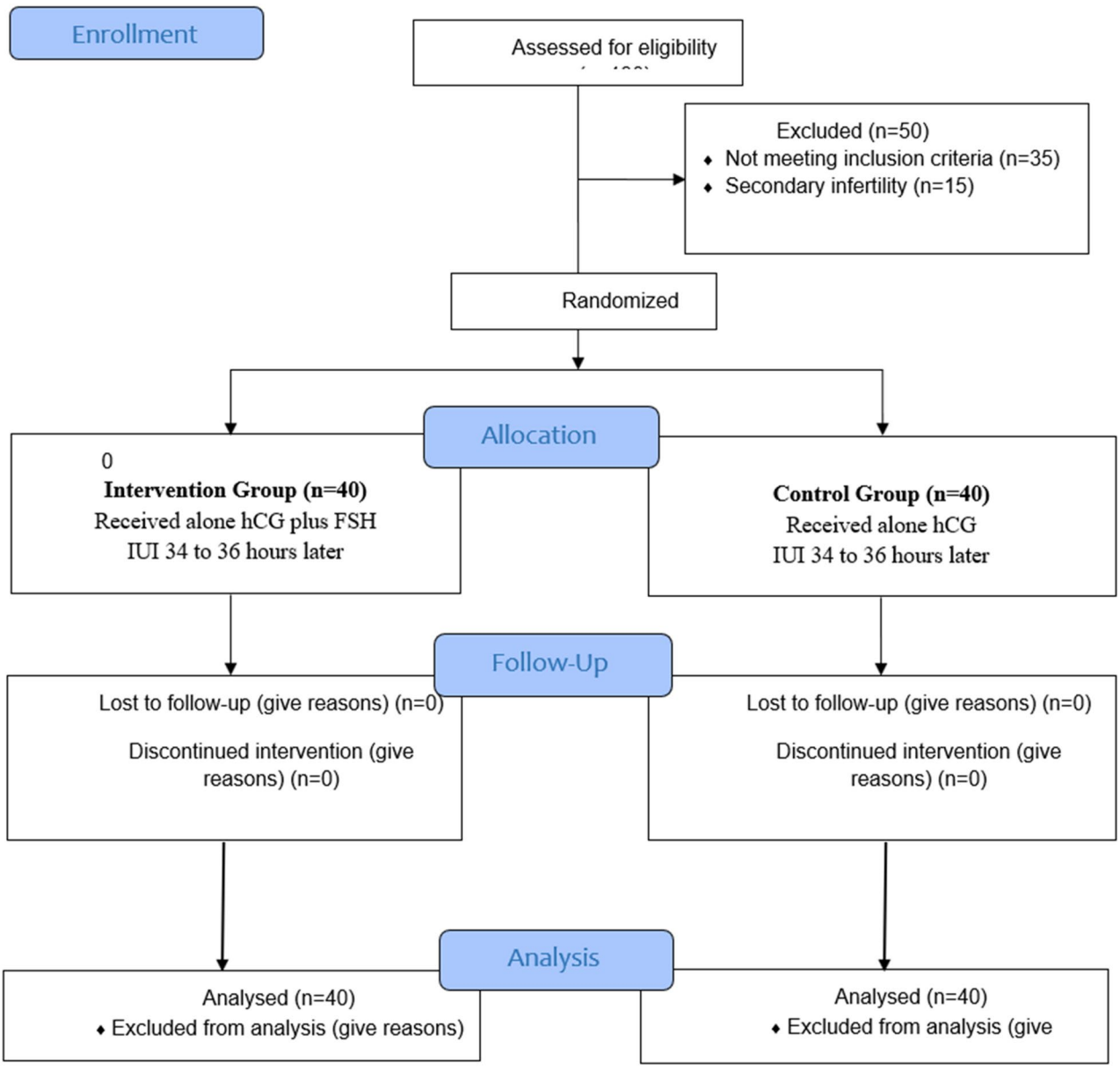

Fig. 1 CONSORT flow diagram of the trial

sonohysterography and hysteroscopy were performed based on the patient's condition. Patients who had been diagnosed with unexplained infertility were subsequently recruited to participate in the research. Before commencing the treatment cycle, it was verified that the patients were indeed infertile. Patient information, including demographic profile, age, pregnancy history, miscarriages, family history of similar problems, underlying diseases of the couple, fertility history, etc., were collected using a checklist.

Transvaginal ultrasound was conducted on the third day of the ovarian cycle and at the onset of menstruation in both the intervention and control groups. For ovulation induction, starting from the third day of menstruation and continuing for 5 days, each patient was prescribed one to two tablets of letrozole 2.5 mg daily (Iran, Arta Pharmed), adjusted according to the age and status of the patient's ovaries. On days 8 and 10 of the cycle, Cinnal-F (r-FSH) ampoules (Iran, CinnaGen), with a dose appropriate to the age and status of the ovaries (75 units subcutaneously), were administered in both groups. To evaluate the ovarian response to the medication, a transvaginal ultrasound was performed again on the 11th and 12th days of the menstrual cycle. Following an evaluation of the ovaries to ensure the presence of a mature follicle measuring 18–20 mm in diameter and endometrium with a minimum thickness of 7 mm, two ampoules containing 5000 units of hCG (Iran, Pooyesh Darou; PD Preg) were administered intramuscularly to both groups. In contrast, 500 units of hCG diluted in 0.5 cc normal saline were administered directly into the uterine cavity of the intervention group, in addition to the 10,000 units of hCG administered intramuscularly. IUI was performed in a standardized and identical approach on both groups approximately 34–36 h following medication injection. Both interventions were performed with the same duration and similar process.

Outcomes

Primary outcomes were clinical and chemical pregnancy rates in the study groups. The secondary outcome was adverse pregnancy outcomes including abortion, anomaly, term delivery, ovarian hyperstimulation syndrome (OHSS), and multiple pregnancies.

Statistical analysis

The data were analyzed using Stata version 14 software. Descriptive statistical measures such as percentage, frequency, mean, and standard deviation were used to describe the data. The Shapiro–Wilk test was employed to assess the normality of the data. The independent t test (for parametric variables) or the Mann–Whitney test (for non-parametric variables) was used to compare quantitative variables between the two groups including age, body mass index (BMI), infertility duration, total sperm count, and sperm mobility. Moreover, the Chi-squared test was utilized to test qualitative or categorized variables such as chemical and clinical pregnancies between the two groups. Fisher's exact test was used when the expected frequency in any cell of a 2×2 table was less than 5. Univariate analysis was carried out using simple logistic regression analysis. Then all significant variables and/or p values <0.2 entered multiple logistic regression analysis to estimate crude and adjusted odds ratios (ORs) of the pregnancy rates with 95% confidence intervals (CIs) [17, 18]. A significance level of less than 5% was considered for all tests.

Results

A total of 80 women with unexplained infertility (40 in each group) were enrolled; all participated until the study's conclusion, with no sample dropout. Table 1 shows the demographic and baseline characteristics of the participants. The mean age ± standard deviation of the participants in the control group was 31.46 ± 4.44 years, and in the intervention group was 30.95 ± 6.15 years. The mean ± standard deviation of body mass index (BMI) in the control group was 24.15 ± 2.12, and in the intervention group was 25.34 ± 4.89, with no statistically significant difference between the two groups regarding age and BMI ($p>0.05$). Furthermore, no statistically significant difference was observed between the two groups regarding other demographic characteristics such as education level and occupation ($p>0.05$). There was no significant difference between the intervention and control groups in terms of family history of infertility and duration of infertility (years) ($p>0.05$).

Table 2 demonstrates the results of multiple logistic regression analysis to estimate crude and adjusted odds ratios (AOR) with 95% confidence intervals (CIs) for pregnancy outcomes after adjusting for potential confounders. Comparing the sperm analysis parameters of participants showed a statistically significant difference in sperm count between the two groups ($p=0.002$). However, no significant relationship was observed between the two groups in other sperm analysis parameters, such as sperm motility and sperm with normal morphology ($p>0.05$). The mean ± standard deviation of the number of follicles before the intervention was 3.30 ± 1.41 in the control group and 1.88 ± 0.72 in the intervention group, which was statistically significant ($p=0.001$). The mean ± standard deviation of endometrial thickness in the control group was 7.40 ± 1.44 mm, and in the intervention group, it was 7.12 ± 1.19 mm, indicating no significant relationship between the two groups ($p>0.05$).

Table 1 Demographic and baseline characteristics of the study participants (before the intervention)

Variable		Control	Intervention	OR; 95% CI	*p* value
		Intramuscular hCG ($n=40$)	Intrauterine and intramuscular hCG ($n=40$)		
Age (year)*		31.4 ± 4.4	30.95 ± 6.15	0.98 (0.90–1.06)	0.649
Body mass index (BMI)*		24.15 ± 2.12	25.3 ± 4.8	1.07 (0.93–1.24)	0.307
Education level	Illiterate	1 (1.6)	2 (2.4)	1.13 (0.91–1.40)	0.258
	Elementary	3 (8.2)	6 (16.7)		
	Middle school and high school	16 (41)	20 (50)		
	University	20 (49.2)	12 (31)		
Employment status	Housewife	24	29	1.14 (0.94–1.37)	0.654
	Employed	16	11		
Familial infertility	Yes	8 (21.3)	6 (16.7)	1.35 (0.49–3.7)	0.558
	No	32 (78.7)	34 (83.3)		
Infertility duration (years)**		5.26 ± 2.8	4.4 ± 2.5	0.85 (0.73–1.05)	0.340

hCG: human chorionic gonadotropin; OR: odds ratio; CI: confidence interval

*Independent *t* test

**Mann–Whitney *U* test

Table 2 Results of multiple logistic regression analysis to estimate crude and adjusted odds ratios (AOR) and 95% confidence intervals (CLs) for pregnancy outcomes in the study groups

Variables		Groups ($n=80$)		Crude OR; 95% CI *p* value	Adjusted OR; 95% CI *p* value
		Control ($n=40$)	Intervention ($n=40$)		
Total sperm count (million)		113.8 ± 109	22.6 ± 57.2	0.989 (0.98–0.99) 0.002	0.88 (0.72–0.95) 0.037
Sperm motility (million)		87.6 ± 95.1	73.08 ± 13.7	0.99 (0.98–1.03) 0.223	1.02 (0.93–1.41) 0.745
Normal sperm morphology (percentage)		9.8 ± 1.3	8.5 ± 1.4	0.75 (0.52–1.07) 0.114	0.96 (0.81–1.82) 0.398
Number of follicles		3.3 ± 1.4	1.8 ± 0.7	0.24 (0.12–0.48) 0.001	0.57 (0.39–0.88) 0.014
Endometrial thickness (mm)		7.4 ± 1.4	7.1 ± 1.2	0.85 (0.61–1.17) 0.333	0.98 (0.84–1.53) 0.512
Chemical pregnancy	Yes	13 (32.5)	16 (40.0)	1.39 (1.11–3.83) 0.041	1.42 (1.31–4.12) 0.036
	No	27 (67.5)	24 (60.0)	Reference	1
Clinical pregnancy	Yes	13 (32.5)	15 (37.5)	1.2 (1.0–3.13) 0.051	1.25 (1.03–3.74) 0.048
	No	27 (67.5)	25 (62.5)	Reference	1
Number of gestational sacs		0.34 ± 0.51	0.36 ± 0.53	1.06 (0.48–2.29) 0.891	1.11 (0.73–1.85) 0.866

OR: odds ratio; CI: confidence interval

More details of crude and adjusted odds ratios (AORs) for each variable are presented in Table 2.

The proportion of chemical and clinical pregnancy in the intervention group was 40% and 37.5%, respectively. In contrast, these values for the control group for both types of pregnancy were reported as 32.5%. In the final analysis, the adjusted odds ratio (AOR) of chemical and clinical pregnancy in the intervention group was 1.42 and 1.25, respectively. The intervention increased the likelihood of chemical pregnancy by 1.42 times AOR = 1.42 (1.31–4.12; $p=0.036$), and clinical pregnancy by AOR = 1.25 (1.03–3.74; $p=0.048$). Similarly, the number of gestational sacs did not significantly differ between the two groups ($p>0.05$).

Table 3 compares adverse pregnancy outcomes in the study groups. The frequency of anomalies was two in the control group and none in the case group, with no significant difference (OR = 0.31; 95% CI 0.05–4.19). Likewise, the number of abortions was 2 and 3 in the control and case groups, respectively. No significant difference was found (OR = 0.65; 95% CI 0.06–6.03). Term delivery in the case group was greater than in the control group (35% vs. 22.5%). However, this difference was not significant (OR = 1.85; 95% CI 0.62–5.6). There were no documented instances of OHSS in either of the groups. Details of adverse pregnancy outcomes are presented in Table 3.

Discussion

The present study aimed to evaluate the impact of intramuscular and intrauterine injection of hCG for ovulation induction on the success rate of IUI in couples diagnosed with unexplained infertility, comparing this method to intramuscular hCG injection alone. In the final analysis, after adjusting for potential confounders, it was found that ovulation induction in IUI with intrauterine hCG injection resulted in a significantly higher likelihood of chemical pregnancy rates (40% compared to 32.5%) and clinical pregnancy rates (37.5% vs. 32.5%) than in the control group.. The practical implications of this study can be especially remarkable and a priority choice for physicians who eagerly seek to increase the chances of assisted reproductive technology for their patients. This issue is particularly significant in contexts, where families cannot afford costly assisted reproductive methods from a financial perspective [19].

It may be argued that IUI with intramuscular–intrauterine hCG injection could be considered a suitable and cost-effective alternative to IVF for individuals with unexplained infertility. Craciunas and Tsampras injected r-hCG during the serum LH surge to induce ovulation. They found a significant increase in fertility rates and follicular growth capability with r-hCG injection compared to conventional hCG alone, similar to our findings [20].

Agrawal et al. (2018) conducted a study examining 624 ovarian cycles. They found that injecting hCG approximately 36–40 h before IUI, compared to a longer time interval of more than 40 h between the two, was associated with increased fertility rates and success in biochemical pregnancy [21]. These findings align with the results of the present study.

Leena Wadhwa and Anupama Rani conducted a study on 200 infertile women to investigate the effect of intrauterine hCG on fertility rates. The intervention group received 500 units of diluted hCG in normal saline intrauterinely. The results showed that intrauterine hCG increased the pregnancy success rate by 26% in the intervention group, whereas the fertility rate in the control group was only 9% [22]. The objectives of their study are similar and aligned with the goals of this research. Studies by Mansour on IVF cycles indicated that injecting intrauterine hCG before embryo transfer significantly increases implantation rates and fertility [23]. On the other hand, Hong and Wirleitner did not find the beneficial effects of intrauterine hCG before blastocyst transfer in IVF [24], which aligns with the results of this study.

Wan and Sheng (2020) found that hCG for ovulation induction resulted in an increased pregnancy rate in IUI with donor sperm in a natural ovarian cycle [25]. Firouzabadi and Janati divided 159 infertile women into three groups. One group received 500 units of hCG intrauterine, the second 1000 units, and individuals in the third group, serving as the control group, did not receive any medication. They found no significant difference among these three groups regarding increasing pregnancy rates [26]. These findings are contrary to the results of the mentioned study. The reasons for such differences between studies, factors such as the study design and method, dose of administration, ethnicity, sample size, and control source can be the causes of the possible difference in the results.

The high biological availability of hCG has led to its use in artificial insemination for the final maturation of oocytes instead of LH. Evidence has shown that intrauterine injection of low-concentration hCG during the luteal phase is an immunomodulator. It increases embryo implantation by decidualizing the endometrial stromal cells, invading trophoblasts, and proliferation of uterine natural killer (u-NK) cells. In addition, this gonadotropin induces immunological modulation at the embryo–maternal interface by stimulating the angiogenesis of

Table 3 Comparison of adverse pregnancy outcomes between the study groups

Pregnancy outcome		Groups		OR; 95% CI	*p* value
		Control (*n* = 40)	Intervention (*n* = 40)		
Anomaly*	Yes	2 (5.0)	0 (0.0)	0.31 (0.05–4.19)	0.152
	No	38 (95)	40 (100)		
Abortion*	Yes	3 (7.5)	2 (5.0)	0.65 (0.06–6.03)	0.644
	No	37 (92.5)	38 (95.0)		
Term delivery*	Yes	9 (22.5)	14 (35)	1.85 (0.62–5.6)	0.216
	No	31 (77.5)	26 (65.0)		
OHSS*	Yes	0 (0.0)	0 (0.0)	0.99 (0.12–8.5)	0.988
	No	40 (100.0)	40 (100.0)		

OR: odds ratio; CI: confidence interval; hCG: human chorionic gonadotropin; OHSS: ovarian hyperstimulation hormone

*Fisher's exact test

endometrial cells and maintaining progesterone secretion from the corpus luteum, thus enhancing the success rate of implantation [13].

There was no significant difference between the two groups in terms of adverse pregnancy outcomes such as various anomalies, preterm birth, miscarriage, and OHSS. In addition, some negative consequences, such as anomalies and miscarriages, had fewer occurrences in the intervention group. Therefore, it can be said that, at least in the present study, the intervention did not lead to adverse pregnancy outcomes.

Limitations and strengthens

Although this trial identified a significant association with an increased likelihood of chemical and clinical pregnancies, the study had several limitations. The first concern was the potential confounding factors that could influence the study interventions. This issue might distort the true effect size of the intervention group compared to the control group. To address this, the study groups were randomly assigned, and the patients were blinded to their group allocations. Second, the study carried out multiple logistic regression analysis to estimate crude and adjusted odds ratios (AOR) with 95% confidence intervals after accounting for the potential confounders.

The next issue was the absence of patient satisfaction measurement in the study groups. However, given that the intervention methods in both groups were non-invasive and similar, it was not feasible to discern any difference for the patients. Therefore, we believe that patient satisfaction in the two groups is likely the same. This issue will serve as a starting point for future studies.

The target group of the present study was patients with unexplained infertility. We do not know how effective it is for other types of infertility. This trial found significant associations, it indicated that the study's power is sufficient. However, larger trials with multicenter patients are needed to provide reliable results.

Conclusion

It seems that ovulation induction through intrauterine and intramuscular hCG injection increased the odds of chemical and clinical pregnancy rates compared with intramuscular hCG alone. Multicenter clinical trials and meta-analysis studies are needed for decision making in clinical settings.

However, findings may be of utmost importance, especially to clinicians who are actively trying to improve their patient's likelihood of becoming fertile (despite having low financial means) using assisted reproductive techniques.

Abbreviations

AOR	Adjusted odds ratio
ART	Assisted reproductive technology
BMI	Body mass index
CI	Confidence interval
IUI	Intrauterine insemination
IVF	In vitro fertilization
OHSS	Ovarian hyperstimulation hormone
OR	Odds ratio
hCG	Human chorionic gonadotropin
LH	Luteinizing hormone
r-FSH	Recombinant follicle stimulating hormone
u-NK	Uterine natural killer

Acknowledgements

The authors express gratitude for the statistical support and epidemiological consultation provided by the Clinical Research Development Unit of Al-Zahra Hospital, Tabriz University of Medical Sciences. This study is derived from Ehsan Nikkhoo's doctoral dissertation.

Author contributions

PH, HA: Developed the original idea, protocol development and interpretation, data analysis, and data collection and drafted all sections of the manuscript. Data collection, data extraction, contributed to the development of the protocol: MA and EN. All the authors approved the final manuscript.

Funding

The study was funded by Tabriz University of Medical Sciences.

Availability of data and materials

The datasets generated and/or analysed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

The study protocol was registered and confirmed in the Iranian Registry of Clinical Trials under the number (IRCT20220702055335N1). The present study has been approved by the Ethics Committee of Tabriz University of Medical Sciences with the approval number (IR.TBZMED.REC.1401.233). Written informed consent was obtained from the participants, granting them the right to participate or withdraw at any research stage. The research groups were assured of the confidentiality of their information. Integrity and honesty were maintained during data collection, review of available sources, etc. Upon request, research results will be made available to health centers. At the beginning of the participant's entry into the study, the researcher's contact number was provided so that they could receive necessary guidance in case of any problems or questions.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

Author details

[1]Women's Reproductive Health Research Center, Tabriz University of Medical Sciences, Tabriz, Iran. [2]Department of Infertility Center, Jahrom University of Medical Sciences, Jahrom, Iran.

Received: 14 September 2024 Accepted: 19 December 2024
Published online: 31 December 2024

References

1. Romualdi D, Ata B, Bhattacharya S, Bosch E, Costello M, Gersak K, Homburg R, Mincheva M, Norman R. Evidence-based guideline: unexplained infertility. Hum Reprod. 2023;38(10):1881–90.
2. Esmaeilzadeh S, Delavar MA, Zeinalzadeh M. Mir M-RA: epidemiology of infertility: a population-based study in Babol, Iran. Women Health. 2012;52(8):744–54.

3. Ling L, Xia D, Jin Y, Hong R, Wang J, Liang Y. Effect of follicle size on pregnancy outcomes in patients undergoing first letrozole-intrauterine insemination. Eur J Med Res. 2024;29(1):184.
4. Carson SA, Kallen AN. Diagnosis and management of infertility: a review. JAMA. 2021;326(1):65–76.
5. Ombelet W, Cooke I, Dyer S, Serour G, Devroey P. Infertility and the provision of infertility medical services in developing countries. Hum Reprod Update. 2008;14(6):605–21.
6. Thoma M, Fledderjohann J, Cox C, Adageba RK. Biological and social aspects of human infertility: a global perspective. In: Oxford research encyclopedia of global public health. edn.; 2021.
7. Van Loendersloot L, Van Wely M, Limpens J, Bossuyt P, Repping S, Van Der Veen F. Predictive factors in in vitro fertilization (IVF): a systematic review and meta-analysis. Hum Reprod Update. 2010;16(6):577–89.
8. Lemmens L, Kos S, Beijer C, Braat D, Nelen W, Wetzels A. Laboratories ssotDFfQAiM: techniques used for IUI: is it time for a change? Hum Reprod. 2017;32(9):1835–45.
9. Danhof N, Wang R, Van Wely M, Van Der Veen F, Mol B, Mochtar M. IUI for unexplained infertility—a network meta-analysis. Hum Reprod Update. 2020;26(1):1–15.
10. Zhao M, Huan Q, Huang L, Yang L, Dong M. Pregnancy outcomes of intrauterine insemination in young patients with diminished ovarian reserve: a multicenter cohort study. Eur J Med Res. 2023;28(1):402.
11. Le MT, Nguyen DN, Zolton J, Nguyen VQH, Truong QV, Cao NT, Decherney A, Hill MJ. GnRH agonist versus hCG trigger in ovulation induction with intrauterine insemination: a randomized controlled trial. Int J Endocrinol. 2019;2019:1–6.
12. Santi D, Casarini L, Alviggi C, Simoni M. Efficacy of follicle-stimulating hormone (FSH) alone, FSH+ luteinizing hormone, human menopausal gonadotropin or FSH+ human chorionic gonadotropin on assisted reproductive technology outcomes in the "personalized" medicine era: a meta-analysis. Front Endocrinol. 2017;8:114.
13. 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(5):532–8.
14. Abdallah KS, Makhlouf A, Badran E, El-Nashar IM, Al-Hussaini TK, Farghaly T, Mohamed HS, Mol BW, Abdelmagied AM. Intrauterine injection of HCG before embryo transfer: a parallel, double-blind randomized trial. Reprod Biomed Online. 2021;43(4):663–9.
15. Aydin Y, Hassa H, Oge T, Tokgoz VY. A randomized study of simultaneous hCG administration with intrauterine insemination in stimulated cycles. Eur J Obstet Gynecol Reprod Biol. 2013;170(2):444–8.
16. Aziz MU, Anwar S, Mahmood S. Hysterosalpingographic evaluation of primary and secondary infertility. Pak J Med Sci. 2015;31(5):1188.
17. Fakhari A, Allahverdipour H, Esmaeili ED, Chattu VK, Salehiniya H, Azizi H. Early marriage, stressful life events and risk of suicide and suicide attempt: a case–control study in Iran. BMC Psychiatry. 2022;22(1):71.
18. Esmaeili ED, Fakhari A, Naghili B, Khodamoradi F, Azizi H. Case fatality and mortality rates, socio-demographic profile, and clinical features of COVID-19 in the elderly population: A population-based registry study in Iran. J Med Virol. 2022;94(5):2126–32. https://doi.org/10.1002/jmv.27594.
19. Souza MCB, Silva LAB, Sequeira FF, Azevedo Antunes R, Souza MM. The management of infertility for primary care physicians. Women Health. 2023;63(3):194–203.
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. https://doi.org/10.1002/14651858.CD011537.pub3.
21. Agrawal S, Das V, Agarwal A, Pandey A. Decoding the effect of time interval between hCG and IUI and sperm preparation and IUI. Int J Reprod Contracept Obstet Gynecol. 2018;7(3):892–7.
22. 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(2):156.
23. Mansour R, Tawab N, Kamal O, El-Faissal Y, Serour A, Aboulghar M, Serour G. 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(6):1370–4.
24. Hong KH, Forman EJ, Werner MD, Upham KM, Gumeny CL, Winslow AD, Kim TJ, Scott RT Jr. 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(6):1591–5.
25. Wan J-P, Wang Z-J, Sheng Y, Chen W, Guo Q-Q, Xu J, Fan H-R, Sun M. Effect of HCG-triggered ovulation on pregnancy outcomes in intrauterine insemination: an analysis of 5,610 first IUI natural cycles with donor sperm in China. Front Endocrinol. 2020;11:423.
26. Firouzabadi RD, Janati S, Razi MH. The effect of intrauterine human chorionic gonadotropin injection before embryo transfer on the implantation and pregnancy rate in infertile patients: a randomized clinical trial. Int J Reprod BioMedicine. 2016;14(10):657.

Publisher's Note