Pregnancy is for the mother a time of profound changes in her immunity. Within 280 days in human and 21 days in mice the mother's immune system has to tolerate antigen-foreign fetus. At the same time it must be in constant readiness to fight pathogens that threaten both the mother and the fetus.

The development of fetal tolerance starts very early already during fertilization, when maternal immune system cells contact with foreign paternal antigens present in semen.

Disorders of pregnancy, such as miscarriage and pre-eclampsia, are associated with disturbed mechanisms of immune tolerance. The problem of miscarriage affects about 1% of all women. Spontaneous abortions occur in the range of 11-20% of all pregnancies and the vast majority of them occur in very early stages of pregnancy. It is reported that 30% of them are lost between the implantation period and the sixth week of pregnancy. Some of these cases are attributed to such factors as anatomical defects, hormonal disorders, age or genetic diseases. The problem of recurrent miscarriages still remains unresolved, and more than 50% of the loss of pregnancies have no clearly defined cause. Researches indicate that a significant part of these abortions is immune mediated. The mechanisms underlying this phenomenon are still poorly understood. Scientist suppose that the abnormal functioning of the cells of the immune system may be responsible for this.

Regulatory T-cells (Treg) are currently considered for responsible for peripheral immune tolerance to fetal antigens among the known types of cells, immune factors and soluble regulatory molecules. It has been proved that in humans and in mice already at a very early stage of normal pregnancy there is an increase in the number of regulatory cells both in decidua and in peripheral blood. However, the decrease in their number and activity is observed in spontaneous abortions. Recently, similar immunosuppressive properties have been observed among some B cell populations in B regulatory lymphocytes (Breg). Both cell populations are capable of secreting anti-inflammatory cytokines incl. interlekin-10 (IL-10) and transforming growth factor β (TGF-β) that inhibit the mother's immune response against developing fetus.

For ethical reasons, most of the experiments aimed at explaining the complex mechanisms leading to fetal tolerance in pregnancy are carried out on animal models. The best known is the murine abortion model of pregnancy which is characterized by an increase in fetal mortality, as well as a decrease in active Treg lymphocytes compared to the normal pregnancy.

Our preliminary studies on this mouse model have demonstrated an increased expression toll-like receptor 9 (TLR9) gene in spleen’s B cells in abortion prone females before embryo implantation. In addition to pathogen associated molecular patterns, these receptors may recognize signals of inborn origin, among which, in the context of pregnancy, paternity antigens (e.g. male DNA present in semen) may be considered. The toll-like receptors present on B lymphocytes may lead to B cell activation manifested by increased expression of cytokines, both pro- and anti-inflammatory.

The main objective of this project is to investigate whether the TLR9 receptor-dependent regulation of regulatory B cells activity may be responsible for the development of fetal tolerance. The research hypothesis assumes that TLR9 receptor expression on B lymphocytes is changed in women suffering from miscarriage and murine abortion model of pregnancy. This in turn leads to an unfavorable co-stimulatory phenotype of these cells and changes in the profile of secreted cytokines, what all together could cause the miscarriages.

In the project, we plan to investigate the expression of TLR9 receptor on selected B cell subpopulations in the blood of normal pregnant women and in women with spontaneous and recurrent abortions. It will be also examined the percentage of Breg cells, level of co-stimulatory molecules and MHC class II on B cells, which we will correlated with the amount of TLR9 receptor on the same cells. We will also verified the percentage of cytotoxic T cells, helper T cell and NK cells in the blood and hormone concentrations and the level of anti- and pro-inflammatory cytokines in serum. It aims to determine the correlation between their number / level and the amount of Breg cells.

The proposed project will contribute to a better understanding the role which Breg cells may play in pregnancy. The B cells TLR9 receptor may become in future an effective target in therapies aimed at restoring impaired immune tolerance in pregnancy. Our study may also become useful in a safe, for the mother and fetus, diagnosis of adverse mother’s immune status which leads to an early miscarriage