



Role of endometrial scratching in IVF- ET cycles in women with recurrent implantation failure

Authors

Avani Pillai¹, Fessylouis T^{2*}, Ramesh P³, Aparna N⁴

¹Assistant Professor, Department of Reproductive Medicine and Surgery, Amrita Institute of Medical Sciences, Kochi, Kerala

²Associate Professor, Dept of Reproductive Medicine and Surgery, Amrita Institute of Medical Sciences, Kochi

^{3,4}MCh Resident, Department of Reproductive Medicine and Surgery, Amrita Institute of Medical Sciences, Kochi

*Corresponding Author

Dr Fessylouis T

Associate Professor, Dept of Reproductive Medicine and Surgery, Amrita Institute of Medical Sciences, Kochi, India

Abstract

Introduction: Developing a receptive endometrium for implantation of the embryo during In vitro fertilization (IVF) treatment is considered the rate-limiting step for its success. Recurrent implantation failure (RIF) has always been a difficult situation to deal with. Our Objective was to evaluate the role of hysteroscopic endometrial scratching in improving the implantation rate and the pregnancy outcome in IVF-ET (embryo transfer) cycles in women with RIF.

Materials and Methods: We did a prospective study including 58 patients with RIF and sonologically normal endometrium, who underwent IVF-ET between September 2016 and August 2018. Of these 32 women underwent hysteroscopic endometrial scratching in the previous cycle whereas 26 of them did not undergo the procedure. The implantation rate, overall pregnancy rate, clinical pregnancy rate and live birth rate were compared between the two groups.

Results: The implantation rate in patients who underwent hysteroscopic endometrial scratching prior to IVF-ET was comparable to that in the subjects who did not undergo the procedure (18.8% versus 13.2%, $p=0.569$). The overall pregnancy rate (40.6% versus 38.5%, $p=0.87$), clinical pregnancy rate (37.5% versus 34.6%, $p=0.82$) and live birth rate (21.9% versus 19.2%, $p=0.80$) were also comparable between the two groups.

Conclusion: Our study did not show statistically significant improvement in the pregnancy outcome after hysteroscopic endometrial injury in IVF-ET cycles. Hysteroscopic endometrial injury in IVF -ET cycles in women with recurrent implantation failure may not be beneficial in improving the pregnancy outcome.

Keywords: Endometrial scratching, Pre – IVF hysteroscopy, Recurrent implantation failure, Endometrial receptivity, IVF embryo transfer, Endometrial injury.

Introduction

Assisted Reproductive technique has become the treatment of choice in indicated cases of female infertility and male sterility. But despite advances in the procedures of In vitro fertilization (IVF) technology, the clinical pregnancy rate has not substantially improved over the last ten years (currently only 32.4~33.0% per IVF transfer as

reported by ESHRE in 2010),¹ and many patients suffer repeated implantation failure even in the most successful *in vitro* fertilization (IVF) clinics.

Implantation of the embryo represents the most critical step of the reproductive process. It is a complex and multistage process which may be one of the causes that may explain outcome failures.² In spite of repeated transfers of embryos with good

morphology into a normal uterus in IVF cycles, implantation may fail. Consequently, a successful implantation depends not only on quality of embryo but also on the acquisition of endometrial receptivity. Thus, developing a receptive endometrium, which is a limiting step for success in IVF- ET (embryo transfer), is at upmost importance for a successful implantation and IVF outcome.^{2,3} Of the various methods evaluated to improve endometrial receptivity, local endometrial injury gaining a lot of recent attention.

There are various proposed hypothesis as to how endometrial injury improves endometrial receptivity. The first hypothesis states that local injury to the endometrium induces endometrial decidualization, which increases the probability of implantation of are placed embryo.⁴ This hypothesis is based on the observation of induction of decidual tissue formation which mimics the endometrial changes of early pregnancy after mechanical endometrial stimulation with a microcurette in guinea pigs.⁵ Second mechanism is the wound healing. Endometrial injury might provoke the wound healing, involving a massive secretion of different cytokines and growth factors. Gnainsky et al.⁶ noted that local injury of the endometrium induced an inflammatory response resulting in increase in the amount of uterine dendritic cells and macrophages (HLA-DR+ CD11c+ cells), thus improving endometrial receptivity and promoting successful implantation. The third mechanism is that endometrial maturation is abnormally advanced when controlled ovarian stimulation is performed during ART.^{7,8} Zhou et al.⁹ postulated that local endometrial injury in stimulated cycle delays the endometrial development because of the wound repair processes correcting the asynchrony between endometrial and embryo stage. The last mechanism is endometrial gene modulation. Kalma et al.¹⁰ reported that the expression of 183 genes increased 2- to 10-fold and the expression of 39 genes was down regulated at least two fold in biopsy-treated patients. Song et al.¹¹ noted that in women lacking PLA2, the initiation of implantation was deferred, shifting the normal window of implantation. MUC1

represents a potential ligand for selectins that are known to be expressed by human blastocysts, and which may have an important role in the adhesion of the blastocyst to the endometrium.¹²

Various methods of endometrial injury have been mentioned in the literature including endometrial biopsy, curettage, hysteroscopic injury/scratching. Of these hysteroscopic endometrial injury seems most promising. Hysteroscopic endometrial injury prior to embryo transfer has been suggested as a means to improve the implantation rate and hence the clinical pregnancy rate in patients with recurrent implantation failure undergoing IVF treatment.

Our objective was to evaluate the role of hysteroscopic endometrial scratching in improving the implantation rate and overall pregnancy outcome in IVF-ET cycles in women with recurrent implantation failure.

Material and Methods

This study was a 2 years prospective cohort study carried out at a tertiary care centre between September 2016 and August 2018. A total of 58 patients with recurrent implantation failure and sonologically normal endometrium who underwent IVF embryo transfer at the centre were included in the study. We included women aged less than 38 years with history of two or more previous implantation failure (no pregnancy after transfer of at least one good quality embryo), undergoing IVF-ET at our centre. Those subjects with poor endometrial thickness (<7mm on day of HCG trigger) and those who had any abnormality evident in ultrasonography were excluded. Of the patients included in the study, 32 women underwent hysteroscopic endometrial scratching in the previous cycle whereas 26 of them did not undergo the procedure.

Demographic characteristics of the couples were recorded. Clinical history including the details of previous IVF implantation failures were noted. Routine transvaginal ultrasound for the female and semen analysis for the male partner was carried out. In patients undergoing hysteroscopy, it was performed in the luteal phase of the cycle. Rigid

hysteroscope (continuous flow, 30 degree forward oblique view) assembled in a 4mm diameter diagnostic sheath with an atraumatic tip (Karl Storz Endoscopy, Germany) was used. An isotonic solution (0.9% Normal saline) administered via a pressure control pump (hysteromat) was used. The pressure was preset between 80-120 mmHg with the aim to use the lowest pressure required to distend the uterine cavity adequately. Procedure was performed after taking informed consent from the women and under antibiotic prophylaxis. Entry into the uterine cavity was under direct vision using gentle manipulation with tenaculum/vulsellum applied to the anterior lip of the cervix to help straighten the cervical canal whenever required.

Long protocol for IVF stimulation was used. Patients were down-regulated with a gonadotrophin-releasing hormone (GnRH) agonist (single dose lupride depot 3.75mg IM or lupride acetate 0.5mg daily injections) in the mid-luteal phase (days 18-21) of the previous cycle. After down regulation was achieved, ovarian stimulation was commenced from cycle day 2-3 with Gonadotropins (recombinant FSH and/or human menopausal gonadotropin [hMG]) at a dose of 150-300 IU daily according to age, antral follicle count, BMI and response in previous cycles; for first 7 days. Thereafter the dose was adapted according to the ovarian response to treatment. The GnRH agonist was continued upto and including the day of administration of HCG. The final oocyte maturation was achieved with 10,000 IU of HCG (human chorionic gonadotropin) when 2 or more follicles reached a diameter of \geq 17mm. Oocyte retrieval was performed 36 hours after HCG administration. After oocyte retrieval, ICSI was performed. Embryo transfer was performed on day 2 or 3 after oocyte retrieval. The luteal phase was supplemented with intramuscular or intravaginal progestones. A quantitative analysis of serum β HCG concentrations was conducted 14 days after embryo transfer. A cut-off level of 50mIU/ml was used to confirm pregnancy. Subsequently, ultrasound was performed after 6 weeks of gestation to confirm number of sacs

implanted and viability. These pregnancies were followed till term.

Outcome measures were overall pregnancy rate, implantation rate, clinical pregnancy rate and live birth rate which were compared between –

Group 1: women undergone hysteroscopic endometrial injury prior to IVF-ET

Group 2: women not undergone hysteroscopic endometrial injury prior to IVF-ET

Definitions:

- Recurrent implantation failure - Failure to achieve a clinical pregnancy after 2 or more attempts of transfer of good quality embryos.
- Implantation rate- The number of embryos which have produced ultrasonographic evidence of an intrauterine gestational sac per the total number of embryos transferred into the uterine cavity.
- Overall pregnancy rate - The number of patients with biochemical / clinical pregnancy divided by the number of patients who had embryo transfer.
- Clinical pregnancy rate - The number of patients with clinical pregnancy divided by the number of patients who had embryo transfer. Clinical pregnancy was defined by the ultrasound evidence of fetal heart beat.
- Live birth rate - The number of patients with live births divided by the number of patients who had embryo transfer.

Statistical analysis was done using IBM SPSS version 17. Continuous variables are shown as mean \pm SD whereas categorical variables are expressed as frequencies and percentages. Categorical data was analysed using Chi-square Analysis / Fisher's Exact test and continuous data were analysed using student t test. A p value of <0.05 was utilized to indicate significant difference.

Observation and Results

The baseline characteristics between the two groups, with endometrial scratching prior to IVF-ET

(Group1) and without endometrial scratching (Group 2) were compared. The mean age of the women in group 1 was 34.76 years and group 2 was 35.12 years (p-value 0.83)

The pregnancy outcome measures were compared between the two groups with and without endometrial injury prior to IVF-ET to evaluate any benefit from the procedure in women with recurrent implantation failure.

On comparing the effect of hysteroscopic endometrial injury done prior to IVF embryo transfer cycle versus no endometrial injury in patients with recurrent implantation failure, Implantation rates were 18.8% versus 13.2% (p-value 0.569), Overall pregnancy rate 40.6% versus 38.5% (p-value 0.872), Clinical pregnancy rate 37.5% versus 34.6% (p-value 0.820) and live birth rate 21.9% versus 19.2% (p-value 0.802).

Table 1: Baseline characteristics of the two groups.

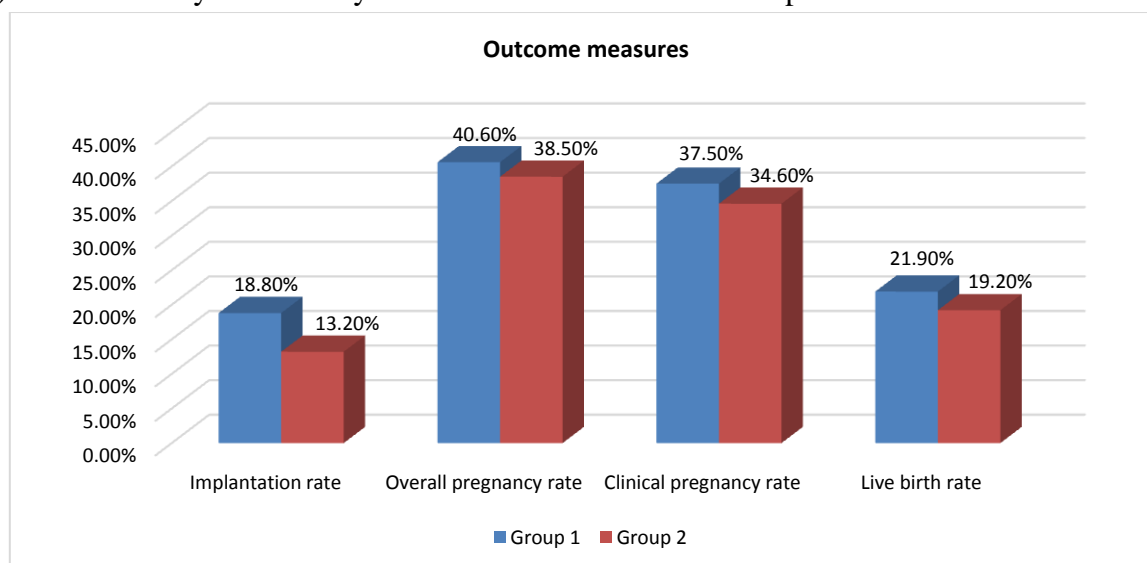
Baseline characteristics of the two groups with and without endometrial injury were comparable.

Baseline Characteristics	With endometrial injury Group 1	Without endometrial injury Group 2	p value
Age (yrs)	34.76±5.65	35.12±7.31	0.83
BMI	25.12±3.81	25.31±4.4	0.86
Duration of Infertility (yrs)	8.8±4.6	9.1±5.7	0.82
ET on the day of transfer (mm)	8.8±1.6	9.2±1.9	0.39
Number of Embryos transferred	2.8±0.87	3.1±0.71	0.16

Table 2: Pregnancy outcome measures of the two groups with endometrial injury (group1) and without endometrial injury (group2) prior to embryo transfer

Outcome Measures	With endometrial injury Group 1(n=32)	Without endometrial injury Group 2 (n=26)	p value
Implantation rate	18.8%	13.2%	0.57
Overall PR	13 (40.6%)	10 (38.5%)	0.87
Clinical PR	12 (37.5%)	9 (34.6%)	0.82
Live BR	7 (21.9%)	5 (19.2%)	0.80

Fig 1: Pregnancy outcome measures with endometrial injury (group 1) and without endometrial injury (group 2) in IVF - embryo transfer cycles of women with recurrent implantation failure.



Discussion

Our study was conducted to evaluate the role of hysteroscopic endometrial injury in improving the pregnancy outcome in women with recurrent implantation failure. In our study, the overall pregnancy rate, implantation rate, clinical pregnancy rate and live birth rate were similar in the two groups.

This was comparable to the study conducted by Tarek et al.¹³ the TROPHY Trial reported at the 30th annual meeting of ESHRE on 30th June 2014 in Munich. It was a large randomised trial performed in eight IVF centres in Europe between 2010 and 2013. More than 700 women were randomised to IVF with hysteroscopy (in the preceding cycle), or IVF without; all were under the age of 38, without known uterine pathology, and had history of unsuccessful IVF (two to four failed cycles). Outcome results following IVF showed no significant difference between the two groups - a live birth rate per patient of 31% in the hysteroscopy group and 29% in the control group. The results indicate that routine outpatient hysteroscopy prior to IVF in women who have experienced two to four failed IVF attempts do not significantly improve the subsequent IVF outcome. A Cochrane review¹⁴ in 2015 included 14 parallel-design randomised controlled trials (RCTs) with 1063 women in the intervention groups and 1065 women in the control groups. There was moderate-quality evidence to indicate that endometrial injury performed between day 7 of the previous cycle and day 7 of the ET cycle, increases the likelihood of live birth/ongoing pregnancy and clinical pregnancy (CPR 38.6% vs 29.8% and LBR 34.2% vs 26%). Although an overall benefit was observed, subgroup analysis suggested that endometrial injury might benefit only women with two or more previous failures. Eight of the 14 included studies were deemed to be at high risk of bias in at least one domain. Hence further research is needed to confirm this observation.

Van Hoogenhuijze et al¹⁵(2018) conducted a systematic review and meta-analysis on the effect of endometrial scratching in patients with or without

prior failed ART cycles on live birth (LBR) and clinical pregnancy rates (CPR) and concluded that it remains unclear whether endometrial scratching improves the chance of pregnancy and, if so, for which group of women. The meta-analysis conducted by Nikoletta et al.¹⁶ (2015) which included 4 RCT's found significant clinical heterogeneities with regards to patient characteristics, intervention used, phase of previous menstrual cycle. The results were inconsistent with regards to clinical pregnancy rate (CPR) and live birth rate (LBR). Thus there is insufficient evidence to support endometrial injury prior to embryo transfer.

Siristatidis et al¹⁷ conducted a prospective non randomised trial in 2017 and found endometrial injury induced through office hysteroscopy in the preceding cycle in subfertile women with recurrent implantation failure improves live birth rates. Yeung et al.¹⁸ conducted a RCT in 2014 including 300 women and found clinical pregnancy rate comparable in cases versus control group (CPR 34% versus 38%). The clinical pregnancy rate in our study was 43.4% in cases versus 44.1% in control group with no statistically significant difference among the groups. Cochrane review 2012 by Nastri al.¹⁹ included 591 women. In the subgroup 'Injury in the previous cycle' it was observed that endometrial injury performed within one month of the embryo transfer cycle was associated with improved live birth and clinical pregnancy (CPR 41% versus 21% and LBR 33% versus 17%).

Thus, the various studies in the literature evaluating the role of endometrial injury in improving the IVF treatment outcome have significant clinical heterogeneity with regards to patient characteristics, intervention used, phase of previous menstrual cycle. There is conflicting evidence in the literature regarding the definite role of endometrial injury / stimulation as a procedure to improve pregnancy outcome in women with recurrent implantation failure. The consensus regarding the patient selection, the timing of endometrial injury, technique and number of endometrial stimulation required are lacking. Well designed RCT's are

required to evaluate the role of hysteroscopic endometrial injury prior to IVF-embryo transfer in patients with recurrent implantation failure prior to making clinical recommendations.

Our study did not show statistically significant improvement in the pregnancy outcome after hysteroscopic endometrial injury in IVF-ET cycles. One of the limitation of the study was small sample size. We also could not randomise the patients into the groups. The subjects fell into the two groups just based on their preferences after proper counselling.

Conclusion

Our study did not show statistically significant improvement in the pregnancy outcome after hysteroscopic endometrial injury in IVF-ET cycles. Therefore, hysteroscopic endometrial injury in IVF-ET cycles in women with recurrent implantation failure may not be beneficial in improving the pregnancy outcome.

References

1. De Mouzon J, Goossens V, Bhattacharya S, Castilla JA, Ferraretti AP, Korsak V, Kupka M, Nygren KG, Nyboe Andersen A: Assisted reproductive technology in Europe, 2006: results generated from European registers by ESHRE. *HumReprod* 2010, 25:1851-1862.
2. Cross JC, Werb Z, Fisher SJ. Implantation and the placenta: key pieces of the development puzzle. *Science* 1994;266: 1508–18.
3. Barash A, Dekel N, Fieldust S, Segal I, Schechtman E, Granot I. Local injury to the endometrium doubles the incidence of successful pregnancies in patients undergoing in vitro fertilization. *FertilSteril* 2003;79(6):1317-22.
4. Li 2009 Li R, Hao G. Local injury to the endometrium: its effect on implantation. *Current Opinion in Obstetrics and Gynecology* 2009;21(3):236–9. [PUBMED: 19352180]
5. Garris 2003 Garris DR, Garris BL. Cytomorphometric analysis and surface ultrastructure of developing decidua. *Tissue and Cell* 2003;35(4):233–42. [PUBMED: 12921706]
6. Gnainsky Y, Granot I, Aldo PB, Barash A, Or Y, Schechtman E et al. Local injury of the endometrium induces an inflammatory response that promotes successful implantation. *FertilSteril* 2010;94(6):2030-6.
7. Lass 1998 Lass A, Peat D, Avery S, Brinsden P. Histological evaluation of endometrium on the day of oocyte retrieval after gonadotrophin-releasing hormone agonist-follicle stimulating hormone ovulation induction for in-vitro fertilization. *Human Reproduction* 1998;13(11):3203–5. [PUBMED: 9853881]
8. Ubaldi 1997 Ubaldi F, Bourgain C, Tournaye H, Smits J, Van Steirteghem A, Devroey P. Endometrial evaluation by aspiration biopsy on the day of oocyte retrieval in the embryo transfer cycles in patients with serum progesterone rise during the follicular phase. *Fertility and Sterility* 1997; 67(3):521–6. [PUBMED: 9091341]
9. Zhou L, Li R, Wang R, Huang HX, Zhong K. Endometrial injury in COH cycle. *FertilSteril* 2008;89(5):1166-76.
10. Kalma Y, Granot I, Gnainsky Y, Or Y, Czernobilsky B, Dekel N, Barash A. Endometrial biopsy-induced gene modulation: first evidence for the expression of bladder-transmembranaluroplakinIb in human endometrium. *Fertil Steril* 2009;91(4)
11. Song H, Lim H, Paria BC, Matsumoto H, Swift LL, Morrow J et al. Cytosolic phospholipase A2 alpha deficiency is crucial for 'on time' embryo implantation that directs subsequent development. *Development* 2002;129:2879-89.
12. Carson DD, Julian J, Lessey BA, Prakobphol A, Fisher SJ. MUC1 is a scaffold for selection ligands in the human uterus. *Front Biosci* 2006;11:2903-8.

13. Tarek El-Toukhy, Rudi Campo, Sesh Kamal Sunkara, Yacoub Khalaf and Arri Coomarasamy. A multi-centre randomised controlled study of pre-IVF outpatient hysteroscopy in women with recurrent IVF implantation failure: Trial of Outpatient Hysteroscopy - [TROPHY] in IVF. *Reproductive Health* 2009, vol.6:20
14. Carolina O Nastri, Sarah F Lensen, Ahmed Gibreel, Nick Raine-Fenning, Rui A Ferriani, Siladitya Bhattacharya, Wellington P Martins. Endometrial injury in women undergoing assisted reproductive techniques. *Cochrane Database of Systematic Reviews* 2015, Issue 3. DOI: 10.1002/14651858.CD009517.pub3
15. Van Hoogenhuijze NE, Kasius JC, Broekmans FJM, Bosteels J, Torrance HL et al. Endometrial scratching prior to IVF; does it help and for whom? A systematic review and meta-analysis. *Hum Reprod Open*. 2019 Jan 29;2019(1):hoy025. doi: 10.1093/hropen/hoy025.
16. Nikoletta Panagiotopoulou, Stamatios Karavolos, Meenakshi Choudhary et al Endometrial injury prior to assisted reproductive techniques for recurrent implantation failure: a systematic literature review. *European journal of obstetrics and gynaecology and reproductive biology*, october 2015, volume 193, pages 27–33
17. Siristatidis C, Kreatsa M, Koutlaki N, Galazios G, Pergialiotis V, Papantoniou N et al. Endometrial injury for RIF patients undergoing IVF/ICSI: a prospective nonrandomized controlled trial. *Gynecol Endocrinol*. 2017 Apr;33(4):297-300. doi: 10.1080/09513590.2016.1255325. Epub 2016 Dec 2.
18. Yeung TW, Chai J, Li RH, Lee VC, Ho PC, Ng EH et al. The effect of endometrial injury on ongoing pregnancy rate in unselected subfertile women undergoing in vitro fertilization: a randomized controlled trial. *Hum Reprod*. 2014 Nov;29(11):2474-81.
19. Nastri CO, Gibreel A, Raine-Fenning N, Maheshwari A, Ferriani RA, Bhattacharya S, Martins WP. Endometrial injury in women undergoing assisted reproductive techniques. *Cochrane Database of Systematic Reviews* 2012, Issue 7. Art. No.: CD009517. DOI: 10.1002/14651858.CD009517.pub2.