

ImmunoTools *special* Award 2023



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Accentuations in endometrial immunophenotype in patients with recurrent implantation failures

The receptivity of the endometrial tissue is a key factor in embryo implantation and subsequent pregnancy outcome. Immune cells shape the receptivity of the endometrium, their proportions and functional conditions are critical for the success of the reproductive process.

We actively investigate the endometrial lymphocytes phenotype and have developed a number of original approaches to the isolation and analysis of endometrial lymphocyte by flow cytometry (*Chernyshov, Viktor P et al. "Comparison of T and NK lymphocyte subsets between human endometrial tissue and peripheral blood." Central-European journal of immunology 2019*). Strong positive correlations of CD8 expression in NK cells ($r = 0.6478$, $p < 0.001$) and HLA DR expression in CD8 T cells ($r = 0.6107$, $p < 0.01$) between peripheral blood and endometrium were registered in fertile women.

Previous studies of the immunophenotype of endometrial tissue in healthy fertile women allowed us to form "reference fertility norms" and to detect numerous immune accentuations in the phenotype of endometrial lymphocytes in patients with Recurrent implantation failures (RIF) on pre-implantation genetic tested [PGT] embryos (*Sudoma, Iryna et al. "Immune phenotype of the endometrium in patients with recurrent implantation failures after the transfer of genetically tested embryos in assisted reproductive technology programs." Journal of reproductive immunology 2023*). Immune abnormalities that were found in study in more than half of the patients (66.7 %) may be the cause of implantation failures in genetically tested embryo transfer programs.

We also showed the dynamics of the formation of the "immunoreceptive window of implantation" (*Dons'koi et al. "Changes in the immunophenotype of endometrium during implantation window receptivity formation in healthy fertile women" Placenta 2023*) and the relationship between immune populations of peripheral blood and endometrial cells. We shown that endometrial immunophenotype is peculiar and specific but not autonomic and isolated. Activity (p46 on NK-cells) of peripheral blood lymphocytes is reflected in endometrial lymphocytes profile.

Recurrent implantation failures (RIF) in assisted reproduction programs are one of the most challenging problems. Among the factors that can adversely affect implantation, endometrial immune structural disorders may be one of the leading causes. The aim of our work was to study the immune features of the endometrium in women with RIF after genetically tested embryo transfer in comparison with fertile control.

We plan to study the endometrial immunophenotype and peripheral blood lymphocytes in women with recurrent embryo implantation failures on PGD-tested embryos. It is planned to examine 40 - 60 such women.

It is planned to investigate the following subpopulations by Flow Cytometry in samples of peripheral blood and endometrium taken in the implantation window. (NK-subsets: CD3-CD56+, CD3-CD56++, CD3-CD56+CD16+, CD3-CD56+CD49a+, CD3-CD56+CD335+CD337+ CD94+, CD3-CD56+CD158a+CD158ah+, CD3-CD56+HLA-DR+, CD3-CD56+CD69+, CD3-CD56+CD8+. T-subsets: CD3+CD4+, CD3+CD4+, CD3+CD8+HLA-DR+, CD3+CD69+, CD3+CD94+. And B cells CD3-CD56-CD19+ and CD45+CD3-CD56-CD19+CD138+.

With the **ImmunoTools** special Award we would like to expand our current project and screen for additional cell populations that might be present in endometrial tissue in patients with RIF. We are interested in the following **ImmunoTools** products: CD2PE, CD4PerCP, CD7PE, CD16PE-Dy647, CD25 PE/Dy647, CD45PerCP, CD49d FITC, CD57PE, TCRabPE, TCRgd FITC.

In fact, there is no information in the scientific literature regarding the expression of CD2 and CD7 on endometrial NK and T cells. So it's important to investigate. Similarly, there is insufficient information regarding the proportions of T/ NKT of NKT-like cells in the endometrium and TCRab /TCRgd/ CD57 will enable us to study these proportions. In endometrial tissue, the role of CD49a on NK cells is well-studied, but there are no data on the expression of CD49d either.

The research will allow a detailed study of accentuations in lymphocyte populations of endometrial tissue that associated with an unfavorable prognosis of embryo implantation. This will help further in the diagnosis and treatment of patients with infertility.

ImmunoTools special AWARD for **Boris Donskoi** includes 10 reagents

FITC - conjugated anti-human CD49d, TCRgd

PE - conjugated anti-human CD2, CD7, CD57, TCRab

PerCP - conjugated anti-human CD4, CD45

PE-Dy647 - conjugated anti-human CD16, CD25

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