

ORIGINAL RESEARCH

Effects of autologous platelet-rich plasma in promoting endometrial thickness on patients with thin endometrium following IVF

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Article Info	ABSTRACT
Received Jun 18, 2024 Revised Sep 4, 2024 Accepted Sep 20, 2024 Published Dec 1, 2024 *Corresponding author: Gita Pratama gitapratama@yahoo.com Keywords: Platelet-rich plasma Endometrial thickness Thin endometrium In vitro fertilization Reproductive health	Objective: The objective of this research was to investigate the impact of autologous platelet-rich plasma (PRP) in enhancing endometrial thickness among individuals experiencing infertility associated with a thin endometrium. Materials and Methods: Nine individuals with a thin endometrium who participated in an in vitro fertilization (IVF) program were enrolled in the study. This study was carried out in Yasmin Clinic, Dr. Cipto Mangunkusumo Hospital, Jakarta, Indonesia. Patients underwent a hormone replacement protocol involving the preparation of the endometrium with estradiol valerate. Treatment with PRP was initiated when the endometrial thickness was less than 7 mm. Autologous PRP was infused into the uterine cavity between the 10th and 12th days after administering estradiol valerate, and the assessment of endometrial thickness was conducted using ultrasound 48 hours later. A second administration of PRP was provided in cases where the endometrial thickness was below 7 mm. Frozen-thawed embryo transfer (FET) was performed if the endometrium reached adequate thickness (minimum 7 mm). Results: Seven of nine patients had adequate endometrial thickness followed by FET. Endometrial thickness was improved in 8 from 9 patients (88.8%). Five patients were improved at the first autologous PRP infusion (62.5%) and three patients (37.5%) at the second PRP infusion. The implantation rate was 33.3-100%, clinical pregnancy was 100%, and ongoing pregnancy rate was 83.3%. Conclusion: The use of autologous platelet-rich plasma (PRP) successfully stimulates endometrial development in individuals with a thin endometrium during frozen-thawed embryo transfer.

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Highlights:

1. It was observed that autologous PRP substantially increased endometrial thickness in individuals experiencing infertility linked to a thin endometrium.
2. The elevated clinical pregnancy rate emphasizes the favorable effectiveness of autologous PRP in addressing issues related to a thin endometrium in IVF programs.



INTRODUCTION

A reduced endometrial thickness, measuring below 7 mm on the day of embryo transfer, is proposed as a factor contributing to diminished pregnancy and live birth rates among patients undergoing IVF. Eftekhari et al.¹ showed that IVF patients with endometrial thickness between 8 to 11 mm on the day of embryo transfer had an optimal pregnancy rate; however, if the endometrial thickness was greater than 14 mm, no pregnancy was achieved.^{1,2} The process of angiogenesis is crucial for the development of the endometrium and the proper functioning of the female reproductive system, including folliculogenesis, corpus luteum formation, differentiation, and implantation, as well as in the maintenance of pregnancy.³ Studies and literatures have demonstrated that VEGF, bFGF, and EGF are several growth factors that play important roles in angiogenesis during follicular development. VEGF is considered the most important growth factor in regulating angiogenesis in the endometrium.⁴⁻⁶

VEGF serves as a potent mitogen for endothelial cells and induces vascular permeability. Its expression within the human endometrium plays a regulatory role in endometrial vascularization. It is thought that the thin endometrium is caused by disturbed angiogenesis and reduced blood flow in the endometrium. A thin endometrium is identified by elevated impedance of blood flow in the radial uterine artery, limited epithelial growth, reduced expression of VEGF, and inadequate vascular development.^{4,7}

Many studies have attempted to increase endometrial thickness using various therapies, such as pentoxifylline, low-dose aspirin, intrauterine G-CSF infusion, and vaginal sildenafil. However, the results are still inconclusive regarding increasing endometrial thickness or increasing pregnancy rates in patients with thin endometrium.⁸⁻¹⁰ Platelet-rich plasma (PRP) refers to the plasma derived from fresh whole blood obtained from peripheral veins, which undergoes centrifugation to concentrate platelets. Upon activation, PRP contains various growth factors, including vascular endothelial growth factor (VEGF), transformation growth factor (TGF), epidermal growth factor (EGF), and platelet-derived growth factor (PDGF). These growth factors are activated after clotting and transformed into the bioactive forms. Because it contains many growth factors, PRP has been widely used in the efforts to regenerate tissues, including the endometrium. PRP can be injected directly to the endometrium and specifically activate the cells within it.¹⁻¹¹

Research has demonstrated that PRP can enhance cell migration, adhesion, proliferation, and differentiation

while facilitating the accumulation of the extracellular matrix. PRP has found extensive applications across diverse medical disciplines, including dermatology, orthopedics, ophthalmology, and surgery. Nevertheless, in the field of gynecology, there has been a scarcity of studies, particularly in exploring the use of PRP to enhance endometrial thickness in individuals facing infertility associated with a thin endometrium.^{11,12} The objective of this study was to explore the impact of autologous PRP on the augmentation of endometrial thickness in individuals with thin endometrium undergoing an IVF program at our institution.

MATERIALS AND METHODS

Nine patients with inadequate endometrial thickness (<7 mm) were recruited on the day of embryo transfer. This study was performed at the Yasmin Clinic IVF Center, Dr. Ciptomangunkusumo Kencana Hospital, Jakarta, from February 2018 to September 2019. The study was approved by the Research Ethics Committee of Universitas Indonesia (Reference number: 850/UN2.FI/ETIK/2017). The research adhered to the ethical guidelines outlined in the 2008 revised Declaration of Helsinki. Patients meeting the inclusion criteria provided their informed consent before participating in the project. The data retrieved are collected and analyzed using SPSS version 27.

A hysteroscopic examination was performed for all the patients to exclude any pathology that might cause thin endometrium, such as intrauterine adhesions or endometritis. Patients with hormonal abnormalities such as hyperprolactinemia and abnormal thyroid hormone levels were also excluded. Patients were evaluated on day 2 or 3 of menstruation via transvaginal ultrasound examination, and an artificial hormone replacement protocol was used before the frozen-thawed embryo transfer (FET) to prepare the endometrium. Estradiol valerate (6 mg/day) was given initially and was increased every three days until the maximum dose was reached (12 mg/day) on days 10 -12. Endometrial thickness was assessed through transvaginal ultrasound, and intrauterine autologous PRP infusion was administered on the next day if the endometrial thickness was below 7 mm. A follow-up transvaginal ultrasound was conducted 48 hours after the PRP infusion to assess post-treatment endometrial thickness.

The second autologous PRP infusion was given if the endometrial thickness did not reach at least 7 mm. However, if the endometrial thickness reached an adequate thickness, 400 mg of micronized progesterone ovule was administered twice daily and 5 mg of dydrogesterone 5 mg was started thrice daily. Frozen-

thawed embryo transfer was performed four days after administration of progesterone for cleavage (day 3) embryos and/or six days after administration of progesterone for blastocyst (day 5 or 6) embryos. Estradiol valerate and progesterone supplementation were maintained for a duration of 14 days post-frozen embryo transfer (FET). In the event of a positive serum β -hCG concentration, hormone supplementation was sustained until week 10 - 12 of gestation. Endometrial thickness was measured via transvaginal ultrasound. Chemical pregnancies were identified by β -hCG serum two weeks after FET, and clinical pregnancies were identified by the presence of fetal heartbeat in 5 weeks after FET.

PRP preparation

A peripheral venous blood sample of 10 ml was drawn and placed in a PRP reagent-BCT tube. Subsequently, at a room temperature of 26°C, immediate centrifugation was carried out at 3500 rpm for 5 minutes. The separated blood components formed three layers: cellular plasma in the supernatant, a buffy coat layer in the middle, and red blood cells settling at the bottom. The PRP was acquired by combining platelets with 1 ml of supernatant, resulting in approximately 1–2 ml of PRP. Following this, 1 ml of PRP was introduced into the uterine cavity using an intrauterine insemination (IUI) catheter.

RESULTS AND DISCUSSION

In this study, six patients were diagnosed with PCOS, two patients with tubal factor infertility, and one patient with primary amenorrhea due to WHO class I anovulation, which is hypogonadotropic-hypogonadism. Polyp resection through office hysteroscopy was performed on two patients as part of the intervention. In the remaining seven patients, who also underwent hysteroscopy, no abnormalities were detected. All the participants underwent autologous PRP intrauterine infusion due to suboptimal endometrial thickness. Endometrial thickness was improved and reached ET 8 mm in eight of nine patients (88.8%). Five patients after the first autologous PRP infusion (62.5%) and three patients (37.5%) after the second PRP infusion ([Table 1](#)). However, one patient did not reach the ideal endometrial thickness after the second PRP infusion.

Frozen-thawed embryo transfers were performed for seven patients with adequate endometrial thickness, while two patients (5 and 8) did not undergo to FET because the endometrial thickness was still ≤ 8 mm ([Table 2](#)). Patient number nine experienced unsuccessful implantation, whereas six other patients achieved pregnancy, with normal progression observed in five of them. However, one of them had a miscarriage. In this study, the range of implantation rate varied from 33.3% to 100%, the ongoing pregnancy rate was 83.3%, and the clinical pregnancy rate reached 100%.

Table 1. Patients' age, diagnosis and improvement of endometrial thickness

No	Age	Diagnosis	Increase of endometrial thickness (mm)
1	39	Tubal factor	0.7/0*
2	32	Primary amenorrhea (WHO class I anovulation)	0.9
3	37	PCOS	0.8
4	38	Tubal factor	0.9/0.5*
5	36	PCOS	-0.78
6	30	PCOS	0.5
7	41	PCOS	0.9
8	41	PCOS	0.2
9	29	PCOS	0.9

*Endometrial thickness: 48 h after the first PRP/48 h after the second PRP injection

Table 2. IVF outcomes after PRP treatment

Patient number	Amount embryo transferred	Chemical pregnancy	Clinical pregnancy	Ongoing pregnancy	Implantation rate
1	2	yes	yes	singleton pregnancy	50%
2	1	yes	yes	singleton pregnancy	100%
3	3	yes	yes	miscarriage on 8 weeks gestational age	33.3%
4	3	yes	yes	singleton pregnancy	33.3%
5*	-	-	-	-	-
6	3	yes	yes	twin pregnancy	67.7%
7	2	yes	yes	singleton pregnancy	50%
8*	-	-	-	-	-
9	2	no	no	no	-

*Patients did not undergo FET

Platelet-Rich Plasma (PRP), alternatively termed autologous conditioned plasma, represents a plasma fraction derived from an individual's blood. This plasma is enriched with platelets at concentrations 4-5 times higher than those found in the circulating blood. Platelets serve essential functions within the body, primarily to prevent excessive blood loss and repair vascular walls and adjacent tissues following injuries. By harnessing the regenerative properties of platelets, PRP has garnered attention as a therapeutic modality in various medical fields, aiming to harness the body's natural healing processes for improved tissue repair and regeneration. This autologous approach, utilizing the patient's blood components, aligns with the pursuit of safer and more personalized medical interventions. PRP is known to contain several growth factors, such as VEGF, TGF, PDGF, and EGF, which could regulate cell differentiation, proliferation, migration, and attachment.^{11,13,14}

Presently, the application of PRP infusion is on the rise across various medical fields, emerging as a treatment modality for individuals with thin endometrium. PRP is recognized for its favorable attributes, including safety, minimal to no side effects, and ease of accessibility. Despite these advantages, there remains a dearth of conclusive evidence regarding the efficacy of PRP in addressing thin endometrium-related issues, particularly in Indonesia. While the utilization of PRP in medical treatments is expanding, more comprehensive studies specific to the Indonesian population are imperative to establish the effectiveness of PRP in enhancing endometrial thickness and addressing infertility concerns related to thin endometrium. This emphasizes

the need for further research and clinical investigations to validate and refine the application of PRP as a therapeutic option for individuals facing challenges associated with thin endometrium in the Indonesian setting.¹⁵

The role of autologous PRP in thin endometrium was first evaluated by Chang et al., who showed that five patients undergoing IVF, who had poor response to standard hormone replacement therapy after PRP treatment, reached their optimum endometrial thickness and all of them were pregnant. Four out of five patients had usually progressing pregnancy, while one patient had a miscarriage.¹¹ Moreover, a study by Molina et al. stated that endometrial thickness increased to more than 7 mm after the first PRP infusion, and after the second infusion of PRP, endometrial thickness reached 9 mm in all cases. After the PRP treatment, the endometrium thickness of all patients is qualified for embryo transfer. The study reported a 73.7% positive pregnancy test rate, with 26.3% resulting in live births and an additional 26.3% indicating ongoing pregnancies.¹⁶

Tandulwadkar et al. evaluated endometrial thickness and vascularity after PRP treatment. They included 68 patients in their study, and 64 achieved optimal thickness for the FET cycle. The average endometrial thickness before PRP was 5 mm, and after PRP, it was 7.22 mm. Seventeen patients with sparse to modest vascularity before PRP resulted in excellent vascularity after PRP treatment, and the other 47 patients showed improvement from sparse to modest endometrial vascularity pattern, while four patients remained having sparse vascularity after PRP treatment.¹⁷

Another study conducted by Kim et al. involved subjects who had experienced failed IVF and had a thin endometrium. These subjects received 2 to 3 intrauterine PRP infusions. Frozen-thawed embryo transfer (FET) was followed, and a follow-up was conducted for up to 20 subjects. The results indicated that 12 patients exhibited an increase in endometrial thickness, while seven experienced a decrease. The ongoing pregnancy rate and live birth rate were 20%, with four out of 20 subjects achieving live births, two experiencing abortion, one having a chemical pregnancy, and 13 not achieving pregnancy.¹⁸

Several studies were recently published (including 2 randomized clinical trials) regarding the effect of PRP infusion in patients with thin endometrium, and all of them suggested that PRP infusion is significant in increasing the endometrial thickness in patients with thin endometrium.^{14,15,18-20} Our study showed that nine patients had poor endometrial thickness, and after PRP infusion, an increase in endometrial thickness was found in eight out of nine patients, which was 88.8%. One patient who got improvement in her endometrial thickness still did not reach optimum endometrial thickness after a second PRP. It is consistent with a meta-analysis that demonstrated patients with an endometrial thickness below 7 mm exhibited a notably reduced likelihood of clinical pregnancy compared to those with an endometrial thickness exceeding 7 mm.²¹ Another study used PRP infused into the endometrium to improve endometrial thickness and proceed to ET. It was found that 86% of subjects achieved an endometrial thickness greater than 7 mm, while 14% had a thickness below 7 mm. However, the reasons for the failure to achieve adequate endometrial thickness were not elaborated.²²

Our study was also in line with previous studies, which resulted in a positive outcome of intrauterine infusion of PRP towards endometrial growth and improvement of pregnancy rate in IVF patients. Out of seven patients who got embryo transfer, six patients were pregnant, and in five of them, the pregnancy went well, but unfortunately, one patient had implantation failure. The implantation rate was 33.3-100%, clinical pregnancy was 100%, and ongoing pregnancy rate was 83.3%.

In the course of our investigation, no adverse effects were discerned after the administration of PRP infusion. Nevertheless, it is noteworthy that one participant encountered a reduction in endometrial thickness, and another experienced a failure in implantation. The precise etiology behind these occurrences remains elusive, necessitating further inquiry. The current body of research needs to be revised to offer comprehensive insights into instances of diminished endometrial

thickness after PRP infusion. Furthermore, there are no research or evidence that found major side effects of PRP or determined whether it also has a role in implantation failure. This underscores the imperative for additional meticulous studies and systematic exploration to clarify the mechanisms and contributing factors associated with the observed outcomes, contributing to the refinement of our understanding and informing clinical practice in the realm of reproductive medicine. Furthermore, this could necessitate additional studies to investigate the outcomes and potential complications of neonates born to mothers with an improved thin endometrium.

This study has a notable limitation due to including a small number of patients. Consequently, additional research is essential to substantiate the effectiveness and safety of PRP in enhancing endometrial thickness among individuals facing infertility related to a thin endometrium. Furthermore, a comprehensive understanding of the precise mechanisms underlying the intrauterine infusion of PRP is crucial, especially in promoting endometrial growth in patients with a thin endometrium. Unveiling these mechanisms will contribute significantly to advancing our knowledge and improving therapeutic approaches for individuals experiencing challenges related to thin endometrium. Therefore, future investigations with larger sample sizes and in-depth mechanistic exploration are warranted to address these critical aspects and enhance the overall understanding of PRP's role in treating thin endometrium-related infertility, particularly in Indonesia.

CONCLUSION

Addressing the thin endometrium in patients poses a persistent challenge for healthcare providers. Various treatment approaches have been employed, but conclusive and positive outcomes are yet to be achieved. Based on the findings of the current investigation, PRP demonstrates encouraging outcomes in enhancing endometrial thickness in individuals experiencing infertility associated with a thin endometrium. Further studies with larger sample sizes and specific molecular basis data are recommended to enrich our comprehension of PRP treatment in patients with thin endometrium.

DISCLOSURES

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Conflict of interest

The authors affirm no conflict of interest in this study.

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Author contribution

GP collected the data, analyzed the statistics, and wrote the manuscript. MM, AKH, KS, RM, EA, and AMY conceptualized and reviewed the manuscript. All of the authors approved the final version of the manuscript.

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