Effects of Intrauterine Infusion of Autologous Platelet-Rich Plasma in Women Undergoing Treatment with Assisted Reproductive Technology: a Meta-Analysis of Randomized Controlled Trials

Auswirkungen einer intrauterinen Infusion von autologem plättchenreichem Plasma bei mit assistierter Reproduktionstechnologie behandelten Frauen: eine Metaanalyse randomisierter kontrollierter Studien

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Key words
platelet-rich plasma (PRP), assisted reproductive technology, randomized controlled trials (RCTs)

Schlüsselwörter
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ABSTRACT

Purpose This meta-analysis was conducted to systematically retrieve relevant randomized controlled trials (RCTs) and evaluate the effects of intrauterine infusion of autologous platelet-rich plasma (PRP) in women with thin endometrium, implantation or pregnancy failure undergoing treatment with assisted reproductive technology (ART).

Methods We conducted a systematic review and meta-analysis of the retrieved RCTs. Studies on the intrauterine infusion of PRP in women undergoing treatment with ART that were published in PubMed, the Cochrane library, Web of Science, and Embase from inception until June 2022 were included. The data were extracted and analyzed independently using the fixed-effects or random-effects model according to heterogeneity.

Results Seven RCTs involving 861 patients (435 in the intervention group and 426 in the control group) were included. The rates of clinical pregnancy (risk ratio [RR]: 2.51; 95% confidence interval [CI]: 2.0–3.13; P < 0.00001), chemical pregnancy (RR: 1.96; 95% CI: 1.58–2.45; P < 0.00001), live births (RR: 7.03; 95% CI: 3.91–12.6; P < 0.00001), and implantation (RR: 3.27; 95% CI: 1.42–7.52; P = 0.005) were significantly higher in the women who received PRP infusion than in the control group. No significant differences were noted in the miscarriage rate (RR: 0.98; 95% CI: 0.39–2.42; P = 0.96) between the two groups.

Conclusion In summary, intrauterine infusion of PRP may be an effective therapy for women with thin endometrium and recurrent implantaion failure (RIF) undergoing treatment with ART. More population-based RCTs are warranted to verify the efficacy of our evidence.

† These authors contributed equally.
Introduction

Infertility is defined as failure to achieve a successful pregnancy after at least 1 year of regular and unprotected intercourse, and its prevalence ranges between 9% and 18% among the general population [1]. Despite recent advancements in the field of assisted reproduction technology (ART), it is challenging to promote embryo implantation and prevent abortion. A thin endometrium, poor endometrial receptivity, embryo defects, and abnormal cross-talk between the endometrium and embryo are the main reasons for recurrent implantation failure (RIF) and recurrent pregnancy loss (RPL) [2, 3]. Endometrial quality is of paramount importance for successful embryo implantation [4].

A large number of individuals suffer from infertility; thus, methods such as the use of vaginal sildenafil, endometrial scratching, the intrauterine administration of granulocyte colony-stimulating factor or stem cells, blastocyst-assisted hatching and pre-implantation genetic diagnosis for aneuploidy, high-dose estrogen therapy, and treatment of thin endometrium have been proposed to improve the pregnancy outcomes in couples with implantation defects and pregnancy failure [5, 6, 7, 8]. However, these treatments do not help to improve the endometrial thickness and/or quality in the affected women. Therefore, a safer and more effective treatment method that can improve the pregnancy outcomes of couples with implantation defects and pregnancy failure is warranted.

Increasing evidence shows that intrauterine infusion of autologous platelet-rich plasma (PRP) is a novel potential method for treating thin endometrium via ART [9, 10]. PRP, also known as autologous conditioned plasma, is prepared by centrifuging patients’ peripheral blood samples and comprises high numbers of platelets [11]. A growing body of evidence suggests that platelets contain numerous proteins; several growth factors (GFs); and cytokines such as platelet-derived GF (PDGF), vascular endothelial GF (VEGF), transforming GF-β1 (TGF-β1), and anti-inflammatory cytokines [12]. These molecules are released upon activation and contribute to cell proliferation, migration, differentiation, chemotaxis, angiogenesis, and anti-inflammatory properties, resulting in improved endometrial growth and receptivity [7, 10]. PRP may thus be a novel treatment for women with a thin endometrium [9]. Moreover, Russell et al. [4] reported the effectiveness of PRP in inducing endometrial growth.

To date, several randomized controlled trials (RCT) have evaluated the efficiency of intrauterine infusion of autologous PRP in women undergoing treatment with ART; however, the results of those RCTs are not consistent. Therefore, the present meta-analysis aimed to screen RCTs that compared the effects of intrauterine infusion of PRP in women undergoing treatment with ART and summarize their results. The results of this meta-analysis will increase awareness among physicians in reproductive medicine, helping to formulate better treatment strategies to improve the pregnancy outcomes of couples with implantation defects and pregnancy failure.

Materials and Methods

Literature search

Two independent reviewers (HSF and JZS) conducted a systematic electronic literature search of PubMed, the Cochrane library, Embase, and Web of Science and identified all relevant studies published in English from inception until June 2022. The search strategy used the following keywords: (“Platelet-rich plasma” OR “Autologous platelet-rich plasma” OR “Platelet-rich plasma gel” OR “PRP”) and (“in vitro fertilization” OR “IVF” OR “Intracytoplasmic sperm injection” OR “ICSI” OR “Embryo transfer” OR “Assisted reproduction technologies” OR “ART”) and (“Randomised controlled trial” OR “RCT”). The end-list references of all relevant papers were also screened to further obtain potentially eligible studies.
Inclusion and exclusion criteria
The studies were included if they
1. were RCTs;
2. included patients undergoing treatment with ARTs, including
   in vitro fertilization (IVF) or intracytoplasmic sperm injection;
3. were already published;
4. compared intrauterine infusion of autologous PRP with no
   injection/placebo; and
5. included at least one of the following reported outcomes:
   chemical pregnancy rate, clinical pregnancy rate, and mis-
   carriage rate.

The studies were excluded if they
1. review articles, commentaries, letters, or observational studies;
2. were non-clinical trials;
3. were not RCTs; and
4. reported inability to extract data from the literature.

Data extraction and quality assessment
Using a standardized extraction form, two review authors (HSF
and JZS) independently extracted the following data from the
included studies: first author, year of publication, country, sample
size, population characteristics, interventions, and main results.
The quality of all of the included studies was appraised by two re-
viewers (JZS and TQQ) in accordance with the Cochrane Collabora-
tion’s tool [13]. A risk-of-bias table including the following ele-
ments was created: random sequence generation, allocation con-
ealment, blinding, incomplete outcome data, selective reporting,
and other bias. Discrepancies, if any, were resolved through con-
sultation with a third reviewer (TQQ).

Statistical analysis
All data were assessed using Review Manager 5.3 (Cochrane Col-
laboration, 2014). Dichotomous data are expressed as risk ratios
(RRs) with 95% confidence intervals (CI). The heterogeneity across
studies was evaluated based on the P and I² values and using stan-
dard chi-square tests. I² < 50% indicated moderate heterogeneity,
and a fixed-effects model was used for the meta-analysis; by con-
trast, a random-effects model was used when severe heterogene-
ity was identified (I² ≥ 50%). Subgroup analyses were conducted
to assess different populations, and sensitivity analysis was con-
ducted by excluding each study one by one. Publication bias was
evaluated by applying funnel plots.

Results
Study characteristics and quality assessment
► Fig. 1 presents a flow chart of the study inclusion process. In
total, 542 published articles were selected upon initial screening
of the electronic databases. Based on the exclusion criteria, 507
obviously irrelevant papers were excluded after scanning the titles
and abstracts. An additional 28 studies were excluded after care-
fully reading the full texts. Finally, seven eligible studies [14, 15,
16, 17, 18, 19, 20] were included for analysis. These seven studies
involved a total of 861 patients (426 in the control group and 435
in the treatment group). The basic characteristics of each study
are presented in ► Table 1. ► Table 2 presents the authors’ judg-
ments regarding the risk of bias across all RCTs.

Clinical pregnancy rate
All seven studies [14, 15, 16, 17, 18, 19, 20] reported on the clin-
cical pregnancy rates of the 861 patients. There was no heteroge-
nity across the studies (I² = 0%; P = 0.39). The pooled analysis
with the fixed-effects model showed a statistically significant in-
crease in the clinical pregnancy rate in the PRP group as compared
with the control group (RR: 2.51; 95% CI: 2.01–3.12; P < 0.00001;
► Fig. 2).

A subgroup analysis was conducted to examine whether a thin
endometrium, RPL, and RIF affected the patient outcomes. Com-
pared with the control group, in the treatment group, patients
with a thin endometrium, RPL, and RIF had RRs of 3.46 (95% CI:
1.58–7.59; two studies), 1.75 (95% CI: 0.61–5.05; one study), and
2.18 (95% CI: 1.93–3.12; four studies), respectively. Similarly, a
subgroup analysis was performed to explore whether the PRP dose
affected the patient outcomes. The results of the meta-analysis
showed that the RRs of the subgroups that were administered
PRP at doses of ≤0.5ml, ≥1ml, and 0.5–1ml were 2.58 (95% CI:
2.01–3.32; P=0.65; I² =0%; five studies), 2.18 (95% CI: 1.22–3.90;
P=0.009; one study), and 2.33 (95% CI: 0.98–5.54; P=0.06; one
study), respectively, relative to the controls. Finally, the stability
of our meta-analysis results was examined using sensitivity analyses
by sequentially excluding each study one by one; the results indi-
cated that our results were stable.
Whether a thin endometrium or RIF would affect the patients’ outcomes. The results of our meta-analysis revealed that patients with a thin endometrium or RIF who were administered PRP had an RR of 1.97 (95% CI: 1.57–2.48; P = 0.73; I² = 0%; three studies) and 1.88 (95% CI: 0.88–4.00; P = 0.73; one study), respectively, as compared with the controls.

**Miscarriage rate**
Three of the reported studies [14, 18, 19] included data on the miscarriage rate for a total of 221 patients (115 in the treatment group and 106 in the control group). As shown in Fig. 4, our meta-analysis results indicated an I² of 0% and P value of 0.64, suggesting that the heterogeneity across the studies was low. Therefore, the fixed-effects model was applied. There was no obvious difference in the miscarriage rate between the two groups (RR: 0.98; 95% CI: 0.39–2.42; P = 0.96; Fig. 4).
Table 2  Quality assessment of the included studies.

<table>
<thead>
<tr>
<th>Author (year)</th>
<th>Random Sequence Generation</th>
<th>Allocation concealment</th>
<th>Blinding of participants and personnel</th>
<th>Blinding of outcome assessment</th>
<th>Incomplete outcome data</th>
<th>Selective reporting</th>
<th>Other bias</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eftekhar (2018) [14]</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Nazari (2022) [18]</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Nazari (2020) [16]</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Nazari (2022) [17]</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Nazari (2019) [15]</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Zamaniyan (2020) [19]</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
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<tr>
<td>Obidniak (2017) [20]</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
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</tbody>
</table>

Fig. 2  Forest plot diagram showing the clinical pregnancy rate in women who received intrauterine platelet-rich plasma versus controls regarding population type (recurrent implantation failure (RIF), recurrent pregnancy loss (RPL) and thin endometrium). CI = confidence intervals.
Implantation rate

Only one of the included studies [19] reported data on the implantation rate. A statistically significant increase in the implantation rate was noted in the PRP group as compared with the control group (RR: 3.27; 95% CI: 1.42–7.52; P = 0.005; ▶ Fig. 5).

Live birth rate

Two studies [17, 18] including 433 patients reported data on the live birth rate. Heterogeneity was not examined (I² = 0% and P = 1.00). A pooled analysis with the fixed-effects model demonstrated a statistically significant increase in the live birth rate in the

<table>
<thead>
<tr>
<th>Study or subgroup</th>
<th>Event</th>
<th>Total</th>
<th>Weight</th>
<th>Risk ratio M-H, fixed, 95% CI</th>
<th>Risk ratio M-H, fixed, 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>1.2.1 RIF</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nazari 2019a</td>
<td>26</td>
<td>49</td>
<td>13</td>
<td>48</td>
<td>16.2%</td>
</tr>
<tr>
<td>Nazari 2021</td>
<td>101</td>
<td>196</td>
<td>49</td>
<td>197</td>
<td>60.4%</td>
</tr>
<tr>
<td>Zamaniyan 2020</td>
<td>20</td>
<td>55</td>
<td>10</td>
<td>43</td>
<td>13.9%</td>
</tr>
<tr>
<td><strong>Subtotal (95% CI)</strong></td>
<td><strong>300</strong></td>
<td><strong>288</strong></td>
<td><strong>90.5%</strong></td>
<td><strong>1.97 (1.57, 2.48)</strong></td>
<td></td>
</tr>
<tr>
<td>Total events</td>
<td>147</td>
<td>72</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heterogeneity: χ² = 0.62, df = 2 (p = 0.73); I² = 0%</td>
<td></td>
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<td></td>
<td></td>
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<tr>
<td>Test for overall effect: Z = 5.79 (p &lt; 0.00001)</td>
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</tr>
</tbody>
</table>

| **1.2.2 Thin endometrium** |       |       |        |                             |                             |
| Eftekhar 2018         | 14    | 40    | 8      | 43                           | 9.5%                         |
| **Subtotal (95% CI)** | **40** | **43** | **9.5%** | **1.88 (0.88, 4.00)** |                     |
| Total events          | 14    | 8     |        |                             |                             |
| Heterogeneity: not applicable |
| Test for overall effect: Z = 1.64 (p = 0.10) |

| **Total (95% CI)**    | **340** | **331** | **100.0%** | **1.96 (1.58, 2.45)** |                     |
| Total events          | 161    | 80     |        |                             |                             |
| Heterogeneity: χ² = 0.63, df = 3 (p = 0.89); I² = 0% |
| Test for overall effect: Z = 5.79 (p < 0.00001) |
| Test for subgroup differences: χ² = 0.01, df = 1 (p = 0.91); I² = 0% |

▶ Fig. 3 Forest plot diagram showing the chemical pregnancy rate in women who received intrauterine platelet-rich plasma versus controls regarding population type (recurrent implantation failure (RIF), and thin endometrium). CI = confidence intervals.

▶ Fig. 4 Forest plot diagram showing the miscarriage rate in women who received intrauterine platelet-rich plasma versus controls. CI = confidence intervals.
PRP group as compared with the control group (RR: 7.03; 95% CI: 3.91–12.6; P < 0.00001; ▶ Fig. 6).

Publication Bias

A funnel plot was applied to qualitatively evaluate the publication bias. The funnel plot presented in ▶ Fig. 7 is symmetrical, indicating that there was no publication bias among the included studies.

Discussion

Previous studies have reported that RIF or RPL may be caused by many factors, including poor endometrial receptivity, anatomic abnormalities, immune factors, endometrial thinning, embryonic quality, and infectious and genetic diseases [3, 21]. Moreover, previous meta-analyses have assessed the effects of PRP infusion in women undergoing treatment with ART [6]. However, the clinical reliability of those meta-analyses is uncertain because of the different article types (three RCTs and four cohort studies), which has increased the risk of bias. RCTs are generally considered the best approach for evaluating the effects of a treatment. In the present meta-analysis, we screened seven RCTs to evaluate the effectiveness of intrauterine infusion of PRP in women undergoing frozen–thawed embryo transfer. The results of our meta-analysis are partially consistent with those of a previous study [6]. We found that the treatment group had an improved clinical pregnancy rate, chemical pregnancy rate, live birth rate, and endometrial thickness as compared with the control group. Furthermore, our subgroup analyses specifically evaluated the effects of different PRP doses on the various outcomes of the patients undergoing treatment with ART. Our data showed that when PRP was administered at a dose of ≤ 0.5 ml or ≥ 1 ml, the clinical pregnancy rate was significantly higher in the treatment group than in the control group. However, the results related to the clinical efficacy of the possible PRP dose response are ambiguous, which may be attributable to differences in the PRP preparation methods.

An optimal endometrial status is important for correct implantation, subsequent embryonic development, and successful pregnancy. An endometrium is considered thin when its thickness is < 7 mm. A thin endometrium is associated with a reduced possibility of pregnancy through IVF [10, 22]. Intrauterine infusion of PRP is a novel approach that was first used in 2015 in the field of infertility for promoting endometrial growth [9]. Chang et al. reported that the intrauterine infusion of autologous PRP can increase the endometrial thickness and improve the pregnancy outcomes of women with inadequate endometrial growth [9]. Similarly, our
study indicated that PRP therapy may be successful in improving the pregnancy outcomes of patients with a thin endometrium. Furthermore, Eftekhar et al. reported that the endometrial thickness increased significantly from 6.09 mm to 8.67 mm in the PRP group and from 6.15 mm to 8.04 mm in the control group [14]. Kusumi et al. recently reported that some patients became pregnant although their endometrium was not receptive to PRP treatment [23]. This indicates that PRP not only improves the endometrial thickness but also enhances the endometrial quality. However, the exact molecular mechanism through which PRP therapy improves patients’ pregnancy outcomes remains unclear. The improvement of endometrial thickness and receptivity is the most accepted theory explaining the positive effects of PRP.

The endometrium starts becoming receptive during the middle-secretory phase of the 19th–23rd days of each IVF cycle; this is defined as the implantation window. Furthermore, GFs, interleukins, cytokines, prostaglandins, and adhesion molecules are expressed throughout the implantation window, and impairment of these agents can decrease the chances of implantation and pregnancy [24]. Indeed, PRP is a plasma fraction of autologous blood with a platelet concentration that is 4–5× greater than that normally contained in whole blood. PRP contains significant concentrations of GFs and cytokines such as vascular endothelial GF, PDGF, TGF, interleukin (IL)-6, and IL-8 [9, 25]. Various cytokine receptors for PDGF, TGF, and PDGF in the human endometrium are considered to promote endometrial tissue healing, play a role in paracrine and autocrine signaling, and be related to endometrial receptivity and embryo implantation and development [26, 27]. Furthermore, the stimulating, proliferation-inducing, and tissue regenerative effects of PRP have been explored in various areas of medicine, including osteoarthritis, ocular epithelial defects, dental disorders, and wound healing [28, 29]. Accordingly, we speculate that the intrauterine infusion of PRP stimulates cell proliferation and regeneration, enhances endometrial receptivity, and promotes implantation.

Although intrauterine infusion of autologous PRP is a novel technique, it is cost-effective and easily accessible for women with a refractory endometrium. However, data on the safety of intrauterine infusion of PRP and research on the possible adverse effects of this therapy on pregnancy-related outcomes are limited. Thus, this issue should be addressed in future studies.

Our study has some strengths. First, our meta-analysis focused on quantitatively evaluating the efficacy of intrauterine infusion of autologous PRP in women undergoing treatment with ART. Second, our meta-analysis involved a rigorous search strategy and included only those studies with a prospective RCT design. Third, all of the included studies were of high quality. Finally, the funnel plot showed no significant asymmetry, indicating the lack of publication bias across the included studies.

However, our study has some limitations as well. First, most of our research was performed in Iran, and our findings may thus not be generalizable to other populations. Furthermore, four of the seven studies were performed by the same first author and their colleagues; this may considerably affect the judgment of the meta-analysis results because there is not only geographical bias but also a great risk of personal systematic bias (for example, all four studies conducted by Nazari et al. used 0.5 ml of PRP). Second, our meta-analysis included only seven RCTs with small numbers of patients. Third, subgroup analyses were not performed for some outcomes because of the limited number of the studies included; therefore, we could not determine the source of heterogeneity. Fourth, only those RCTs published in English were included;
Thus, relevant studies in other languages may have been missed, which may have introduced a language bias. Fifth, to produce consistent and accurate results, a standardized PRP preparation scheme is needed. Finally, although all of the included studies were RCTs, some did not adequately describe the randomization methods, allocation concealment, blinding procedures, or missing data, thus conferring high risks of publication, selection, and reporting biases. Therefore, large, well-designed, and multi-center RCTs are warranted to obtain further evidence.

In conclusion, despite the aforementioned limitations of this meta-analysis, our results suggest that the intrauterine infusion of PRP increases the clinical pregnancy rate, chemical pregnancy rate, live birth rate, and implantation rate among women with thin endometrium and recurrent implantation failure (RIF) undergoing treatment with ART. However, these findings need to be verified through larger, more elegantly designed RCTs.

**Funding**

Not applicable.

**Contributors’ Statement**

HSF and JZS conceived and designed the study. HSF and TQQ conducted the data searches. SFH and JZS performed the analysis, wrote and revised the manuscript. TQQ gave the final approval of the manuscript.

**Conflict of Interest**

The authors declare that they have no conflict of interest.

**References**


