Journal of Reproductive Immunology 173 (2026) 104835

Contents lists available at ScienceDirect

Journal of Reproductive Immunology

journal homepage: www.elsevier.com/locate/jri

Intrauterine administration of granulocyte colony-stimulating factor promotes the transformation of endometrium immune phenotype and the onset of pregnancy in patients with recurrent implantation failures

Iryna Sudoma [a,1,2], Yana Goncharova [a,3,4,*], Boris Dons'koi [b,5,6]

[a] Clinic of Reproductive Medicine "Nadiya", Kyiv, Ukraine
[b] Victor Chernyshov" Laboratory, State Institution «Ukrainian Center of Maternity and Childhood of the National Academy of Medical Sciences of Ukraine», Kyiv, Ukraine

ARTICLE INFO

Keywords:
In vitro fertilization
Recurrent implantation failure
Immune phenotype of endometrium
Granulocyte colony-stimulating factor

ABSTRACT

Aim of the study: The aim of the study was to determine the effectiveness of intrauterine administration of granulocyte-colony-stimulating factor (G-CSF) depending on immune phenotype of endometrium in patients with recurrent implantation failures in embryo transfer programs.
Material and methods: Patients with recurrent implantation failure underwent endometrial biopsy during the implantation window in an artificial cycle before and (in some patients) after intrauterine administration of G-CSF. Pregnancy and live birth rates were determined depending on the immune phenotype of the endometrium in patients with repeated implantation failures after intrauterine administration of G-CSF in an embryo transfer program. Flow cytometry was used to count the classes and subclasses of endometrial lymphocytes in endometrial tissue samples.
Results: The endometria of patients who became pregnant as a result of embryo transfer after G-CSF irrigation of the uterine cavity showed significantly higher expression of HLA-DR on T cells and Th, and HLA-DR and CD16 on uterine natural killers (uNK). There was a high correlation between not transformed endometrium, that is, increased levels of HLA-DR (>55 %) and CD16 (>50 %) on uNK, with successful implantation after G-CSF irrigation of the uterine cavity (OR=7.700, p = 0.0174). In patients with a not transformed immune endometrial phenotype, the pregnancy (53.8 %) and live birth (46.1 %) rates were twice as high as those in patients with other variants of immune abnormalities or without immune profile changes (pregnancy and live birth rates - 19.2 %). Intrauterine G-CSF caused a significant decrease in the high expression of HLA-DR and CD16 in uNK cells without affecting other immune markers.
Conclusions: Elevated expression of HLA-DR and CD16 on uNK was associated with successful implantation after intrauterine use of G-CSF in embryo transfer program.

1. Introduction

Recurrent implantation failure (RIF) after embryo transfer remains an unresolved issue in reproductive medicine. The diagnosis of "RIF" is made, as a rule, after at least three to four transfers of good-quality embryos (Coughlan et al., 2014; Nardo et al., 2015; Bashiri et al., 2018; Shaulov et al., 2020; Mascarenhas et al., 2022; Wang et al., 2022), and according to ESHRE guideline (2023) even after the second transfer

* Corresponding author.
E-mail addresses: i.sudoma@ivf.com.ua (I. Sudoma), y.goncharova@ivf.com.ua (Y. Goncharova), boris_donskoy@ukr.net (B. Dons'koi).
[1] https://orcid.org/0000-0003-4847-9543
[2] Clinic of Reproductive Medicine "Nadiya", 03037 Ukraine, Kyiv, M. Kryvonosa str, 19A
[3] https://orcid.org/0000-0002-6776-4641
[4] Fetal Medicine Department, Clinic of Reproductive Medicine "Nadiya", 03037 Ukraine, Kyiv, M.Kryvonosa str, 19A
[5] https://orcid.org/0000-0001-9434-2826
[6] "VICTOR CHERNYSHOV" laboratory, State Institution «UKRAINIAN CENTER OF MATERNITY AND CHILDHOOD of the National Academy of Medical Sciences of Ukraine», 04050, Ukraine, Kyiv, P. Mayborody str, 8

https://doi.org/10.1016/j.jri.2026.104835
Received 10 August 2024; Received in revised form 18 December 2025; Accepted 8 January 2026
Available online 9 January 2026
0165-0378/© 2026 Elsevier B.V. All rights are reserved, including those for text and data mining, AI training, and similar technologies.

of euploid embryos (after preimplantation genetic testing for aneuploidy - PGT-A) (ESHRE Working Group on Recurrent Implantation Failure et al., 2023).

Many methods are used in clinical practice to increase endometrial receptivity, but there is a lack of evidence regarding their effectiveness (Bashiri et al., 2018; Cavalcante et al., 2020; Sfakianoudis et al., 2021; Stope et al., 2023; Kuroda, 2024). Systemic (subcutaneous or local - into the uterine cavity) administration of granulocyte colony-stimulating factor (G-CSF) is considered a promising means to improve implantation (Rutella et al., 2002; Cavalcante et al., 2015; Zeyneloglu et al., 2020; Hou et al., 2021; Kong et al., 2023; Su et al., 2024; Xie et al., 2024). G-CSF belongs to the family of colony-stimulating factors and is synthesized by many types of cells (endothelial cells, fibroblasts, macrophages, and lymphocytes), including the cells of the reproductive organs (ovary and endometrium). There is evidence that G-CSF regulates processes important for endometrial receptivity, including vascular remodeling, local immune changes, and cell adhesion (Rahmati et al., 2014).

G-CSF was first used in patients with RIF in assisted reproductive technology (ART) programs in 2000. Würfel, (2000)) showed that systemic administration of G-CSF substantially improved embryo implantation rates. Eleven years later, Gleicher et al. (2011) showed that intrauterine administration of G-CSF in four patients with thin hormone-resistant endometrium promoted its growth.

Since then, number of studies have been published on systemic or local administration of G-CSF in patients with RIF (Scarpellini, Sbracia, 2012; Barad et al., 2014; Singh et al., 2015; Aleyasin et al., 2016; Eftekhar et al., 2016; Obidniak et al., 2016; Su et al., 2024). The results of these studies are ambiguous; some show an improvement in the rates of implantation, pregnancy, and live births, whereas others do not.

In our opinion, this may be explained by the fact that the causes of implantation failure differ; therefore, there is no universal means of influence. Among the factors that can negatively affect implantation, disorders of the endometrial immune structure are prominent (Makrigiannakis et al., 2011; Wang et al., 2020; Kwak-Kim et al., 2021; Pantos et al., 2022; Von Woon et al., 2022; Chen et al., 2023; Mukherjee et al., 2023).

There is a shift from a pro-inflammatory to a regulatory phenotype in the endometrium during the implantation window, and the composition and functioning of many cells, including immune cells, change (Parham, 2004; Richman, Naftolin, 2006; Guerin et al., 2009; Saito et al., 2010; Arck, Hecher, 2013; Yang et al., 2019). One of the key cells involved in the creation of an environment tolerant to embryos, remodeling of the vascular bed, and recognition of trophoblasts are uterine natural killers (uNK) (King et al., 1989; Parham, 2004; Guerin et al., 2009; Arck, Hecher, 2013; Moffett et al., 2016; Díaz-Hernández et al., 2021; Fraser, Zenclussen, 2022). Several studies have shown that the quantity and ratio of uNK subtypes may differ between patients with RIF and healthy fertile women (Marron, Harrity, 2019; Marron et al., 2019; Von Woon et al., 2022).

We previously conducted a flow cytometry study on endometrial biopsy material from patients with RIF and fertile women who were gestational carriers. In some patients with RIF, the uNK population showed abnormally high expression of HLA-DR, CD16, CD158a, and CD335, which was not observed in gestation carriers. High expression of HLA-DR on T lymphocytes, HLA-DR, CD16, and uNK was almost always detected in the same patients (15 out of 42). We established this as a specific immune phenotype of a not transformed endometrium, that is, one in which the changes in uNK, typical for the implantation window, did not occur (Sudoma et al., 2023).

As a continuation of our previous study in the same group of women with RIF, we decided to analyze the effectiveness of intrauterine administration of G-CSF for improving embryo implantation depending on the endometrial immune phenotype and to study whether the immune endometrial profile changes under the influence of intrauterine administration of G-CSF.

The aims of our study were: 1) to investigate the pregnancy rate (PR) and live birth rate (LBR) depending on the endometrial immune phenotype in patients with RIF after intrauterine administration of G-CSF in an embryo transfer program; and 2) to determine how intrauterine administration of G-CSF affects the immune phenotype of the endometrium.

2. Material and methods

The study group (RIF) consisted of 42 women, aged 27–45 years, with repeated (at least three) failed transfers of at least two genetically tested embryos; among them, donor egg embryos were transferred to nine patients over 39 years of age. All women had previously undergone endometrial biopsy and evaluation of the immune endometrial phenotype (Sudoma et al., 2023). In 15 women, a specific, not transformed endometrial immune phenotype was observed, with high expression of HLA-DR on T lymphocytes and HLA-DR and CD16 on uNK. Reference ranges were determined on our previous published data in fertile women (oocyte donors and gestational carriers). We considered the normal values to be within 95 % confidence interval (CI) in fertile women (Chernyshov et al., 2019; Sudoma et al., 2023). The study group has been described in detail in a previous study (Sudoma et al., 2023) (these data are shown in the supplementary materials (Table 1, Table 2). Repeated mock-cycle biopsy after intrauterine G-CSF was offered to all participants; seven patients agreed and underwent the procedure; six of these seven belonged to the previously identified not transformed subgroup. Clinical parameters of these 7 women are shown in supplementary materials (Table 3). All women had vitrified euploid embryos for future transfers. PGT-A was performed using next-generation

Table 1
T lymphocytes and NK cells (%) in the endometrium of patients with RIF that became pregnant and did not become pregnant after the embryo transfer with G-CSF irrigation of the uterine cavity.

Groups and subgroups of T lymphocytes and NK cells (%)	P n = 12 Mean±SD	NP n = 27 Mean±SD	p (CI 95 %)
CD3 + (T cells) %	38.1 ± 14.5	39.8 ± 9.9	ns
% HLA DR+ in T cells	**64.9*** ±17.3	51.7 ± 18.3	**p = 0.05**
CD3 +CD4 + (T helper cells) %	16.2 ± 8.8	15.4 ± 5.5	ns
% HLA DR+ in T helper cells	**54.7*** ±16.8	40.6 ± 14.9	**p = 0.029**
CD3 +CD8 + (T cytotoxic cells) %	23.6 ± 11.3	24.6 ± 8.9	ns
% HLA DR+ in T cytotoxic cells	67.7 ± 15.7	56.4 ± 18.6	ns
% CD56 + in T cytotoxic cells	24.6 ± 18.4	20.9 ± 14.3	ns
CD56 +CD3- (NK cells) %	50.5 ± 15.4	50.7 ± 11.7	ns
% CD158a+ in NK cells	53.1 ± 18.4	40.3 ± 17.9	p = 0.051
% HLA DR+ in NK cells	**41.9*** ±21.1	25.7 ± 20.2	**p = 0.035**
% CD8 + in NK cells	**52.6*** ±18.3	40.8 ± 15.4	**p = 0.045**
% CD56 + + bright in NK cells	69.5 ± 19.9	68.0 ± 13.5	ns
CD16 + on NK	**44.1*** ±20.4	25.6 ± 18.8	**p = 0,02**
CD16 + on NK CD56 + +	**40.2*** ±22.5	22.2 ± 17.9	**p = 0.015**
CD16 + on NK CD56 +	**52.1*** ±17.2	33.8 ± 20.0	**p = 0.018**
NK CD335+	95.1 ± 2.1	90.1 ± 8.6	ns

P, patients, who became pregnant; NP, patients, who did not become pregnant; RIF, recurrent implantation failure
*p< 0.05 compared to NP group, significant difference
ns – p > 0.1
Notably, in the endometrium of patients, who became pregnant, significantly higher expression of HLA-DR on T cells and Th, and HLA-DR and CD16 on uNK was observed (Table 1). The most reliable correlations with pregnancy onset were for HLA-DR on Th cells, HLA-DR, and CD16 on uNK cells (Table 2).

Table 2
Expression of HLA-DR on Th, HLA-DR, CD16 on uNK in the endometrium of patients who became pregnant and did not become pregnant as a result of embryo transfer after G-CSF irrigation of the uterine cavity.

Group	ET result	%CD16 + on uNK bright (CD3-CD56 ++) (>15)	%CD16 + on uNK		%HLA-DR on uNK		%HLA-DR on Th (CD3 +CD4 +) (>66)
			(>20)	(>55)	(>16)	(>50)	
G-CSF treatment	P (n = 12)	(10/12) 83.3 %	(9/12) 75 %	(5/12) 41.6 %	(9/12) 75 %	(7/12) 58.3 %	(8/11) 72.7 %
	NP (n = 27)	(13/27) 48.1 %	(12/27) 44.4 %	(3/27) 11.1 %	(12/26) 46.1 %	(4/26) 15.3 %	(7/27) 25.9 %
Significance for ET success		OR= 5.385 p = 0.0410 95 %CI (0.9877–29.355)	OR= 3.750 p = 0.0768 95 %CI (0.8272–17.001)	OR= 5.714 p = 0.0433 95 %CI (1.085–30.084)	OR= 3.500 p = 0.0939 95 %CI (0.7674–15.964)	OR= 7.350 p = 0.0183 95 %CI (1.531–35.292)	OR= 7.619 p = 0.0117 95 %CI (1.566 –37.065)

ET, embryo transfer; G-CSF, granulocyte colony-stimulating factor; NP, did not become pregnant; P, became pregnant; uNK, uterine natural killer; OR, odds ratio

sequencing.

2.1. Preimplantation genetic testing -PGT-A

The trophectodermal biopsy for preimplantation genetic testing (PGT-A) was performed on the blastocyst stage embryos before vitrification. DNA was extracted and amplified using the EmbryoMap Sample Prep Kit (Vitrolife, Sweden AB). Next-generation sequencing (NGS) was performed with the Illumina MiSeq Reagent Kit v3. The sequencing data were analyzed using eMap software, and embryos were classified as euploid, aneuploid, or mosaic according to established diagnostic thresholds.

2.2. Embryo transfer protocol

Cryopreserved and thawed embryos were transferred at least one menstrual cycle after endometrial biopsy. From the third day of the cycle, estrogens were prescribed: estradiol valerate (Proginova Delpharm Lille, France for Schering AG, Germany; 2 mg) at a dose of 6 mg per day. On days 8–11 of estrogen supplementation, an ultrasound examination was performed to assess the thickness of the endometrium. When the endometrium reached at least 7 mm, dydrogesterone (Dufaston, Solvay, Netherlands; 10 mg) was administered daily at a dose of 60 mg. At the beginning of dydrogesterone intake (day 1 or 2), G-CSF (Filstim 0.48 mg, 48 M IU, 1.6 mL, Biopharma, Ukraine) was introduced into the uterus with a transfer catheter (COOK, Guardia, Medical, USA). Embryo(s) transfer was performed on the day 7 of dydrogesterone administration. Medical support with dydrogesterone and estradiol valerate at the same doses was continued for another 14 days after transfer. Subsequently, chorionic gonadotropin testing was done; in the case of a positive test, hormonal support continued for another 14 days, then an ultrasound examination was performed to detect the presence, number, and location of gestational sacs. Pregnant women were referred for further follow-up, and data concerning pregnancy course and outcomes were monitored and recorded.

Embryo transfer was performed in 39 women, three patients dropped out of the study at this stage for personal reasons. Previously vitrified and thawed blastocyst stage euploid embryos of good and excellent quality were transferred. In most cases, single embryo transfer was performed, in 5 cases – double embryo transfer was done. In total 44 embryos were transferred. Blastocysts were scored according to Gardner and Schoolcraft scoring system. Only high-quality blastocysts were vitrified: A-B (Gardner, Schoolcraft, 1999). Vitrification and thawing of embryos were performed by CryoTop method by M. Kuwayama using the Cryotech vitrification and warming kit (Kuwayama, 2007). For embryo transfer we used catheters Guardia TM Access, Cook Medical, Limerick, Ireland.

2.3. Protocol of endometrial preparation before biopsy. G-CSF administration procedure

Seven patients agreed to undergo a repeated biopsy of the endometrium in a mock cycle with an additional instillation of G-CSF. This procedure was performed for at least one menstrual cycle after the previous biopsy, and embryo transfer was performed for at least one menstrual cycle later.

The protocol of endometrial preparation for biopsy was identical to that used for embryo transfer and close to that used for the previous biopsy: estrogens were administered from the third day of the cycle: estradiol valerate (Proginova; 2 mg) at a dose of 6 mg per day. When the endometrium reached at least 7 mm, dydrogesterone (Dufaston; 10 mg) was administered at a daily dose of 60 mg. At the beginning of dydrogesterone admission (day 1 or 2), G-CSF (Filstim 0.48 mg, 48 M IU, 1.6 mL, Biopharma, Ukraine) was introduced into the uterine cavity through a catheter for embryo transfer. The patient remained supine for 5 min to minimize leakage. The catheter was withdrawn immediately after instillation. Pipelle biopsy of the endometrium was performed on the day 7–8 of progesterone intake.

2.4. Endometrial lymphocytes study

The method of researching the immune phenotype of the endometrium at repeated biopsy in a mock cycle with the instillation of G-CSF was identical to that used previously (Sudoma et al., 2023). The endometrial cell isolation method has been previously described (Chernyshov et al., 2019). Endometrial tissue samples were gently washed three times in phosphate-buffered saline (PBS). A 1-mL pipette tip was cut at the edge so that the diameter of the hole was 4 mm and endometrial tissue was washed 10–15 times by pipetting in cool (+4 to +9 °C) PBS to obtain free endometrial cells in PBS solution. The next pipette tip had a hole diameter of 2–3 mm and the tissue pieces were pipetted again. In the last step of pipetting, the tip hole was 1–2 mm and pieces were pipetted without resistance; that is, every next step was performed with a smaller diameter of the tip hole. Between pipetting steps, the endometrium was washed with cold PBS using centrifugation. Endometrial tissue was disaggregated and flake-shaped. PBS (7 mL; +4 °C) was added, mixed with the obtained suspension, and left for 3–5 min for sedimentation of large aggregates. The cells were then washed twice with 10 mL of PBS. The cell pellet was resuspended in PBS (0.5 mL) containing 10 % of the patient serum. The final cell suspension (80 μL) was analyzed by three-color flow cytometry after staining with 5 μL monoclonal antibodies conjugated with fluorochromes FITC, PE, and PE-Cy 5 (BD Biosciences, USA) in the following combinations: CD158aFITC/CD56PE/CD3PECy5, CD8FITC/CD56PE/CD3PECy5, HLADRFITC/CD56PE/CD3PECy5, HLADRFITC/CD8PE/CD3PECy5, HLADRFITC/CD4PE/CD3PECy5, CD16FITC/CD56PE/CD3PECy5, and CD3/CD335/CD56 (BD Biosciences). Samples were vortexed for 30 s with 0.5 mL Lysing solution (BD Biosciences) and washed with 5 mL of

PBS. The samples were analyzed using a FACScan flow cytometer (BD Biosciences). In addition, the samples were incubated with SimulTEST control (IgG1-FITC + IgG2-PE) as a background control. The gating strategy is illustrated in Supplementary Fig. A, B.

2.5. Statistical analysis

Statistical analysis was performed using InStat software for Windows (version 3.0, Graph Pad Software Inc., San Diego, CA, USA). The two-sided p was calculated using Fisher's Exact Test, 95 % CI, using the approximation of Woolf. Differences with $p < 0.05$ were considered statistically significant.

The study was approved by the Ethics Committee of Nadiya Clinic (Protocol № 3–18 of the meeting of Ethics Committee at LLC "Clinic of Reproductive Medicine "Nadiya" from January 26, 2018). All women were informed of all stages of the study and provided written informed consent for participation.

3. Results

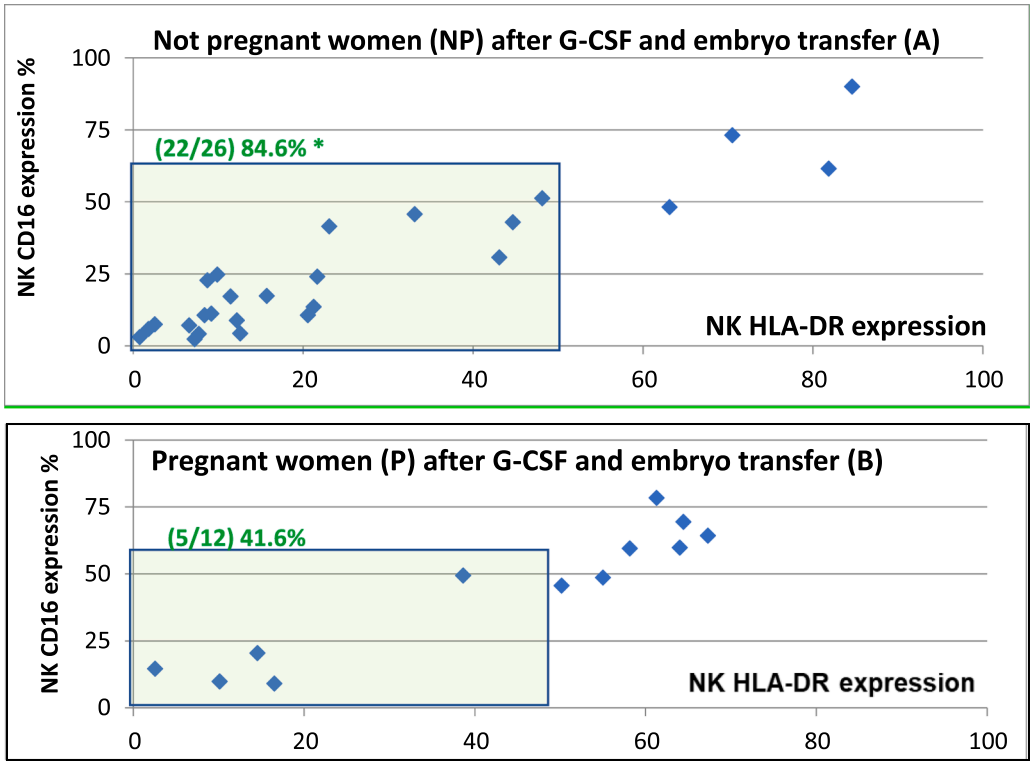
Transfer of the thawed embryo/embryos was performed in 39 women. A clinical uterine pregnancy occurred in 12 patients (PR, 30.7 %; implantation rate (IR), 29.5 %). There were two first-trimester miscarriages. Another two women underwent implantation in the fallopian tube and surgery. Eleven live children were born, including one twin pregnancy (LBR, 28.2 %).

Subpopulations of lymphocytes in the endometrium of the subgroups of women who became pregnant (P) and those who did not become pregnant (NP) after irrigation of the uterine cavity with G-CSF before embryo transfer are presented in Table 1.

As previously mentioned, high levels of HLA-DR on Th, HLA-DR, and CD16 on uNK, that is, the not transformed immune phenotype of the endometrium, were detected in 15 women. Of these women, embryo transfer was carried out in 13 patients, seven of whom became pregnant (uterine pregnancy), (PR, 53.8 %), five of whom had term delivery, including one twin pregnancy, and gave birth to healthy children (LBR, 46.1 %). Two women had a miscarriage. Among the remaining patients (those who did not have the above-mentioned immune accentuations, namely high HLA-DR, and CD16 on uNK) (26 women), uterine pregnancy occurred in only five cases. All of them had term delivery, and healthy children were born (PR, LBR, 19.2 %). There was no significant difference for PR ($p = 0.062$, OR= 4.900, CI95 %: 1.134–21.169) and LBR ($p = 0.13$, OR=3.6, CI95 %: 0.8333–15.553) because of the small number of patients in the study; however, a trend toward better outcomes in the group with a not transformed endometrial immune phenotype (with high expression of HLA-DR and CD16 on uNK) was evident. There was a strong correlation between increased levels of HLA-DR and CD16 in uNK cells (>55 % HLA-DR+, >50 % CD16 +) and successful implantation after G-CSF irrigation of the uterine cavity (OR=7.7, $p = 0.0174$; Fig. 1).

In seven women (six of them were with not transformed endometrium), we obtained biopsy material from the endometrium for research after G-CSF introduction into the uterus. The levels of most of the studied endometrial lymphocyte populations did not change significantly after intrauterine infusion of G-CSF. However, in patients with high expression of HLA-DR and CD16 on uNK cells, a decrease in these markers was observed (six patients), and in one woman with lower expression of these markers, they did not change much or even increased (Fig. 2). High expression of HLA-DR in T lymphocytes in some cases did not change at all; in other cases, it changed towards an insignificant increase or decrease.

Of the 6 women who experienced a decrease in HLA-DR and CD16 expression levels on uNK after G-CSF administration, 4 became pregnant after subsequent embryo transfer. In 2 patients, embryo transfer was not performed (these were two of the three women who dropped out of the study).



** Fisher's Exact Test OR=7.700; 95%CI: 1.608 to 36.874; The two-sided p=0.0174*

Fig. 1. Distribution of CD16 and HLA-DR expression levels in patients with subsequent embryo transfer failures (A) and pregnancy (B) after G-CSF use. * Fisher's Exact Test OR= 7.700; 95 %CI: 1.608–36.874; The two-sided p = 0.0174.

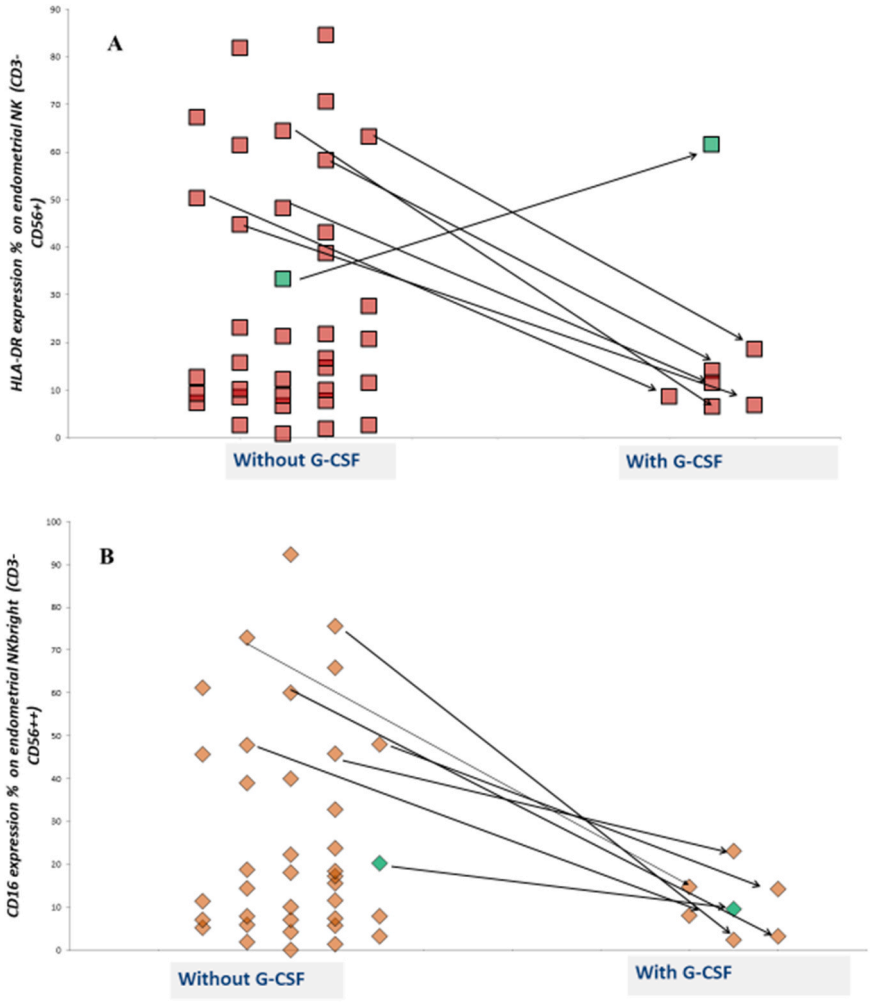


Fig. 2. Expression of HLA-DR (A) and CD16 (B) on uNK in cycles without and with G-CSF use.

4. Discussion

Intrauterine or subcutaneous G-CSF is considered a promising treatment for implantation failure in ART programs (Eftekhar et al., 2018). More than 2000 studies on that topic were published during last 25 years. Mostly the women with at least two to three unexplained implantation failures were included. Typically, cycles using G-CSF were compared with a control group (no treatment or placebo) and/or another intervention. It was found, that G-CSF administration improved the clinical PR, IR, biochemical PR, and LBR in women with at least one implantation failure (Su Q et al., 2024). The reasons for this improvement may include induction of local immune regulation of the endometrium, embryo adhesion and implantation, proliferation of trophoblasts and endometrial vascular remodeling by G-CSF (Su Q et al., 2024). However, there are still no exact indications for who may benefit from G-CSF.

Studies on the effectiveness of G-CSF in patients with RIF and habitual miscarriage typically use doses of 100–300 μg for intrauterine administration and 300 μg subcutaneously as a single dose or 60–100 μg subcutaneously daily from the day of embryo transfer till the hCG test or 9 weeks of pregnancy (total course dose - up to 1400 μg) (Hou Z et al., 2021; Su Q et al., 2024). In a meta-analysis by Su Q et al. (2024), it was shown that at a dose of more than 150 μg G-CSF increases PR and IR. Since the use of this drug in reproductive medicine is “off-label” and there are no established recommendations for dosage, time, and method of administration, we chose a domestic drug that has 480 μg (48 M IU) for intrauterine infusion. This dose is higher than that proposed for intrauterine administration, but lower than that used for the whole course of subcutaneous administration. This dose was chosen taking into account the dose-dependent effect on PR and IR, as well as the fact that when administered into the uterus, part of the active substance is lost due to leakage and incomplete absorption.

In a previous study on the endometrial immune phenotype, we found several peculiar immune features in most patients with RIF (Sudoma et al., 2023). In a quarter of the patients with RIF, the NK cell population had abnormally high expression of HLA-DR and CD16, which was not observed in fertile women. These changes correlated with each other and appeared in the same patient. In this study, we found, that high levels of HLA-DR and CD16 on uNK cells were significantly more frequent in women who became pregnant after intrauterine administration of G-CSF during the embryo transfer cycle. In this group of patients, there was a trend towards higher PR and LBR after intrauterine G-CSF use.

The phenomenon of an increase in the percentage of CD16 positive uNK is relatively well-known and is associated with an unfavorable reproductive prognosis. An increase in the proportion of CD16 +CD56dim cells and a decrease in the population of CD16 −CD56bright cells were found in women with habitual miscarriage (Lachapelle et al., 1996; Quenby et al., 1999; Giuliani et al., 2014), and miscarriage after in vitro fertilization (Fukui et al., 1999). Marron and Harrity (2019) found an increase in the proportion of "blood" type NK, i.e. CD16 +CD56 + , exactly in the subgroups of patients with RIF and other reproductive disorders. Junovich et al. (2013) showed an increase in the proportion of CD16 positive uNK in women with

infertility, which was correlated with a decrease in IL-6 and VEGF levels in the endometrium. It is believed that CD16-positive uNK cells can produce pro-inflammatory cytokines and cytotoxic factors (Sharma, 2014; Huber et al., 2021). Therefore, as their population increases, they create an unfavorable environment for implantation and pregnancy development.

Little is known about the function of HLA-DR-positive NK cells in the endometrium. HLA-DR belongs to the second class of the major histocompatibility complex and is mainly expressed by antigen-presenting cells. However, a small proportion of the peripheral blood and tissue NK cells also express this marker. The expansion of HLA-DR-positive NK cells is associated with inflammation and autoimmune processes (Yano et al., 1996; Hanna et al., 2004; Erokhina et al., 2021) In our previous study, we showed that the implantation window formation in healthy women was associated with decreased HLA-DR expression in endometrial NK (Anoshko et al., 2023). We also showed that elevated HLA-DR expression on peripheral blood NK cells corelated with NK cell accumulation (Dons'koi, 2023). Thus, it can be hypothesized that the increased number of HLA-DR positive uNK in women with RIF may favor a proliferative (not transformed), pro-inflammatory, "threat-fighting" phenotype. Our previous studies on the oocyte donor endometrium in cycles of controlled ovarian stimulation showed that such immune structure was often found in healthy women on the day of oocyte retrieval, but not on the days corresponding to the implantation window (Chernyshov et al., 2019). It can be assumed that the endometrium, from the moment of ovulation (or egg collection) to the implantation window, evolves and transforms, and the percentage of HLA-DR-positive NK cells decreases.

Thus, in 6 patients with a combination of signs, such as an increase in the proportion of CD16-positive and HLA-DR-positive uNK, confirming not transformed, pro-inflammatory endometrium, G-CSF corrects these immune defects, thereby, possibly, contributes to embryo implantation. However, the specific mechanism underlying the corrective effects of G-CSF has not yet been elucidated.

Although there is much evidence supporting the importance of G-CSF in reproduction, we found only a small number of studies addressing aspects of this cytokine physiology other than neutrophil/granulocyte cell line stimulation. G-CSF could be found in trophoblastic cells and endometrial glandular cells, G-CSF receptor is expressed on leukocytes, trophoblasts, macrophages of the decidual membrane, indicating the importance of this cytokine in implantation (Uzumaki et al.,1989; Salmassi et al., 2004). G-CSF could also recruit dendritic cells, activate Th2 cells, Tregs and promote the secretion of Th2 cytokines (Moldenhauer et al., 2010). Ding et al. (2021) demonstrated that G-CSF derived from M2 macrophage could promote trophoblasts invasion and migration through activating PI3K/AKT/Erk1/2 pathway. Tanaka et al. (2000) showed that human endometrial stromal cells produce a small amount of G-CSF, but after stimulation of the cell culture with a derivative of cyclic adenosine monophosphate (8-bromo-cyclic adenosine 3′,5′-monophosphate-8-Br-CAMP) which contributes to the decidualization of stromal cells, their G-CSF production substantially increases. When cells were stimulated with 8-Br-CAMP and G-CSF, the amount of prolactin (a decidualisation marker) produced by these cells significantly increased. Therefore, it can be assumed that a similar phenomenon occurs in vivo in the endometrium. Progesterone and estradiol contribute to the differentiation of stromal cells into decidual stromal cells, which begin to produce more G-CSF, which in turn enhances the process of decidualization. In a favorable environment created by decidual stromal cells, the expansion of leukocyte populations, including uNK, opens the "immune window of implantation". In some women with RIF immune reconstruction does not occur in the full extent. One form of incomplete reconstruction is not transformed immune phenotype of the endometrium. Additional administration of G-CSF can enhance the decidualization of stromal cells, Treg recruitment, Th2 type activation, production of regulatory cytokines, which contributes to the "maturation" and transformation of uNK. It was found, that G-CSF upregulate m-RNA expression of Plasminogen Activator Urokinase Receptor (PLAUR or uPAR) expression by uNK (Rahmati et al., 2014). So, it could regulate trophoblast invasion via the uPAR system, a cascade of proteolytic activities, enabling cell migration (Rahmati et al., 2014).

Based on our data, patients with not transformed immune phenotype of the endometrium may benefit from the use of intrauterine G-CSF to increase the probability of pregnancy after embryo transfer. In our previous study, we showed that high levels of HLA-DR and CD16 on NK cells were extremely unfavorable for implantation and associated with RIF (Sudoma et al., 2023). However, it seems that these unfavorable accentuations are sensitive to G-CSF therapy and may be a target marker for treatment.

As far as post-G-CSF biopsies were obtained predominantly in patients with the not transformed phenotype (6/7), we cannot determine whether G-CSF would alter immune markers in RIF patients without this phenotype. The absence of post-treatment sampling in the larger not transformed subgroup limits the conclusions about the mechanisms in those patients. Therefore, we cannot explain why these RIF patients do not respond to G-CSF treatment. We suppose that they may lack the target marker or have a different cause of implantation failure.

Certain limitations of this investigation, such as small sample size and lack of the control group are due to the fact, that this work is a continuation of our previous study in which the immune phenotype of the endometrium was investigated. Thus, only the women who participated in the previous study were included. Larger trials (blind, randomized, prospective) are required to work out the standardization of endometrial immune phenotype criteria and determination of optimal modes of G-CSF use.

5. Conclusions

1. Increased levels of HLA-DR and CD16 expressing uNK are the markers of non-receptive endometrium and are found in patients with RIF. But, at the same time, elevated expression of HLA-DR and CD16 on uNK was reliably associated with successful implantation after intrauterine infusion of G-CSF in embryo transfer program. In the group of patients with not transformed immune endometrial phenotype, the PR (53.8 %) and the LBR (46.1 %) were more than twice as high compared to those in patients with other variants of immune abnormalities or without immune profile changes (PR and LBR, 19.2 %).
2. Intrauterine G-CSF causes a significant decrease of the expression of HLA-DR and CD16 on uNK, without substantial changes of other immune markers.
3. Elevated HLA-DR and CD16 on NK, as markers of not transformed endometrium, may identify a subgroup of RIF patients treatable with intrauterine G-CSF, but prospective validation with prespecified thresholds, standard procedures, and control arms is required before clinical adoption.

CRediT authorship contribution statement

Iryna Sudoma: Writing – original draft, Project administration, Methodology, Formal analysis, Data curation, Conceptualization. **Dons'koi Boris:** Visualization, Methodology, Investigation, Formal analysis, Data curation. **Yana Goncharova:** Writing – review & editing.

Ethical

The authors confirm that the ethical policies of the journal, as noted on the journal's author guidelines page, have been adhered to and the appropriate ethical review committee approval has been received. We confirm that neither the manuscript nor any parts of its content are currently under consideration or published in another journal. All authors have approved the manuscript and agree with its submission.

Declaration of Competing Interest

The authors have no conflict of interest to declare.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.jri.2026.104835.

References

Aleyasin, A., Abediasl, Z., Nazari, A., Sheikh, M., 2016. Granulocyte colony-stimulating factor in repeated IVF failure, a randomized trial. Reproduction 151 (6), 637–642. https://doi.org/10.1530/REP-16-0046.

Anoshko, Y., Dons'koi, B., Sudoma, I., Khazhylenko, K., Zabara, D., Goncharova, Y., 2023. Changes in the immunophenotype of endometrium during implantation window receptivity formation in healthy fertile women. Placenta 143, 62–68. https://doi.org/10.1016/j.placenta.2023.10.002.

Arck, P.C., Hecher, K., 2013. Fetomaternal immune cross-talk and its consequences for maternal and offspring's health. Nat. Med 19 (5), 548–556. https://doi.org/10.1038/nm.3160.

Barad, D.H., Yu, Y., Kushnir, V.A., Shohat-Tal, A., Lazzaroni, E., Lee, H.J., et al., 2014. A randomized clinical trial of endometrial perfusion with granulocyte colony-stimulating factor in in vitro fertilization cycles: impact on endometrial thickness and clinical pregnancy rates. Fertil. Steril. 101 (3), 710–715. https://doi.org/10.1016/j.fertnstert.2013.12.016.

Bashiri, A., Halper, K.I., Orvieto, R., 2018. Recurrent implantation failure-update overview on etiology, diagnosis, treatment and future directions. Reprod. Biol. Endocrinol. 16 (1), 121. https://doi.org/10.1186/s12958-018-0414-2.

Cavalcante, M.B., Cavalcante, C.T.M.B., Sarno, M., Barini, R., 2020. Intrauterine perfusion immunotherapies in recurrent implantation failures: systematic review. Am. J. Reprod. Immunol. 83 (6), e13242. https://doi.org/10.1111/aji.13242.

Cavalcante, M.B., Costa Fda, S., Barini, R., Araujo Júnior, E., 2015. Granulocyte colony-stimulating factor and reproductive medicine: a review. Iran. J. Reprod. Med 13 (4), 195–202. PMID: 26131007; PMCID: PMC4475767.

Chen, Z., Huang, J., Kwak-Kim, J., Wang, W., 2023. Immune checkpoint inhibitors and reproductive failures. J. Reprod. Immunol. 156, 103799. https://doi.org/10.1016/j.jri.2023.103799.

Chernyshov, V.P., Dons'koi, B.V., Sudoma, I.O., Goncharova, Y.O., 2019. Comparison of T and NK lymphocyte subsets between human endometrial tissue and peripheral blood. Cent. Eur. J. Immunol. 44 (3), 316–321. https://doi.org/10.5114/ceji.2019.89610.

ESHRE Working Group on Recurrent Implantation Failure, Cimadomo, D., de Los Santos, M.J., Griesinger, G., Lainas, G., Le Clef, N., McLernon, D.J., et al., 2023. ESHRE good practice recommendations on recurrent implantation failure. Jun 15 Hum. Reprod. Open 2023 (3), hoad023. https://doi.org/10.1093/hropen/hoad023.

Coughlan, C., Ledger, W., Wang, Q., Liu, F., Demirol, A., Gurgan, T., et al., 2014. Recurrent implantation failure: definition and management (Jan). Reprod. Biomed. Online 28 (1), 14–38. https://doi.org/10.1016/j.rbmo.2013.08.011.

Díaz-Hernández, I., Alecsandru, D., García-Velasco, J.A., Domínguez, F., 2021. Uterine natural killer cells: from foe to friend in reproduction. Jun 22 Hum. Reprod. Update 27 (4), 720–746. https://doi.org/10.1093/humupd/dmaa062.

Ding, J., Yang, C., Zhang, Y., Wang, J., Zhang, S., Guo, D., et al., 2021. M2 macrophage-derived G-CSF promotes trophoblasts EMT, invasion and migration via activating PI3K/Akt/Erk1/2 pathway to mediate normal pregnancy (Feb). J. Cell Mol. Med 25 (4), 2136–2147. https://doi.org/10.1111/jcmm.16191.

Dons'koi, B.V., 2023. Theory of immune accentuations: formation of accentuated immunophenotype. Likars'ka Sprav. (2), 5–13. https://doi.org/10.31640/LS-2023-2-01.

Eftekhar, M., Miraj, S., Farid Mojtahedi, M., Neghab, N., 2016. Efficacy of Intrauterine infusion of granulocyte colony stimulating factor on patients with history of implantation failure: a randomized control trial. Int J. Reprod. Biomed. 14 (11), 687–690 (Nov).

Eftekhar, M., Naghshineh, E., Khani, P., 2018. Role of granulocyte colony-stimulating factor in human reproduction. Jan 29 J. Res Med Sci. 23, 7. https://doi.org/10.4103/jrms.JRMS_628_17.

Erokhina, S.A., Streltsova, M.A., Kanevskiy, L.M., Grechikhina, M.V., Sapozhnikov, A.M., Kovalenko, E.I., 2021. HLA-DR-expressing NK cells: effective killers suspected for antigen presentation (Feb). J. Leukoc. Biol. 109 (2), 327–337. https://doi.org/10.1002/JLB.3RU0420-668RR.

Fraser, R., Zenclussen, A.C., 2022. Killer Timing: The Temporal Uterine Natural Killer Cell Differentiation Pathway and Implications for Female Reproductive Health. Jun 27 Front Endocrinol. (Lausanne) 13, 904744. https://doi.org/10.3389/fendo.2022.904744.

Fukui, A., Fujii, S., Yamaguchi, E., Kimura, H., Sato, S., Saito, Y., 1999. Natural killer cell subpopulations and cytotoxicity for infertile patients undergoing in vitro fertilization (Jun). Am. J. Reprod. Immunol. 41 (6), 413–422. https://doi.org/10.1111/j.1600-0897.1999.tb00456.x.

Gardner, D.K., Schoolcraft, W.B., 1999. Culture and transfer of human blastocysts (Jun). Curr. Opin. Obstet. Gynecol. 11 (3), 307–311. https://doi.org/10.1097/00001703-199906000-00013.

Giuliani, E., Parkin, K.L., Lessey, B.A., Young, S.L., Fazleabas, A.T., 2014. Characterization of uterine NK cells in women with infertility or recurrent pregnancy loss and associated endometriosis (Sep). Am. J. Reprod. Immunol. 72 (3), 262–269. https://doi.org/10.1111/aji.12259.

Gleicher, N., Vidali, A., Barad, D.H., 2011. Successful treatment of unresponsive thin endometrium (May). Fertil. Steril. 95 (6). https://doi.org/10.1016/j.fertnstert.2011.01.143.

Guerin, L.R., Prins, J.R., Robertson, S.A., 2009. Regulatory T-cells and immune tolerance in pregnancy: a new target for infertility treatment? (Sep-Oct). Hum. Reprod. Update 15 (5), 517–535. https://doi.org/10.1093/humupd/dmp004.

Hanna, J., Gonen-Gross, T., Fitchett, J., Rowe, T., Daniels, M., Arnon, T.I., et al., 2004. Novel APC-like properties of human NK cells directly regulate T cell activation (Dec). J. Clin. Invest 114 (11), 1612–1623. https://doi.org/10.1172/JCI22787.

Hou, Z., Jiang, F., Yang, J., Liu, Y., Zha, H., Yang, X., et al., 2021. What is the impact of granulocyte colony-stimulating factor (G-CSF) in subcutaneous injection or intrauterine infusion and during both the fresh and frozen embryo transfer cycles on recurrent implantation failure: a systematic review and meta-analysis? Aug 13 Reprod. Biol. Endocrinol. 19 (1), 125. https://doi.org/10.1186/s12958-021-00810-4.

Huber, W.J., 3rd, Sauerbrun-Cutler, M.T., Krueger, P.M., Sharma, S., 2021. Novel predictive and therapeutic options for better pregnancy outcome in frozen embryo transfer cycles (Apr). Am. J. Reprod. Immunol. 85 (4), e13300. https://doi.org/10.1111/aji.13300.

Junovich, G., Azpiroz, A., Incera, E., Ferrer, C., Pasqualini, A., Gutierrez, G., 2013. Endometrial CD16(+) and CD16(-) NK cell count in fertility and unexplained infertility (Sep). Am. J. Reprod. Immunol. 70 (3), 182–189. https://doi.org/10.1111/aji.12132.

King, A., Wellings, V., Gardner, L., Loke, Y.W., 1989. Immunocytochemical characterization of the unusual large granular lymphocytes in human endometrium throughout the menstrual cycle (Mar). Hum. Immunol. 24 (3), 195–205. https://doi.org/10.1016/0198-8859(89)90060-8.

Kong, X., Tang, G., Liu, Y., Zheng, Z., Li, Y., Yan, F., 2023. Efficacy of intrauterine infusion therapy before embryo transfer in recurrent implantation failure: A systematic review and network meta-analysis (Mar). J. Reprod. Immunol. 156, 103819. https://doi.org/10.1016/j.jri.2023.103819.

Kuroda, K., 2024. Management strategies following implantation failure of euploid embryos. Apr 8 Reprod. Med Biol. 23 (1), e12576. https://doi.org/10.1002/rmb2.12576.

Kuwayama, M., 2007. Highly efficient vitrification for cryopreservation of human oocytes and embryos: the Cryotop method. Jan 1 Theriogenology 67 (1), 73–80. https://doi.org/10.1016/j.theriogenology.2006.09.014.

Kwak-Kim, J., Sung, N., Saab, W., Fukui, A., 2021. Introduction of the special issue, "Clinical reproductive immunology" (Apr). Am. J. Reprod. Immunol. 85 (4), e13415. https://doi.org/10.1111/aji.13415.

Lachapelle, M.H., Miron, P., Hemmings, R., Roy, D.C., 1996. Endometrial T, B, and NK cells in patients with recurrent spontaneous abortion. Altered profile and pregnancy outcome. J. Immunol. 156 (10), 4027–4034. May 15.

Makrigiannakis, A., Petsas, G., Toth, B., Relakis, K., Jeschke, U., 2011. Recent advances in understanding immunology of reproductive failure (Jun). J. Reprod. Immunol. 90 (1), 96–104. https://doi.org/10.1016/j.jri.2011.03.006.

Marron, K., Harrity, C., 2019. Endometrial lymphocyte concentrations in adverse reproductive outcome populations (May). J. Assist Reprod. Genet 36 (5), 837–846. https://doi.org/10.1007/s10815-019-01427-8.

Marron, K., Walsh, D., Harrity, C., 2019. Detailed endometrial immune assessment of both normal and adverse reproductive outcome populations (Feb). J. Assist Reprod. Genet 36 (2), 199–210. https://doi.org/10.1007/s10815-018-1300-8.

Mascarenhas, M., Jeve, Y., Polanski, L., Sharpe, A., Yasmin, E., Bhandari, H.M., 2022. British fertility society. Management of recurrent implantation failure: British fertility society policy and practice guideline (Dec). Hum. Fertil. (Camb.) 25 (5), 813–837. https://doi.org/10.1080/14647273.2021.1905886.

Moffett, A., Chazara, O., Colucci, F., Johnson, M.H., 2016. Variation of maternal KIR and fetal HLA-C genes in reproductive failure: too early for clinical intervention (Dec). Reprod. Biomed. Online 33 (6), 763–769. https://doi.org/10.1016/j.rbmo.2016.08.019.

Moldenhauer, L.M., Keenihan, S.N., Hayball, J.D., Robertson, S.A., 2010. GM-CSF is an essential regulator of T cell activation competence in uterine dendritic cells during early pregnancy in mice. Dec 1 J. Immunol. 185 (11), 7085–7096. https://doi.org/10.4049/jimmunol.1001374.

Mukherjee, N., Sharma, R., Modi, D., 2023. Immune alterations in recurrent implantation failure (Feb). Am. J. Reprod. Immunol. 89 (2), e13563. https://doi.org/10.1111/aji.13563.

Nardo, L.G., El-Toukhy, T., Stewart, J., Balen, A.H., Potdar, N., 2015. British Fertility Society Policy and Practice Committee: adjuvants in IVF: evidence for good clinical practice (Mar). Hum. Fertil. 18 (1), 2–15. https://doi.org/10.3109/14647273.2015.985454.

Obidniak, D., Gzgzyan, A., Dzhemlikhanova, L., Feoktistov, A., 2016. Effect of colony-stimulating growth factor on outcome of frozen-thawed embryo transfer in patients with repeated implantation failure. Fertil. Steril. 106 (3), e134–e135. https://doi.org/10.1016/j.fertnstert.2016.07.401.

Pantos, K., Grigoriadis, S., Maziotis, E., Pistola, K., Xystra, P., Pantou, A., et al., 2022. The Role of interleukins in recurrent implantation failure: a comprehensive review of the literature. Feb 16 Int. J. Mol. Sci. 23 (4), 2198. https://doi.org/10.3390/ijms23042198.

Parham, P., 2004. NK cells and trophoblasts: partners in pregnancy. Oct 18 J. Exp. Med 200 (8), 951–955. https://doi.org/10.1084/jem.20041783.

Quenby, S., Bates, M., Doig, T., Brewster, J., Lewis-Jones, D.I., Johnson, P.M., et al., 1999. Pre-implantation endometrial leukocytes in women with recurrent

miscarriage (Sep). Hum. Reprod. 14 (9), 2386–2391. https://doi.org/10.1093/humrep/14.9.2386.
Rahmati, M., Petitbarat, M., Dubanchet, S., Bensussan, A., Chaouat, G., Ledee, N., 2014. Granulocyte-colony stimulating factor related pathways tested on an endometrial ex-vivo model. Oct 2 PLoS One 9 (9), e102286. https://doi.org/10.1371/journal.pone.0102286.
Richman, S., Naftolin, F., 2006. Evolution of the Mammalian Reproductive Tract and Placentation. In: Mor, G. (Ed.), Immunology of Pregnancy. Medical Intelligence Unit. Springer, New York, pp. 7–14.
Rutella, S., Pierelli, L., Bonanno, G., Sica, S., Ameglio, F., Capoluongo, E., et al., 2002. Role for granulocyte colony-stimulating factor in the generation of human T regulatory type 1 cells. Oct 1 Blood 100 (7), 2562–2571. https://doi.org/10.1182/blood-2001-12-0291.
Saito, S., Nakashima, A., Shima, T., Ito, M., 2010. Th1/Th2/Th17 and regulatory T-cell paradigm in pregnancy (Jun). Am. J. Reprod. Immunol. 63 (6), 601–610. https://doi.org/10.1111/j.1600-0897.2010.00852.x.
Salmassi, A., Schmutzler, A.G., Huang, L., Hedderich, J., Jonat, W., Mettler, L., 2004. Detection of granulocyte colony-stimulating factor and its receptor in human follicular luteinized granulosa cells (Mar). Fertil. Steril. 81 (1), 786–791. https://doi.org/10.1016/j.fertnstert.2003.09.039.
Scarpellini, F., Sbracia, M., 2012. G-CSF treatment improves IVF outcome in women with recurrent implantation failure in IVF. J. Reprod. Immunol. 94, 103. https://doi.org/10.1016/J.JRI.2012.03.435.
Sfakianoudis, K., Rapani, A., Grigoriadis, S., Pantou, A., Maziotis, E., Kokkini, G., et al., 2021. The role of uterine natural killer cells on recurrent miscarriage and recurrent implantation failure: from pathophysiology to treatment. Oct 9 Biomedicines 9 (10), 1425. https://doi.org/10.3390/biomedicines9101425.
Sharma, S., 2014. Natural killer cells and regulatory T cells in early pregnancy loss. Int. J. Dev. Biol. 58 (2-4), 219–229. https://doi.org/10.1387/ijdb.140109ss.
Shaulov, T., Sierra, S., Sylvestre, C., 2020. Recurrent implantation failure in IVF: a Canadian fertility and andrology society clinical practice guideline (Nov). Reprod. Biomed. Online 41 (5), 819–833. https://doi.org/10.1016/j.rbmo.2020.08.007.
Singh, R., Singh, M., Jindal, A., Jindal, P.C., 2015. A prospective randomized controlled study (RCT) of intrauterine administration of granulocyte colony-stimulating factor (G-CSF) before embryo-transfer on resistant thin endometrium in IVF cycles. Hum. Reprod. 30, i280. https://doi.org/10.1093/humrep/30.Supplement-1.1.
Stope, M.B., Mustea, A., Sänger, N., Einenkel, R., 2023. Immune cell functionality during decidualization and potential clinical application. Apr 27 Life 13 (5), 1097. https://doi.org/10.3390/life13051097.
Su, Q., Pan, Z., Yin, R., Li, X., 2024. The value of G-CSF in women experienced at least one implantation failure: a systematic review and meta-analysis. Apr 16 Front. Endocrinol. (Lausanne) 15, 1370114. https://doi.org/10.3389/fendo.2024.1370114.
Sudoma, I., Goncharova, Y., Dons'koy, B., Mykytenko, D., 2023. Immune phenotype of the endometrium in patients with recurrent implantation failures after the transfer of genetically tested embryos in assisted reproductive technology programs (Jun). J. Reprod. Immunol. 157, 103943. https://doi.org/10.1016/j.jri.2023.103943.
Tanaka, T., Miyama, M., Masuda, M., Mizuno, K., Sakamoto, T., Umesaki, N., et al., 2000. Production and physiological function of granulocyte colony-stimulating factor in non-pregnant human endometrial stromal cells (Dec). Gynecol. Endocrinol. 14 (6), 399–404. https://doi.org/10.3109/09513590009167710.
Uzumaki, H., Okabe, T., Sasaki, N., Hagiwara, K., Takaku, F., Tobita, M., et al., 1989. Identification and characterization of receptors for granulocyte colony-stimulating factor on human placenta and trophoblastic cells (Dec). Proc. Natl. Acad. Sci. 86 (23), 9323–9326. https://doi.org/10.1073/pnas.86.23.9323.
Von Woon, E., Greer, O., Shah, N., Nikolaou, D., Johnson, M., Male, V., 2022. Number and function of uterine natural killer cells in recurrent miscarriage and implantation failure: a systematic review and meta-analysis. Jun 30 Hum. Reprod. Update 28 (4), 548–582. https://doi.org/10.1093/humupd/dmac006.
Wang, C., Guan, D., Li, Z., Yang, Y., Yang, K., 2022. Emerging trends and frontier research on recurrent implantation failure: a bibliometric analysis (Mar). Ann. Transl. Med. 10 (6), 307. https://doi.org/10.21037/atm-22-703.
Wang, W., Sung, N., Gilman-Sachs, A., Kwak-Kim, J., 2020. T Helper (Th) cell profiles in pregnancy and recurrent pregnancy losses: Th1/Th2/Th9/Th17/Th22/Tfh Cells. Aug 18 Front. Immunol. 11, 2025. https://doi.org/10.3389/fimmu.2020.02025.
Würfel, W., 2000. Approaches to a better implantation. J. Assist Reprod. Genet. 17, 473.
Xie, Q., Quan, X., Lan, Y., Yang, X., 2024. Uterine infusion strategies for infertile patients with recurrent implantation failure: a systematic review and network meta-analysis. Apr 16 Reprod. Biol. Endocrinol. 22 (1), 44. https://doi.org/10.1186/s12958-024-01221-x.
Yang, X., Gilman-Sachs, A., Kwak-Kim, J., 2019. Ovarian and endometrial immunity during the ovarian cycle (Jun). J. Reprod. Immunol. 133, 7–14. https://doi.org/10.1016/j.jri.2019.04.001.
Yano, N., Endoh, M., Nomoto, Y., Sakai, H., Rifai, A., 1996. Increase of HLA-DR-positive natural killer cells in peripheral blood from patients with IgA nephropathy (Aug). Hum. Immunol. 49 (1), 64–70. https://doi.org/10.1016/0198-8859(96)00057-2.
Zeyneloglu, H.B., Tohma, Y.A., Onalan, G., Moran, U., 2020. Granulocyte colony-stimulating factor for intracytoplasmic sperm injection patients with repeated implantation failure: which route is best? (May). J. Obstet. Gynaecol. 40 (4), 526–530. https://doi.org/10.1080/01443615.2019.1631772.