First-Trimester Serum Cytokine Profile in Pregnancies Conceived After Assisted Reproductive Technology (ART) With Subsequent Pregnancy-Induced Hypertension

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Pregnancy-induced hypertension (PIH) is one of the most common pregnancy complications that seriously affects the mother and fetus. The incidence of PIH is higher in pregnancies conceived after assisted reproductive technology (ART) than in spontaneous pregnancies; thus, exploring potential serum biomarkers before PIH onset is of great significance for effective early prediction and prevention of PIH in the ART population. Cytokines are involved in the inflammatory response and immune regulation, which play an essential role in the pathogenesis of PIH. A description of the cytokine profile in the first trimester of pregnancy could help identify new diagnostic tools and develop targeted therapies for PIH in the ART population. The concentrations of classical predictive markers for PIH and another 48 cytokines were measured in the first-trimester pregnancy serum samples from 33 PIH patients and 33 matched normotensive controls (NC), both of whom conceived after ART treatment. The measured values were compared and analyzed between NC and PIH, followed by comprehensive bioinformatic analysis and logistic regression analysis. There was no significant difference in classical predictive markers, including Activin A, PlGF, sFLT1 (VEGFR), and sFLT1/PlGF, between the PIH and NC groups (P > 0.05), while 29 cytokines were significantly lower in the PIH group than in the NC group (P < 0.05). Logistic regression analysis revealed that 17 cytokines (IL-2Rα, M-CSF, IL-6, IL-2, β-NGF, IL-7, IL-12 (p70), SCF, IL-10, IL-9, MIG, GM-CSF, LIF, IL-1α, MCP-3, IL-4, and HGF) in the first-trimester pregnancy serum were significantly negatively correlated with the subsequent onset of PIH. With the top 3 cytokines (IL-7, MIG, and SCF) of receiver operating characteristic (ROC) analysis, we constructed an efficient multifactor combined detection and prediction model for PIH in ART pregnancy. Classical early predictors for
INTRODUCTION

Pregnancy-induced hypertension (PIH) is one of the most common pregnancy complications, affecting 6% to 10% of pregnancies (1, 2). It is defined as a new onset of hypertension (systolic pressure ≥ 140 mmHg and/or diastolic pressure ≥ 90 mmHg) with or without proteinuria after the 20th week of gestation, which includes preeclampsia (3). PIH is characterized by maternal hypertension and multiorgan involvement resulting from systemic endothelial dysfunction, and it is also a major cause of maternal and fetal morbidity and mortality (4, 5). However, the clinical intervention of PIH stagnates in passive treatment after symptoms appear, resulting in poor prognosis. Hence, early prediction markers for PIH are of great significance for presymptomatic prophylaxis.

To date, there are several well-studied molecular markers related to placental hypoperfusion, such as placental growth factor (PIGF), soluble Fms-like tyrosine kinase-1 (sFlt-1) or vascular endothelial growth factor receptor (VEGFR) and Activin A. Although a high sFlt-1:PIGF ratio (6–8), high Activin A serum level (9) and low PIGF serum level (8) have been proven to be able to separate PIH from normotensive pregnant women, they all share low predictive accuracy in the first trimester of pregnancy (10, 11). Unfortunately, it has recently been found that the incidence of PIH in the ART population is higher than that in spontaneous pregnancy (12). Guidelines suggest that oral aspirin should be started at 11-13+6 gestational weeks in high-risk populations to achieve effective prevention (13). Therefore, it is urgent to explore biomarkers for the first-trimester prediction of PIH in the ART population.

The pathophysiological mechanisms of the development of PIH remain unclear. One of the more deeply studied mechanisms is fetal-maternal immunity (14). There is increasing evidence that PIH may originate from poor maternal immune tolerance and abnormal interactions between immune cells, trophoblast cells, and decidual stromal cells after embryo implantation. Immune cells such as natural killer cells, dendritic cells, and T regulatory lymphocytes in the decidua have impaired immune tolerance to spiral artery remodeling and the emergence of fetal trophoblasts, leading to increased placental shedding, exaggerated systemic inflammation, and subsequent endothelial dysfunction (15).

Cytokines are a family of small soluble proteins expressed by various cell and tissue types that serve as immune mediators. Their expression profiles have been used to classify immune responses and functional states of the immune system. Evidence suggests that cytokines play a crucial role in ovulation, implantation, placentation, and parturition during pregnancy (16, 17). Recent research indicates that PIH is related to immune dysregulation and that components of the immune system may interact with angiogenic and antiangiogenic factors (12). In this study, the cytokine profiles were characterized in paired PIH and normotensive women following ART treatment to determine the specific cytokine signature involved, which would be of great value to explain the occurrence of PIH after ART and provide new avenues for early diagnosis, monitoring, and intervention.

MATERIALS AND METHODS

Participants

In this study, 33 women diagnosed with PIH and 33 controls were selected from patients who underwent in vitro fertilization (IVF)/intracytoplasmic sperm injection (ICSI) cycles based on a large prospective cohort conducted at the Research Center of the Reproductive Hospital Affiliated to Shandong University from January 2018 to June 2019. All patients were <40 years and achieved singleton pregnancy by assisted reproductive technology in this pregnancy. Patients diagnosed with chronic autoimmune disease (such as systemic lupus erythematosus, thyroid autoimmunity, or antiphospholipid syndrome), preexisting secondary/essential hypertension, gestational diabetes mellitus, and other diseases that may affect the inflammatory process were excluded. Pregnancies after embryo transfer were followed by periodic reviews of electronic medical records until delivery. PIH was defined as a new onset of hypertension (systolic pressure ≥ 140 mmHg and/or diastolic pressure ≥ 90 mmHg) with or without proteinuria after the 20th week of gestation. According to clinical and laboratory evaluations, 33 patients diagnosed with PIH were included in the PIH group. To achieve a balanced baseline characteristics, propensity score matching (PSM) was applied to control for potential confounders, including age, BMI, cause of infertility, fertilization method, and embryo transfer proposal, which were weighted equally. The NC group included 33 healthy women who were matched in a 1:1 ratio to PIH based on the propensity score with a standard caliper width of 0.1. This study protocol was approved by the Ethical Committee of Reproductive Medicine of Shandong University, and all participants signed informed consent forms.

Sample Collection

At 11-13 gestational weeks following IVF/ICSI, 6 ml fasting blood samples of the participants were collected by venipuncture.
After centrifugation at 1300 × g for 10 minutes, serum samples were separated and stored at −80°C until measurement.

**ELISA Analysis**

The serum levels of Activin A, PlGF and sFLT1 were measured using commercial ELISA kits (R&D, Cat # DAC00B, Human/Mouse/Rat Activin A Quantikine ELISA Kit; Cat # DPG00, Human PLGF Quantikine ELISA Kit; Cat # DVR100C, Human VEGFR1/Flt-1 Quantikine ELISA Kit).

**Cytokine Profiling**

Individual serum samples were subjected to cytokine profile measurement by the Bio-Plex Pro Human Cytokine Screening Panel, 48-plex (BioRad). The protocol of the kit was followed for the analysis of a total of 48 cytokines, chemokines or growth factors. These included interleukin-1β (IL-1β), IL-1α, IL-1Ra, IL-2, IL-2Rα, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-12 (p40), IL-12 (p70), IL-13, IL-15, IL-16, IL-17A, IL-18, interferon-α2 (IFN-α2), interferon-γ (IFN-γ), tumor necrosis factor-α (TNF-α), tumor necrosis factor-β (TNF-β), granulocyte colony-stimulating factor (G-CSF), macrophage colony-stimulating factor (M-CSF), granulocyte-macrophage colony-stimulating factor (GM-CSF), leukemia inhibitory factor (LIF), stem cell factor (SCF), vascular endothelial growth factor (VEGF), Eotaxin, macrophage inflammatory protein-1α (MIP-1α), macrophage inflammatory protein-1β (MIP-1β), basic fibroblast growth factor (FGF basic), monocyte chemotactic protein-1 (MCP-1) or monocyte chemoattractant proteinactivating factor (MCAF), monocyte chemotactic protein-3 (MCP-3), nerve growth factor-β (β-NGF), RANTES, stromal cell derived factor-1α (SDF-1α), platelet-derived growth factor-BB (PDGF-BB), growth related oncogene-α (GRO-α), hepatocyte growth factor (HGF), interferon inducible protein-10 (IP-10), cutaneous T-cell attracting chemokine (CTACK), mifepristone (MIF), gamma-interferon-induced monokine (MIG), stem cell growth factor-β (SCGF-β), and TNF-related apoptosis-inducing ligand (TRAIL).

**Statistical Analysis**

Statistical analysis and graphical representations were completed in IBM SPSS Statistics 21 and GraphPad Prism. Normality was assessed by Kolmogorov–Smirnov tests. Normally distributed data are presented as the mean ± SEM, and the t test was used to compare the significant differences between groups. For data with a nonnormal distribution, the median with interquartile range was applied, and the Mann–Whitney U test was used for data comparison. Values of P < 0.05 were considered statistically significant. Principal component analysis (PCA), heatmap and cluster analysis, forest map, and half-violin plots were conducted using the website www.bioinformaticsm.com.cn. Univariate logistic regression analysis was performed to assess the longitudinal association between first-trimester serum cytokine levels and the occurrence of PIH. OR > 1 indicates a risk factor, and OR < 1 indicates a protective factor. Cytokines with P < 0.05 were considered to be related to the occurrence of PIH and included in Receiver operating characteristic (ROC) analysis. ROC curves were drawn to analyze the independent predictive value of specific cytokines for PIH. Additionally, IL-7, MIG, and SCF were selected to build a combined prediction model, and the combined diagnostic value of the prediction model for PIH was analyzed.

**RESULTS**

**Baseline Characteristics and Perinatal Outcomes of the Participants**

Table 1 displays the baseline characteristics and perinatal outcomes of 33 women who developed PIH in later pregnancy and their 33 matched normotensive controls (NC). The systolic pressure (NC 115.48 ± 2.15 vs. PIH 127.07 ± 2.17, P < 0.01) and mean arterial pressure (NC 85.30 ± 1.86 vs. PIH 92.66 ± 1.83, P = 0.01) before pregnancy were higher in the PIH group than in the NC group. There were no significant differences between the two groups in other prepregnancy baseline characteristics. The incidence of cesarean section was significantly higher in the PIH group than in the NC group (NC 66.70% vs. PIH 96.97%, P < 0.01). The gestational age at delivery was lower in the PIH group than in the NC group (NC 275 days vs. PIH 269 days, P = 0.02). Lower gestational age and higher cesarean section rate are related to each other and exactly reflect the characteristics of PIH.

**Classical Serum Biomarker Levels in Pregnancy After Assisted Reproductive Technology**

First, we wanted to verify whether classical serum biomarkers can distinguish PIH from NC in ART pregnancy. Activin A, PlGF and sFLT1 levels tested by ELISA and accordingly calculated sFlt1/PlGF are shown in Table 2. In the first-trimester serum of ART pregnancy, classical biomarkers appeared to have no predictive validity with no significant difference between NC and the women who developed PIH later in pregnancy.

**Cytokine Profile in First-Trimester Serum in ART Pregnancy**

To explore serum markers with potential predictive/diagnostic value for PIH in the first trimester of ART pregnancy, 48 cytokine expression levels were detected in patients with PIH and NC. PCA showed that the NC and PIH groups could be well distinguished by the first principal component (PC1), and a variety of cytokines contributed to PC1 and served as the main reason for the variation (Figure 1A). Heatmap and cluster analysis showed that the expression level of first-trimester serum cytokines in the PIH group was lower than that in the NC group (Figure 1B). In total, we found 29 differentially expressed cytokines between the two groups: IL-1β, IL-1α, IL-2, IL-2Rα, IL-4, IL-5, IL-6, IL-7, IL-9, IL-12 (p40), IL-12 (p70), IL-16, IL-17A, TNF-α, M-CSF, GM-CSF, LIF, SCF, Eotaxin, MIP-1β, FGF basic, MCP-1 (MCAF), β-NGF, SDF-1α, PDGF-BB, GRO-α, HGF, MIG, and TRAIL (Figure 1C and Table 3). Notably, compared with the NC group, the expression levels of all these cytokines were significantly lower in the PIH group (P < 0.05). In addition, we generated ROC curves for every cytokine that showed a significant difference (Supplementary Figure 1).
We further analyzed the relationship between serum cytokine levels in the first trimester of pregnancy and the occurrence of PIH in later pregnancy by logistic regression analysis. Univariate logistic regression analysis showed that 35 cytokines were associated with subsequent PIH (Figure 2A). After adjusting for the influence of prepregnancy mean arterial pressure, 17 cytokines in early pregnancy serum were found to be negatively associated with an increased risk of PIH in later pregnancy, including IL-2Rα, M-CSF, IL-6, IL-2, β-NGF, IL-7, IL-12 (p70), SCF, IL-10, IL-9, MIG, GM-CSF, LIF, IL-1α, MCP-3, IL-4, and HGF (Figure 2B).

Predictive Value of Serum Cytokines in Early Pregnancy for PIH in ART Pregnancy

We selected the above 17 cytokines related to PIH and further explored their predictive value as potential target molecules for PIH. The ROC curve analysis results are shown in Table 4. Among the above 17 PIH-related cytokines, IL-7, MIG and SCF are the top three cytokines with pronounced predictive values for PIH. We then selected IL-7, MIG, and SCF to construct a combined prediction model, which was found to have good predictive value for PIH (AUC 0.821, 95% CI: 0.718-0.924, P < 0.001) (Figure 3). The AUC of the combination of IL-7, MIG, and SCF was larger than that of every single cytokine, which proved that the combined prediction model had a better ability to predict PIH than other individual indicators. The sensitivity (true-positive rate) was 81.8%, and the specificity (true-negative rate) was 72.7%.

Relationship Between the First-Trimester Serum Cytokine Profile and Subsequent PIH

We further analyzed the relationship between serum cytokine levels in the first trimester of pregnancy and the occurrence of PIH in later pregnancy by logistic regression analysis. Univariate logistic regression analysis showed that 35 cytokines were associated with subsequent PIH (Figure 2A). After adjusting for the influence of prepregnancy mean arterial pressure, 17 cytokines in early pregnancy serum were found to be negatively associated with an increased risk of PIH in later pregnancy, including IL-2Rα, M-CSF, IL-6, IL-2, β-NGF, IL-7, IL-12 (p70), SCF, IL-10, IL-9, MIG, GM-CSF, LIF, IL-1α, MCP-3, IL-4, and HGF (Figure 2B).

### Table 1 | Baseline characteristics and perinatal outcomes of the participants.

| Prepregnancy baseline characteristics | NC (N = 33) | PIH (N = 33) | P Value |
|--------------------------------------|-------------|-------------|---------|
| Age (yr)-Mean ± SD                   | 30.21 ± 4.29 | 29.94 ± 3.75 | 0.78    |
| BMI (kg/m²)-Mean ± SD               | 25.17 ± 3.92 | 25.21 ± 3.94 | 0.97    |
| Blood pressure (mmHg)-Mean ± SD     |             |             |         |
| Systolic pressure                   | 115.48 ± 2.15 | 127.07 ± 2.17 | < 0.01* |
| Diastolic pressure                  | 70.21 ± 1.90  | 75.45 ± 1.87  | 0.06    |
| Mean arterial pressure              | 85.30 ± 1.86  | 92.68 ± 1.83  | 0.01*   |
| Primigravida-no. (%)                | 29 (87.88)   | 29 (87.88)   | > 0.99  |
| PCOS-no. (%)                        | 6 (46.2)     | 7 (53.8)     | 0.80    |
| Cause of infertility-no. (%)        |              |             | 0.96    |
| Pelvic factor                       | 10 (30.30)   | 9 (27.27)    |         |
| Male factor                         | 2 (6.06)     | 2 (6.06)     |         |
| Controlling factor                  | 21 (63.64)   | 22 (66.67)   |         |
| Endometrial preparation protocol-no. (%) |     |             |         |
| Natural cycle                       | 14 (42.42)   | 13 (39.39)   | 0.80    |
| Hormonally controlled               | 19 (57.58)   | 20 (60.61)   |         |
| Mode of fertilization-no. (%)       |              |             | 0.60    |
| IVF                                  | 21 (63.64)   | 23 (69.70)   |         |
| ICSI                                 | 12 (36.36)   | 10 (30.30)   |         |
| Perinatal outcomes                  |              |             |         |
| Delivery mode-no. (%)               |              |             | < 0.01* |
| Vaginal delivery                    | 11 (33.33)   | 1 (3.03)     |         |
| Cesarean delivery                   | 22 (66.67)   | 32 (96.97)   |         |
| Gestational age at delivery (day)-Median (P25, P75) | 275.00 (269.00,279.50) | 269.00 (263.50, 273.00) | 0.02* |
| Delivery at <37 wk-no. (%)          | 4 (12.12)    | 5 (15.15)    | > 0.99  |
| Neonatal birth weight (g)-Mean ± SD | 3319.05 ± 107.72 | 3137.19 ± 93.48 | 0.21    |
| Low birth weight infants (<2500 g)-no. (%) | 3 (9.09) | 2 (6.06)     | 0.64    |

Values are the mean ± SD/median (P25, P75) or n. (%). BMI, body mass index; PCOS, polycystic ovarian syndrome; IVF, in vitro fertilization; ICSI, intracytoplasmic sperm injection. Significant difference (P < 0.05, marked by *).

### Table 2 | Classical serum biomarker levels in pregnancy after assisted reproductive technology.

| Cytokine                  | NC (N = 33) | PIH (N = 33) | P Value |
|---------------------------|-------------|-------------|---------|
| Activin A (pg/mL)         | 2168.54 (1247.21, 3105.89) | 1917.64 (1340.23, 2754.65) | 0.621   |
| PlGF (pg/mL)              | 56.43 (43.23, 71.14)     | 44.48 (36.06, 71.30)     | 0.221   |
| sFlt1 (VEGFR) (pg/mL)     | 14401.52 (9859.96, 19593.74) | 11592.32 (7951.62, 14677.62) | 0.052   |
| sFlt1/PlGF                | 243.43 (178.78, 315.70)   | 229.72 (163.65, 314.83)   | 0.485   |

Values are median (P25, P75). PlGF, placental growth factor; sFlt1, soluble fms-like tyrosine kinase-1. Significant difference (P < 0.05).
DISCUSSION

PIH is a common pregnancy-specific complication that threatens both maternal and fetal safety (18–20). As a result of the lack of definitive therapies except for early delivery of the feto-placental unit once PIH occurs, in recent years, guidelines have placed greater emphasis on early warning, early detection and early intervention (21–23). Low-dose aspirin (LDA) is suggested to be used beginning from 11 gestational weeks for better prevention efficacy in high-risk women (13, 23–25). In regard to ART pregnancy, the evaluation and prediction methods of PIH remain to be elucidated. In this study, we brought forward the serum cytokine testing time to as early as 11-13 gestational weeks for a potential timely prediction for PIH after ART. It is worth noting that even the routinely used predictive parameters, such as Activin A, PIGF, sFlt1 and sFlt1/PIGF, were not able to differ PIH from controls in ART women, which is in line with another recent study targeting the IVF population, reporting no
significant difference in first-trimester serum Activin A between normotensive controls and pregnancies with subsequent PE/PIH (26). This is possibly ascribed to the potentially undefined population specificity, emphasizing the importance of exploring novel biomarkers for PIH in the ART population. Our results show that the cytokine profile in the first trimester of ART pregnancy demonstrated a significantly downregulated trend in ART women destined to develop PIH. We found 17 cytokines in the serum of first-trimester pregnancy related to the increased risk of PIH, including 10 interleukins (IL): IL-1α, IL-1β, IL-2, IL-4, IL-6, LIF, IL-7, IL-9, IL-10, and IL-12 (p70); three colony stimulating factors (CSF): M-CSF, GM-CSF, and SCF; two chemokines: MIG and MCP-3; and two growth factors (GF): β-NGF and HGF. Moreover, we constructed a first-trimester prediction model using IL-7, MIG, and SCF for screening out the cases at high risk of developing PIH in the early pregnancy, are significantly downregulated.

With increasing knowledge, the antecedents of poor placentaion of PIH are considered to be immunological in origin (14, 27, 28). At the maternal-fetal interface during early pregnancy, the adaptive regulation of maternal immunity is mainly characterized by the transformation of T helper type 1 (Th1) cytokines to Th2 cytokines (29, 30). Studies have shown that the shift toward Th2 may be hindered in PIH, with the cytokine profile in peripheral blood being mainly Th1 (31, 32). Our results support this theory; that is, IL-4, IL-9, and IL-10, which are related to Th2 in the serum of PIH patients during early pregnancy, are significantly downregulated.

The cytokine profile of PIH in previous studies was usually demonstrated during middle and late pregnancy, which is generally characterized by a pro-inflammatory state, showing a higher level of proinflammatory cytokines, such as IL-1, IL-6, IL-8, IL-17, and TNF-α, and a lower level of anti-inflammatory interleukins, especially IL-10, IL-33, and IL-35 (15, 33–35). Compared to the pro-inflammatory state in middle and late pregnancy, our assay in the ART population showed that proinflammatory factors such as IL-6 and IL-12 were downregulated in the first-trimester serum of PIH patients. A study in rats indicated that IL-6-mediated arterial pressure elevation is due to a response to chronic reductions in uterine perfusion pressure during pregnancy (36). Therefore, we hypothesized that in ART pregnancy, the shift toward a
proinflammatory phenotype in PIH probably does not occur in the very early stages of the disease. Similarly, the association of IL-12 serum levels with the development of PIH has been assessed in various studies, drawing no certain conclusions because of conflicting evidence (37–41). In fact, a proinflammatory phenotype is the result of compensation for insufficient immune activation and early placentation establishment in the first trimester. Therefore, most studies devoted to finding biomarkers in middle and late pregnancy to predict PIH failed to achieve good clinical application effects (42). The difference in serum proinflammatory factors in PIH patients at different pregnancy stages and the dynamic changes need to be further explored.

In spontaneous pregnancy, semen exposure between coitus and conception primes innate and adaptive immune cells for prepared maternal immune tolerance in advance of potential implantation (43–45). In contrast, ART pregnancy tends to directly transfer embryos under the premise of prohibiting sexual intercourse in the transfer cycle. This may at least partially explain why ART is associated with an increased risk of PIH and at the same time support our results that the PIH

![FIGURE 2](image)

**TABLE 4** | ROC analysis of serum cytokines in early pregnancy for PIH predictive/diagnostic value.

| Cytokines | AUC (95% CI) | P value |
|-----------|-------------|---------|
| IL-7      | 0.787 (0.676-0.899) | < 0.001 |
| MIF       | 0.786 (0.651-0.882) | < 0.001 |
| SCF       | 0.785 (0.649-0.876) | < 0.001 |
| M-CSF     | 0.759 (0.644-0.875) | < 0.001 |
| GM-CSF    | 0.759 (0.637-0.881) | < 0.001 |
| IL-2      | 0.758 (0.641-0.876) | < 0.001 |
| IL-2R    | 0.756 (0.64-0.873) | < 0.001 |
| IL-6      | 0.737 (0.617-0.857) | < 0.001 |
| IL-4      | 0.736 (0.616-0.857) | < 0.001 |
| IL-12 (p70)| 0.729 (0.607-0.852) | 0.001 |
| β-NGF     | 0.729 (0.607-0.850) | 0.001 |
| LIF       | 0.723 (0.599-0.847) | 0.002 |
| HGF       | 0.721 (0.593-0.849) | 0.002 |
| IL-1α     | 0.716 (0.693-0.839) | 0.003 |
| IL-10     | 0.697 (0.567-0.826) | 0.006 |
| MCP-3     | 0.675 (0.545-0.805) | 0.015 |
| IL-9      | 0.664 (0.533-0.796) | 0.022 |

AUC: area under the curve; CI: confidence interval. Significant difference (P < 0.05).
group showed lower serum cytokine levels than the normotensive group at 11-13 weeks gestation. In other words, inadequate tolerance induction before embryo transfer results in a universally low level of serum cytokines in the first trimester of pregnancy, which may disrupt the inflammatory process of embryo implantation and is in turn involved in the shallow invasion of trophoblasts in early PIH (46).

The present study is based on the human biobank of our large-scale assisted reproductive cohort platform and is the first to delineate the first-trimester serum cytokine profile of PIH, especially for the ART population. We found that maternal serum concentrations of multiple immune-related cytokines were significantly decreased in the first trimester of ART pregnancy with subsequent PIH. With conjoint analysis of serum IL-7, MIG and SCF, we demonstrated a first-trimester prediction model before PIH onset and subsequent compensatory response. Admittedly, the number of participants included in the current study is still limited and further research with a large sample size is needed to support our findings.

The establishment and maintenance of pregnancy poses great challenges to the maternal immune system. Uncoordinated maternal immunization, especially in the first trimester of pregnancy, will be a potential cause of a series of pregnancy complications (47, 48). Currently, immunotherapy has been applied to the treatment of a variety of reproductive diseases, such as recurrent spontaneous abortion (RSA) and recurrent implantation failure (RIF) (49), while the immunotherapy against PIH still needs a long way to go. Our study described the first-trimester serum cytokine profile in pregnancies conceived after ART and provided a better understanding of the immunological etiology and pathophysiology of PIH in the specific population. Additionally, the first-trimester immune disturbance enables early prediction, which guarantees enough time for low-dose aspirin (LDA) prevention before the appearance of hypertension symptom and hopefully may also promote novel therapeutic strategies on immune regulation and shed light on the clinical management of PIH in the ART population.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/Supplementary Material. Further inquiries can be directed to the corresponding authors.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by Ethical Committee of Reproductive Medicine of Shandong University. The patients/participants provided their written informed consent to participate in this study. Written informed consent was obtained from the individual(s) for the publication of any potentially identifiable images or data included in this article.

AUTHOR CONTRIBUTIONS

YL conceived and designed the project. XL, LG, and SZ performed the experiments and analyzed the data. XL, YC, and YN collected the clinical serum samples and information. XL, LG, SZ, SH, and ZL wrote the manuscript. YL and JY critically revised the manuscript. All authors have been involved in interpreting the data and approved the final version.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fimmu.2022.930582/full#supplementary-material
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