

Study of Effects on Endometrial Thickness and Pregnancy Outcome of Intrauterine Instillation of Autologous Platelet-Rich PlasmaHena Jabin¹, Homa Imam², Anupama Sinha³^{1,2}Senior Resident, Department of Obstetrics and Gynaecology, Jawaharlal Nehru Medical College and Hospital, Bhagalpur, Bihar³Professor and Head of Department, Department of Obstetrics and Gynaecology, Jawaharlal Nehru Medical College and Hospital, Bhagalpur, Bihar

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Abstract:**Background:** As infertility becomes a big issue in reproductive lives globally, many females are diagnosed as infertile each year. Examining the efficacy of intrauterine infusion of autologous platelet-rich plasma (PRP) in infertile women undergoing frozen embryo transfer cycles with poor endometrium is the aim of this study.**Methods:** In all, 61 women between the ages of 22 and 40 were involved in the research. They were split into two groups, A representing normal hysteroscopy findings and B representing abnormal hysteroscopy findings, based on the results of the hysteroscopy. Even after receiving estradiol valerate therapy, women with an endometrial thickness of ≤ 7 underwent intrauterine instillation of autologous PRP. Following PRP infusion for 48 and 72 hours, an increase in EMT was assessed. When the endometrium reached its ideal pattern of thickness, appearance, and vascularity, embryo transfer was carried out.**Result:** In comparison to Group A, EMT increased dramatically ($p < 0.05$) in Group B. There were no discernible differences in CPR, IR, or LBR between the two groups ($p < 0.001$). Clinical pregnancy and LBR both rose overall.**Conclusion:** Patients with low EMT undertaking IVF treatment and those who have had two or more prior IVF failures benefit from autologous platelet-rich plasma (PRP) infusion, which enhances endometrial thickness and the success rate in pregnancy.**Keywords:** Endometrial thickness, Hysteroscopy, Fertility-outcome, In vitro fertilization, Embryo transfer, intrauterine instillation, Thin endometrium, Ultrasonography.This is an Open Access article that uses a funding model which does not charge readers or their institutions for access and distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>) and the Budapest Open Access Initiative (<http://www.budapestopenaccessinitiative.org/read>), which permit unrestricted use, distribution, and reproduction in any medium, provided original work is properly credited.**Introduction**

Adult infertility is a serious issue that causes social and psychological suffering. A World Health Organization (WHO) survey states that 8–10% of couples worldwide struggle with infertility at some point in their reproductive careers. [1]. Many women are found to be infertile each year for a variety of causes, including endometriosis, pelvic adhesions, ovulatory problems, bilateral tubal blockage, hyperprolactinemia, and acquired tubal abnormalities. Advancement and usage of assisted reproductive technology (ART) like in-vitro fertilization (IVF) have helped to combat this problem to a certain extent. [2]

A phenomenon that is found to be associated with the less probability of implantation of an embryo is the low endometrial thickness (EMT). It is observed that low endometrial thickness (< 7 mm) at the end of the follicular phase is negatively associated with the benefit of IVF treatment. It accounts for affecting 5% of the women

undergoing IVF treatment. [3] Multiple treatment methods are suggested to increase the endometrial thickness. A Few such methods are Platelet-rich plasma (PRP) treatment, stem cell therapy, low-dose Human chorionic gonadotropin (HCG) administration, and Granulocyte colony-stimulating factor (G-CSF) treatment. [4] Under PRP therapy, the platelet-rich plasma of autologous blood is administered to the patient. This PRP is 4-5 times more concentrated with platelets than blood. [5] Multiple studies had demonstrated that administering PRP conjugated with Hormone Replacement Therapy (HRT) prior to embryo transfer enhances the likelihood of conception. [6]

Autologous PRP is the stimulator of many growth factors such as vascular endothelial growth factors (VEGF), epidermal growth factor (EGF), platelet-derived growth factor (PDGF), and transforming growth factor (TFG), fibroblast growth factor (FGF), insulin-like growth factor I, II (IGFI,II),

connective tissue growth factor (CTGF), and cytokines. Hence it increases endometrial thickness, resulting in successful embryo implantation. [7] Platelets also activate peripheral blood mononuclear cell (PBMC), which releases IL-10. IL-10 is a cytokine involved in tissue regeneration. [8] A significant concentration of PDGF-AB, PDGF-BB, and TGF- were found in PRP solution during the therapy. [9]

PRP therapy is a low-cost, low-risk treatment module and advancement in the field of this therapy will result in increased benefits in ART. Our study had shown improvement in endometrial thickness and pregnancy rates after PRP administration in patients with low EMT undergoing IVF treatment. The objective of this study was to evaluate the efficacy of intrauterine infusion of PRP on endometrial thickness and safety in women undergoing infertility treatments. The secondary objective was to study the implantation rate and clinical pregnancy rate.

Material and Methods

This prospective study was conducted from November 2022 to October 2023 at Obstetrics and Gynaecology department of Jawaharlal Nehru Medical College and Hospital, Bhagalpur, Bihar.

All infertile women attending JLNCH were started on oral Estradiol valerate from day 1 of their menses in a dose of 6 to 8 mg/day; the amount was progressively increased to 12 mg/day in divided doses as needed.

Serial transvaginal ultrasound examinations were done by using a transvaginal probe of 5 to 9 MHz, on Voluson E (Voluson, GE Healthcare, India) starting from day 7 or 8, and repeated as required to measure the endometrial thickness. Serum E2 level should be >250ng/ml. For patients who did not respond to oral therapy, transdermal estrogen (17-

□ Estradiol) has been given. If ET still remains ≤ 7 mm then that patients were included in the study. Patients included in this study were divided into two major groups based on hysteroscopy findings:

Group A: Patients having endometrium thickness <7mm with normal hysteroscopy findings.

Group B: Patients having endometrium thickness <7mm with abnormal hysteroscopy i.e. having polypectomy, adhesiolysis, and endometritis.

After obtaining written informed consent from the selected women, PRP was done in the stimulated cycle on the day of the trigger in both groups. Endometrial thickness was measured after 48 and 72 hours of the PRP treatment. Patients who did not show response in the form of increased thickness were submitted for repeat PRP in the next cycle.

Data collected were analyzed using the Graph pad prism software version 9. Paired t-test was used, and a P value of <0.01 was considered to be statistically significant.

Results

In this study total of 61 patients were included. Out of these 44 patients were in group A having normal hysteroscopy findings, and 17 patients were in group B having abnormal hysteroscopy findings. The average age of patients in group A was 33.3 years and it was 36.87 years in group B.

The BMI of all patients in both groups was comparable. The average years of infertility were 6.5 years in group A and 8.5 years in group B. In group A, three previous IVF attempts failed only in one patient, nine patients had two failures, and 13 patient's just one failure. In group B, there was 1 case where three previous IVF attempts failed, 7 cases where it failed twice, and 7 cases where it failed just once.

Table 1: Characterization of the study groups according to the parameters

Parameters	Group A (Normal Hysteroscopy) (n=44)	Group B (Abnormal Hysteroscopy) (n=17)	p-value
	Mean±SD		
Age (years)	33.3±5.3	36.87±4.69	0.018
BMI (kg/m ²)	27.2±7.34	28.63±5.53	0.470
Duration of infertility (years)	6.5±2.88	8.25±3.37	0.047
Previous IVF failure (attempt)	1.55±0.97	1±0.85	0.044
Basal Pre PRP E2 (ng/dl)	553±428	546.02±430.6	0.954

As per Table 2 Mean increase in EMT was significantly increased following PRP administration ($p < 0.001$) with an average increase of 1.62 mm (24.96% rise) after 48 hours and 2.42mm (37.28% rise) after 72 hours of PRP instillation in the normal hysteroscopy group and an increase of 2.61 mm (43.5% rise) after 48 hours and 3.28 mm (54.66% rise) after 72 hours in abnormal hysteroscopy group.

Table 2: Comparison of EMT in both the study groups

Study Group	Pre PRP EMT	EMT after 48 hrs of PRP	EMT after 72 hrs of PRP	Mean and % increase in EMT after 48 hrs if PRP	Mean and % increase in EMT after 72 hrs if PRP	p-value
Group A Normal Hysteroscopy (n=44)	6.49±0.72	8.11±0.78	8.91±1.35	1.62 mm & 24.96%	2.42 mm & 37.28%	<0.05
Group B Abnormal Hysteroscopy (n=17)	6.00±0.58	8.61±2.25	9.28±2.47	2.61 mm & 43.5%	3.28 mm & 54.66%	
p-value	0.01	0.20	0.46			

Table 3: Showing various parameters during the IVF cycle

	Normal Hysteroscopy Group A (N=44)	Abnormal Hysteroscopy Group B (N=17)
Embryo transfer performed on day 3	25	8
Embryo transfer performed on day5	19	9
Fresh et	12	9
Thaw et	32	13
Total embryo transferred	D3-79 D5-19	D3-22 D5-19
Average embryo Transferred per cycle	2.81±0.84	2.06±1.11

Table 4: Characterization of study groups according to parameters

Study groups	Positive b-HCG		Negative b-HCG		Clinical pregnancy		Live birth		Live birth rate per embryo	Implantation rate per embryo
	No.	%	No.	%	No.	%	No.	%		
Group A Normal Hysteroscopy(N=44)	24	54.55%	20	45.45%	22	50%	30	47%	24.8%	25.2%
Group B Abnormal Hysteroscopy(N=17)	10	58.82%	7	41.18%	9	52.94%	10	40%	24.4%	26.82%

Discussion

The incidence of thin endometrium varies from 1% to 2.5% in ART. [5,6] The most common pathological causes of thin endometrium may include Asherman syndrome, history of uterine surgery, infection, and radiation. [6] This research was conducted to determine the efficacy of autologous PRP in the study groups. Most of the studies have excluded patients having thin endometrium with abnormal hysteroscopy findings and cases in which operative hysteroscopy was done. But in this study, we have included all of these patients in group B and the results were compared for the outcome in both the groups. Adequate endometrial thickness is the main factor for implantation and pregnancy.

Women with persistent thin endometrium often do not undergo embryo transfer. Although several methods have been adopted for endometrial preparation like low dose Aspirin, Pentoxifylin, Low molecular weight heparin, Sildenafil, Vaginal estrogens, Nitro-glycerine patch, etc., but the success rate is insignificant mostly. Recently, local infusion of G-CSF is introduced, which is a cytokine that stimulates endometrial

proliferation.[5] However, PRP infusion stimulates proliferation and regeneration of endometrium with a large number of growth factors and cytokines. [7] In the present study Autologous platelet-rich plasma was infused, which contains 5 to 6 times more concentration of platelets / ml of plasma and its effect was observed. In this study, 80% of patients have shown a significant increase in endometrial thickness, hence successful embryo transfer could be completed in the same cycle. Patients who did not show an increase in endometrial thickness were subjected to repeat PRP in the next cycle and they were excluded from the study to check secondary outcomes.

In both the study groups after embryo transfer, 50% of the patients conceive successfully and our results are in accordance to the previous studies. Though, Poor response to PRP infusion cannot be explained fully, but the literature says severe damage to the basal layer may be responsible for it. Various factors influence the yield of PRP such as the drawing of blood; centrifugation speed and time, Temperature of centrifugation, and use of anticoagulants, therefore the yield of PRP can also influence the results of PRP therapy. [10]

For the first time, Chang reported the efficacy of intrauterine infusion of PRP for endometrial growth in women with thin endometrium. In that trial, PRP was infused in 5 Women with adequate endometrium who had a poor response to conventional therapy during the FET cycle. The proper response to treatment was reported in all of them, and normal pregnancy was reported in 4 women (Changetal., 2015).[9] Gokalp Oner (Department of Obstetrics and Gynaecology, Kayseri Acibadem Hospital, Turkey) has published three case reports of hysteroscopic guided the patients conceived. In our study, the patients who did not show an increase in endometrial thickness were subjected to repeat PRP in the next cycle. [8]

Research conducted by Maryam Eftekhari et al., found that pregnancy rate and LBR were 20%. The implantation and clinical pregnancy rates were 12.7% and 30%, respectively, and the difference was statistically significant.

The average increase in the EMT was 0.6 mm compared with the EMT of their previous cycle. However, this difference was not statistically significant. Further, the EMT of 12 patients increased (mean difference: 1.3mm), while that of seven patients decreased (mean difference: 0.7mm); the EMT of one patient did not change. There were no adverse effects reported by the patients who were treated with Autologous PRP. [12]

Zadehmodarres et al. in a pilot study revealed the efficacy of PRP on endometrial growth. Adequate endometrial growth was found in all the participants after two PRP infusions in all patients who had a history of the cycle cancellation due to the thin endometrium. [13] PRP is an effective, safe, low-cost, easy treatment with no major side effects to improve endometrial thickness and help to reduce the cycle cancellation rate in IVF practice.

Conclusion

Autologous PRP instillation in the uterine cavity significantly increases endometrial thickness in patients with endometrium and also found to be effective in reducing the cycle cancellation rate. PRP Instillation is found more efficacious in women having abnormal hysteroscopic finding (group B) as compared to women having normal hysteroscopic finding (group A).

Increased in endometrial thickness was statistically significant ($P < 0.05$). There was no significant difference in pregnancy outcome (clinical pregnancy rate and LBR) in both groups.

There was no significant difference in CPR, IR, and LBR among both groups ($p < 0.001$). Overall, clinical pregnancy and LBR increased respectively. No adverse reactions were reported.

From the present study it is reported that PRP is an effective, safe, affordable and convenient method with no major side effects to improve endometrial thickness and help to reduce the cycle cancellation rate in IVF practice.

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