

The endometrium is the lining of the body of the uterus. Its morphology and functions periodically change in the subsequent phases of the menstrual /estrus cycle in both women and cows. Proper activity of the endometrium is necessary for the implantation of the embryo after fertilization and its proper development with the course of pregnancy. Disruption of the endometrial function, both during the estrus cycle and during pregnancy, carries a high risk of fertility disorders. The endometrium, in response to hormonal signals, undergoes cyclical processes of cell growth, proliferation and degeneration. These mechanisms are activated by hormones, growth factors and cytokines, but many elements of this process remain unexplained. Neuropeptides are included as the molecules potentially involved in the regulation of endometrial function. One of such peptides, is the recently discovered neuropeptide - phoenixin 14, which expression we confirmed in the luminal and glandular epithelial cells and the endothelial cells of blood vessels of cows' endometrium. To date, phoenixin 14 has not been studied in the uterus, and there is a lack of data on the role of this factor during cycle and pregnancy in the endometrium in cows and other species. The results of preliminary experiments suggest that phoenixin 14 regulates the secretory and proliferative processes in the endometrium. The aim of the project is to determine the role of phoenixin 14 in the endometrium function during the estrus cycle.

In the planned tasks, we will want to define: (1) mRNA and protein expression profile of phoenixin 14 and its receptor (GPR173) in the endometrium during the estrus cycle; (2) factors, that can regulate the expression of phoenixin-14 and its receptor in the endometrium; (3) impact of phoenixin 14 on the secretory (synthesis and secretion of steroid hormones and prostaglandins) and proliferative (growth, proliferation, migration and angiogenesis) activity of the endometrium. In addition, (4) we will check whether the effect of phoenixin 14 modulates the expression of genes involved in the control of studied physiological processes in endometrial cells/ slices, i.e. proliferation, migration, apoptosis, prostaglandin synthesis, steroid hormones, factors controlling uterine receptivity and epithelial adhesion. (5) In the next step we will identify intracellular signaling pathways modulated by phoenixin-14 in the endometrial cells.

The experimental material will be: (i) uteri collected from different stage of estrous cycle, (ii) the endometrial slices and (iii) the luminal epithelial cells and (iv) the endothelial cells of blood vessels cultured in vitro. Molecular biology techniques, such as: Real Time PCR and Western blot for expression studies, microscopic and time-laps technique for analysis of proliferation, migration, apoptosis, angiogenesis, and immunoenzymatic analysis will be used in this project.

The results obtained from these studies will determine the role of phoenixin-14 in endometrial function. Data obtained from such comprehensive studies will contain innovative elements, and therefore we expect that they will extend our knowledge in the field of endocrinology and reproductive physiology. Moreover, these studies extend our earlier studies on the function of uterine and ovarian cells using cow tissue/cells as a model. We assume these studies will help to understand better the changes in the reproductive system during the estrous cycle and pregnancy. Especially that, dysfunction of corpus luteum and a reduction of P4 level in the blood may affect the so-called "early embryos mortality", that in farm animals is up to 40%. From this point of view, better understanding the mechanisms regulating the function of these organs at the level of cellular and sub-cellular regulation may also have some practical significance in medical and veterinary practice.