Distribution of Growth Stimulating and Degradation Factors in Cow Endometrium in Postparturition Period
Augšanu stimulējošo un degradācijas faktoru izplatība govs endometrijā pēcdzemdību periodā

Ilga Šematoviča, Aleksandrs Jemeļjanovs
Research Institute of Biotechnology and Veterinary Medicine „Sigra", LLU
LLU Biotehnoloģijas un veterinārmedicīnas zinātniskais institūts „Sigra"
e-mail: sigra@lis.lv
Māra Pilmane
Institute of Anatomy and Anthropology, Riga Stradins University
Rīgas Stradiņa universitātes Anatomijas un antropoloģijas institūts
e-mail: pilmane@latnet.lv

Abstract. The aim of the research was to reveal distribution of the growth stimulating and degradation factors, inflammatory factors, and neuropeptide-containing innervation, as well as apoptosis in the cow's endometrium in postparturition period. Haematoxylin and eosin were used for routine staining of each sample. Matrix metalloproteinases type 2 and type 9 (MMP-2 un MMP-9, working dilution (w.d.) 1:100, *R&D*, England), tumor necrosis factor-α (TNF-α, w.d. 1:100, *Abcam*, England), interleukin-10 (IL-10, w.d. 1:400, *Abcam*, England), vascular endothelial growth factor (VEGF, w.d. 1:50, *DakoCytomation*, Denmark), nerve growth factor receptors p75 (NGFR p75, w.d. 1:150, *DakoCytomation*, Denmark), and protein gene product 9.5 (PGP 9.5, w.d. 1:1600, *DakoCytomation*, Denmark) were detected immunohistochemically (IMH). TUNEL method was used for detection of apoptosis (programmed cells death – PCD). A mild positive correlation ($p<0.05$) was observed between amount of inflammatory cells and TNF-α ($r=0.52$), PCD and TNF-α ($r= 0.58$), TNF-α and NGFR p75 ($r=0.51$), NGFR p75 and PGP 9.5 ($r=0.49$), VEGF and PCD ($r=0.59$), and VEGF and NGFR p75 ($r= 0.64$). A strong positive correlation ($p<0.01$) was observed between TNF-α and VEGF ($p<0.01$; $r=0.72$), as well as between VEGF and NGFR p75 ($r=0.64$). The research suggests that uterus endometrial tissue degradation and repairing were in balance during postparturition. The intensity level of tissue repairing process in the cow's endometrium was specific for the individual organism, which might be connected with immunity of each of them.
Key words: cow's endometrium, matrix degradation factors, growth factors, inflammatory factors.

Introduction

Time after parturition regarding successful involution is one of the most critical for optimal function of the reproductive system in the subsequent lactations and gestation periods (Twardoň et al., 2002). Substantial morpho-physiological changes occur in uterus tissue, blood vessels, and nerves during the reproductive cycle and gestation (Zoubina et al., 1998; Hickey et al., 2003). Insufficient awareness still persists concerning endometrium changes, distribution of the growth stimulating factors, and apoptosis in the cow's endometrium in postparturition period. Varied alterations happen exactly in extracellular matrices (ECM) of the uterus cells regulated by matrix metalloproteinases (MMP) and other factors during each reproductive cycle and depending on the physiological stage (CurryJr and Osteen, 2001). These proteolyses regulate composition of ECM by releasing (making place for) the biologically active sites on cells' surfaces (Jacob, 2003). Thereby MMP as zinc-depending endopeptidases provide and maintain homeostasis of ECM in cells. There are more than 20 members of MMP known in mammals that take part in ECM degradation in different physiological and pathological processes (CurryJr and Osteen, 2001; Kliem, 2006). Genetically determined cells death termed apoptosis or programmed cells death (PCD) is one of the homeostatic mechanisms that results in disposing of tissues from old, defective, deteriorated, and needless cells (Kliem, 2006).

Inflammatory factors have an important role in successful involution process, because increased infiltration of endometrium with inflammatory cells, like macrophages, lymphocytes, etc., is characteristic

for postparturition period (Емельянова, 1974), as well as number of inflammatory cells in the endometrium are closely related with the different stages of the oestrus cycle (Kaoket, 2003).

Interleukin-10 (IL-10) and tumor necrosis factor-α (TNF-α) as inflammatory markers take part in different processes in cow's uterus in postparturition period; besides, TNF-α is mentioned to be one of inductors of apoptosis (Kliem, 2006).

In the uterus tissue nerve fibres, changes in the amount of protein gene product (PGP 9.5) are observed in correlation with different biological processes (Tingaker et al., 2006). Nerve growth factors (NGF) stimulate alimentation, survival, and differentiation of the neurons (Davidson et al., 2003; Wilcox et al., 2004), but innervation is important for metabolic processes in all types of tissues.

Increased expression of the vascular endothelial growth factor (VEGF) observed in repairing processes in tissues is described as a response to hypoxic stimuli (Scott et al., 1998; Ankoma–Sey et al., 2000).

The aim of the investigation was to find out presence and distribution of growth stimulating and degradation factors, as well as qualitative and quantitative correlations between those factors and substances in cow endometrium in postparturition period.

Materials and Methods

Biopsy samples from the cow's uterus were taken in winter 2004/2005 on the research and training farm "Vecauce". Nine cows were biopsied twice – in the first and fifth week of postparturition period. Histological investigations were performed at the Institute of Anatomy and Anthropology of Riga Stradins University. Routine staining with haematoxylin and eosin was performed for each endometrium sample (Aughey, Frye, 2001). Immunohistochemically (IMH) were detected: matrix metalloproteinases type 2 and type 9 (Anti-human MMP-2 antibody / AF902 and Anti-mouse MMP-9 antibody / AF909, working dilution (w.d.) 1:100, *R&D*, England), tumor necrosis factor-α (Rabbit Polyclonal to TNF-α / ab6671, w.d. 1:100, *Abcam*, England), interleukin-10 (Rabbit Polyclonal to IL-10 / ab34843, w.d. 1:400, *Abcam*, England), vascular endothelial growth factor (Anti-human VEGF / M 7273, w.d. 1:50, *DakoCytomation*, Denmark), nerve growth factor receptors p75 (Anti-human NGFR p75 / M3507, w.d. 1:150, *DakoCytomation*, Denmark), and protein gene product 9.5 (Polyclonal Rabbit Anti-PGP 9.5 / Z 5116, w.d. 1:1600, *DakoCytomation*, Denmark) (Hsu et al., 1981). TUNEL method was used for detection of apoptosis (Negoescu et al., 1998). Distribution of these factors and substances was determined semiquantitatively by counting positive structures in visual fields (0 – occasional, + – few, ++ – moderate, +++ – numerous positive structures). For the data statistical processing the following methods were used: Student's t-test, Wilkinson's test, and statistical correlation analyses (Paura, Arhipova, 2002; Arhipova, Bāliņa, 2003).

Results

Distribution of the researched factors in cow's endometrium in the first and fifth week after parturition is displayed in Tables 1 and 2, respectively.

A significantly increased number of inflammatory cells ($p<0.01$), especially the number of macrophages ($p<0.05$), intensity of TNF-α ($p<0.01$) and VEGF expressing ($p<0.05$), as well as amount of NGFR p75 ($p<0.05$) were observed in cow's endometrium from the first up to the fifth week after parturition.

A mild positive correlation ($p<0.05$) was observed between amount of inflammatory cells and TNF-α ($r=0.52$), number of apoptotic cells and TNF-α ($r=0.58$), TNF-α and NGFR p75 ($r=0.51$), NGFR p75 and PGP 9.5 ($r=0.49$), VEGF and PCD ($r=0.59$), and VEGF and NGFR p75 ($r=0.64$). A strong positive correlation ($p<0.01$) was detected between TNF-α and VEGF containing structures ($p<0.01$; $r=0.72$), as well as between VEGF and NGFR p75 ($r=0.64$).

Discussion

The intensity of metabolic processes regarding tissue repairing processes is more intensive in the fifth week after parturition than in the first week, because at that time a statistically significant increase in the number of inflammatory cells is observed in cow's endometrium. Infiltration of endometrium with inflammatory cells has always been observed in early postparturition period, as a wide spectrum of microorganisms are found in cow's uterus after parturition (Зверева, Хомин, 1976; Twardon et al., 2002; Lewis, 2003).

In the literature, different data are found on the resumption of cyclicity in dairy cows – in the first four weeks (Dzenīte, Jonins, 1989), and in six up to seven weeks (Kask et al., 1998; 1999; 2003) after parturition; whereas increase in infiltration with inflammatory cells depending on the phase of the reproductive cycle has been detected only by a few authors, for example, Kaoket (2003).

Cytokine IL-10, mediator of inflammation, is released not only by $CD4^{+}$T cells, but also by endometrial glandular and epithelial cells (Sallinen et al., 2000; El-Sherif et al., 2001). Expression of IL-10 is related to the reproductive cycle phases but not to inflammatory processes in the uterus, namely, expression of IL-10 increases in the secretory phase (Gamo et al., 2007).

Table 1

Distribution of the growth stimulating and degradation factors, inflammatory factors, and neuropeptide-containing innervation, as well as apoptosis in the cow endometrium in the 1st week after parturition

Cow's name	No. of a sample / week	Inflammatory cells in endometrium	IL-10	TNF-α	Apoptotic cells in endometrium, %	MMP-9	MMP-2	VEGF	NGFR (p75)	PGP 9.5
Tapa	F2 / 1	3.3±2.73	+	–	34.5±3.20	+++	+	–	–	++
Esma	F3 / 1	14.7±7.63	–	–	25.2±3.15	+++	+	–	–	+
Dakota	F4 / 1	13.7±12.94	+	–	……..	++	+	–	++	++
Tērvete	F5 / 1	15.0±4.20	++	–	30.2±5.50	++	+	–	+	–
Vaida	F6 / 1	23.7±8.36	–	–	14.0±0.3	+++	+	–	+	+
Okence	F7 / 1	16.5±2.43	–	++	……..	+	+	–	–	+
Elba	F8 / 1	11.2±7.19	+	+	……..	+++	+	–	+	+
Akāce	F9 / 1	27.2±4.02	–	++	……..	++	+	–	–	+
Loda	F10 / 1	4.8±1.19	+++	–	21.6±28.85	++	+	–	–	+

Notations: IL-10 – interleukin-10; TNF–α – tumor necrosis factor-α; MMP-9 and MMP-2 – matrix metalloproteinases type 2 and type 9; VEGF – vascular endothelial growth factor; NGFR p75 – nerve growth factor receptors p75; PGP 9.5 – protein gene product 9.5;
– – no cells contain VEGF, NGFR p75, PGP 9.5, and TNF-α;
+ – few cells contain VEGF, NGFR p75, PGP 9.5, and TNF-α;
++ – moderate number of cells conatin VEGF, NGFR p75, PGP 9.5, and TNF-α;
+++ – numerous cells contain VEGF, NGFR p75, PGP 9.5, and TNF–α.

Table 2

Distribution of the growth stimulating and degradation factors, inflammatory factors, and neuropeptide-containing innervation, as well as apoptosis in the cow endometrium in the 5th week after parturition

Cow's name	No. of a sample / week	Inflammatory cells in endometrium	IL-10	TNF-α	Apoptotic cells in endometrium, %	MMP-9	MMP-2	VEGF	NGFR (p75)	PGP 9.5
Tapa	1 / 5	32.5±7.18	–	+	28.9±6.15	++	+	–	++	+++
Esma	7 / 5	45.2±32.02	+	+	63.2±1.50	++	+	–	–	+
Dakota	3 / 5	34.5±11.18	+++	+++	…….	+++	+	+	++	+
Tērvete	2 / 5	25.0±4.15	++	++	12.9±0.85	++	+	–	+	+
Vaida	6 / 5	46.2±7.17	–	++	62.5±7.8	++	+	–	–	+
Okence	8 / 5	19.3±13.87	+	+++	74.0±14.35	+++	+	+	++	+
Elba	9 / 5	29.3±6.02	+	+++	……..	++	+	+	+++	++
Akāce	5 / 5	29.5±4.76	–	+	……..	++	+	+	+++	+
Loda	4 / 5	45.3±13.06	++	++	23.8±4.00	++	+	–	+++	+

Notations: see Table 1.

Cytokine TNF-α, besides its other functions, improves growth, differentiation and functioning of cells in the uterus during the reproductive cycle and gestation; however, it has no influence on expression and action of MMP-9 in endometrial stromal cells (Gamo et al., 2007).

We determined an increased number of apoptotic cells in endometrium from the first up to fifth week

after parturition, which might be explained by the fact that tissues of the uterus are at the end of involution in the fifth week after parturition, and are more compact than in the first week when they are outstretched and with oedema. Apoptosis is described as a physiological process in different types of glandular tissues of the reproductive system during their involution processes (Walker et al., 2005). A mild positive correlation between the number of apoptotic cells and number of macrophages in endometrium ($r= 0.54$; $p<0.1$) can be explained by the fact that TNF-α is one of inducers of apoptosis (Zhdanov et al., 2003).

The amount of MMP-2 positive structures in cow's endometrium in postparturition period practically did not change. Intensity of MMP-9 expression statistically significantly decreased from the first to the fifth week after parturition, which proves decreasing in the degenerative processes during that time. Our research results partly agree with conclusions found in the literature that during the first ten days of postparturition period, degenerative processes in endometrium prevail, whereas regenerative processes become active later after parturition (Shea, Wright, 1984; Емельянова, 1974). It should be noted that the research made by other authors is based only on histological investigations.

More recent investigations indicate the important role of MMP-2 and MMP-9 in delivering fetal membranes after parturition (Takagi et al., 2006). Other authors have found that MMP-9 is regulated by estrogens, and it is significant for uterus tissue remodeling regarding the reproductive cycle (Zhang et al., 2006), which was not researched in our present study.

The research suggests that increase in PGP9.5 and NGFR p75 in endometrium at that time is induced by inflammatory mediators like IL-10 and TNF-α, because the PGP9.5 and NGFR increase in tissues is characteristic of regenerative processes as well as processes of tissue innervation reconstruction. The changes in innervation regarding reproductive cycle were proved (Zoubina et al., 1998), as well as the fact that increased innervation promotes blood flow in tissues (Blacklock, Smith, 2004).

VEGF-containing structures were found in endometrial blood vessels and under epithelium only in the fifth week after parturition. VEGF is an important signal protein – cytokine – involved in angiogenesis of the embryonic as well as adult organism from the existing vascular tissues (Wei et al., 2004; Wang et al., 2003; Halder et al., 2000; Guidi et al., 1998). The activity of VEGF is oriented to the endothelial cells of blood vessels and its influence on other types of cells as monocyte/macrophage stimulation was established. The ability of VEGF to stimulate mitogenesis of endothelial cells and migration of cells was also observed. That also enhances the microvascular permeability *in vitro*, which is why VEGF is sometimes referred to as a vascular permeability factor (Halder et al., 2000). It is probable that VEGF expression found only in the fifth week after parturition in our investigation can be associated with the increased amount of inflammatory cells in endometrium at that time. The production of VEGF can be induced in the cells which have not received enough oxygen, and then these cells produce transcription factor, namely, hypoxia inducible factor (HIF). Among other functions (modulation of erythropoesis), HIF stimulates VEGF production (Ankoma-Sey et al., 2000).

An ischemia of endometrial tissues in the fifth week after parturition can be raised by intensive metabolic processes connected with inflammatory processes in uterus tissue in postparturition period. One of the reasons of increased expression of VEGF in inflammatory process is the already mentioned activity of TNF-α (Scott et al., 1998). Whereas in our investigation, statistically significant ($p<0.01$) increase in TNF-α positive structures, as well as strong positive correlation ($p<0.01$; $r=0.72$) among TNF-α and VEGF positive structures in cow's endometrium were observed in the fifth week after parturition. Several authors have found that the VEGF expression, localization, and intensity in endometrium depend on the phase of reproductive cycle in the woman and in the cow (Wijayagunawardane et al., 2005; Wei et al., 2004; Zhang et al., 2006). Investigations on the resumption of the reproductive cycle after parturition were not the aim of our research.

Conclusions

1. Expression of NGFR p75, VEGF, and TNF-α positive structures as well as correlation between these structures indicate their reciprocal stimulation in the endometrium of the cow from the first to fifth week after parturition.
2. The increased amount of VEGF, NGFR p75, TNF-α, and PGP 9.5 positive structures in the endometrium of the cow indicate the intensification of regenerative processes in connection with inflammatory process in cow's endometrium in the fifth week after parturition.
3. The decrease in the amount of MMP-9 positive structures in the cow's endometrium from the first to fifth week after parturition indicates the attenuation of degenerative processes and the role of MMP-9 in fetal membrane delivering processes. The unchangeable amount of MMP-2 positive structures in the cow endometrium indicates stability of respective structures at that period.

References

1. Ankoma–Sey, V.M.D., Wang, Y., Dai, Z. (2000) Hypoxic stimulation of vascular endothelial growth factor expression in activated rat hepatic stellate cells. *Hepatology*, Volume 31, Issue 1, 141-148.
2. Arhipova, I., Bāliņa, S. (2003) *Statistika Ekonomikā. Risinājumi ar SPSS un Mikrosoft Exel.* Rīga: Datorzinību centrs, 352 lpp.
3. Aughey, E., Frye, F.L. (2001) *Comparative veterinary histology with clinical orrelates.* Manson Pub.Ltd., London, 14-127.
4. Blacklock, A.D., Smith, P.G. (2004) Estrogen increases calcitonin gene-related peptide-immunoreactive sensory innervation of rat mammary gland. *Journal of neurobiology*, Vol. 59, Issue 2, 192-204.
5. CurryJr, T.E., Osteen, K.G. (2001) Cyclic changes in the matrix metalloproteinases system in the ovary and uterus. *Biology of reproduction,* Vol. 64, 1285-1296.
6. Davidson, B., Reich, R., Lazarovici, P., Nesland, J.M., Skrede, M., Risberg, B., Tropé, C.G., Florenes, V.A. (2003) Expression and activation of the nerve growth factor receptor TrkA in serous ovarian carcinoma. *Clinical cancer research*, Volume 9, 2248-2259.
7. Dzenīte, A., Jonins, V. (1989) *Govju mākslīgā apsēklošana*. Rīga: Avots, 150 lpp.
8. El-Sherif, A.M., Seth, R., Tighe, P.J., Jenkins, D. (2001) Quantitative analysis of IL-10 and IFN-gamma mRNA levels in normal cervix and human papillomavirus type 16 associated cervical precancer. *Journal of pathology*, 195(2):179-85.
9. Gamo, T., Yamauchi, N., Nishimura, K., Watanabe, R., Matsumoto, K., Oozono, S., Kubota, K., He, P., Soh, T., Hattori, M. (2007) Effects of tumor necrosis factor-α on cell proliferation, prostaglandins and matrix-metalloproteinases production in rat endometrial stromal cells cultured in vitro. *Journal of experimental zoological genetics and physiology*, Vol. 307A, Issue 12, 699-707.
10. Guidi, A.J., Abu-Jawdeh, G., Tognazzi, K., Dvorak, H.F., Brown, L.F. (1998) Expression of vascular permeability factor (vascular endothelial growth factor) and its receptors in endometrial carcinoma. *Cancer*, Volume 78, Issue 3, 454-460.
11. Halder, J.B., Zhao, X., Soker, S., Paria, B.C., Kagsbrun, M., Das, S.K., Dey, S.K. (2000) Differential expression of VEGF isoforms and $VEGF_{164}$-specific receptor neuropilin-1 in the mouse uterus suggests a role for $VEGF_{164}$ in vascular permeability and angiogenesis during implantation. *Genesis,* Volume 26, Issue 3, 213-224.
12. Hickey, M., Fraser, I. (2003) Human uterine vascular structures in normal and diseased states. *Microscopy research and technique*, Volume 60, Issue 4, 377-389.
13. Hsu, S.M., Raine, L., Fanger, H. (1981) The use of antiavidin antibody and biotin-streptavidin peroxydase complex in immunoperoksidase technics. *Am. J. Clin. Pathol*., Vol. 75, p. 816.
14. Jacob, P.M. (2003) Extracellular matrix remodeling and matrix metalloproteinases in the vascular wall during aging and in pathological conditions. *Biomedicine & Pharmacotherapy*, Vol. 57, Issue 5, 195-202.
15. Kaoket, K. (2003) Infiltration by cells of the immune system in the sow endometrium. With special reference to different stages of the oestrus cycle and after pre- and post-ovulatory insemination. *Acta universitatis agriculturae sueciae Veterinaria,* Vol. 136, p. 170.
16. Kask, K., Gustafsson, H., Magnusson, U., Bertilsson, J., Gunnarsson, A., Kindahl, H. (1999) Uterine bacteriology, histology, resumption of ovarian activity and granulocyte function of the postpartum cow in different milking frequencies. *Acta Vet. Scand.,* 40(4), 287-297.
17. Kask, K., Kindahl, H., Gustafsson, H. (1998) Bacteriological and histological investigation of the postpartum bovine uterus in two Estonian dairy herds. *Acta Vet. Scan.,* 39 (4): 423-432.
18. Kask, K., Kurykin, J., Lindjärv, R., Kask, A., Kindahl, H. (2003) Assessment of early postpartum reproductive performance in two high producing Estonian dairy herds. *Acta Vet. Scand.*, 44, 131-143.
19. Kliem, H.S. (2006) *Investigations of extracellular matrix proteases, apoptotic and anti-apoptotic factors in the bovine corpus luteum. Inaugural–Dissertation.* Friedberg/Hessen, München, 155 p.
20. Lewis, G.S. (2003) Steroidal regulation of uterine resistance to bacterial infection in livestock. *Reproductive biology and endocrinology*, 1:117: http://www.rbej.com/content/1/1/117/abstract – Accessed on 25.03.2008.
21. Negoescu, A., Guillermet, Ch., Lorimer, Ph., Robert, C., Lantuejoul, S., Brambilla, E., Labat-moleur, F. (1998) *Biochemica*, Vol. 3, 36-41.
22. Paura, L., Arhipova, I. (2002) *Neparametriskās metodes. SPSS datorprogramma.* Jelgava, 148 lpp.
23. Sallinen, K., Veräjänkorva, E., Pöllänen, P. (2000) Expression of antigens involved in the presentation of lipid antigens and induction of clonal anenergy in the female reproductive tract.

Journal of reproductive immunology, Vol. 46, No. 2, 91-101.
24. Scott, P.A.E., Gleadle, J.M., Bicknell, R., Harris, A.L. (1998) Role of the hypoxia sensing system, acidity and reproductive hormones in the variability of vascular endothelial growth factor induction in human breast carcinoma cell lines. *International journal of cancer*, Vol. 75, Issue 5, 706-712.
25. Shea, J.D.O., Wright, P.J. (1984) Involution and regeneration of the endometrium following parturition in the ewe**.** *Cell and Tissue Research*, Vol. 236, No. 2, 477-485.
26. Takagi, M., Yamamoto, D., Ohtani, M., Miyamoto, A. (2006) Quantitative analysis of messenger RNA expression of matrix metalloproteinases (MMP-2 and MMP-9), tissue inhibitor-2 of matrix metalloproteinases (TIMP-2), and steriodogenic enzymes in bovine placentomes during gestation and postpartum. *Molecular reproduction and development*, Vol. 74, Issue 7, 801-807.
27. Tingaker, B.K., Johansson, O., Cluff, A.H., Ekman-Ordeberg, G. (2006) Unaltered innervation of the human cervix uteri in contrast to the corpus during pregnancy and labor as revealed by PGP 9.5 immunohistochemistry. *European Journal Obstetric Gynecology Reproduction Biology*, 125 (1):66-71.
28. Twardon, J., Dejneka, G.J., Błaszkowszowska, M., Trzęsowski, P. (2002) The influence of simpanorm (carazolol) on puerperal period course in dairy cows. Wrocław, Poland: http://216.239.59.104/search ?q=cache:vqm2wmjRKn4J:wwwfatro.it/fatro_ gb/News/Vetupdates/Files/DOC/SIMPANOR.D OC+The+influence+of+Simpanorum&hl=lv&ct =clnk&cd=1&gb=lv – Acessed on 01.02.2007.
29. Walker, N.I., Bennet, R.E., Kerr, J.F.R. (2005) Cell death by apoptosis during involution of the lactating breast in mice and rats. *American journal of anatomy*, Vol. 185, Issue 1, 19-32.
30. Wang, H., Li, Q., Lin, H., Yu, X., Qian, D., Dai, J., Duan, E., Zhu, C. (2003) Expression of vascular endothelial growth factor and its receptors in the rhesus monkey (*Macaca mulata*) endometrium and placenta during early pregnancy. *Molecular reproduction and development,* Vol. 65, Issue 2, 123-131.
31. Wei, P., Chen, X., Song, X., Han, C., Liu, Y. (2004) VEGF, bFGF, and their receptors in the endometrium of rhesus monkey during menstrual cycle and early pregnancy. *Molecular reproduction and development,* Vol. 68, Issue 4, 456-462.
32. Wijayagunawardane, M.P.B., Kodithuwakku, S.P., Yamamoto, D., Miamoto, A. (2005) Vascular endothelial growth system in the cow oviduct: a possible involvment in the regulation of oviductal motility and embryo transport. *Molecular reproduction and development,* Vol. 72, Issue 4, 511-520.
33. Wilcox, B.J., Applegate, M.D., Portera-Cailliau, C., Koliatsos, V.E. (2004) Nerve growth factor prevents apoptotic cell death in injured central cholinergic neurons. *The journal of comparative neurology*, Vol. 359, Issue 4, 573-585.
34. Zhang, X., Christenson, L.K., Nothnick, W.B. (2006) Regulation of MMP-9 expression and activity in the mouse uterus by oestrogen. *Molecular reproduction and development*, Vol. 74, Issue 3, 321-331.
35. Zhdanov, A. V., Kurbanova, D.F., Davydova, M. P., Sosulina, L. Yu., Ezhova, L.S., Sukhikh, G.T. (2003) Apoptosis in fimbriae of fallopian tubes and endometrium in pyoinflammatory adnexal diseases. *Bulletin of experimental biology and medicine*, Vol. 135, No. 2/Febr., 150-153.
36. Zoubina, E.V., Fan, Q., Smith, P.G. (1998) Variations in uterine innervation during the estrus cycle in rat. *The journal of comparative neurology*, Vol. 397, Issue 4, 561-571.
37. Емельянова, М. (1974) Гистоморфологические и гистохимические изменения основных структур эндометрия после отёла. *Диссертация на соискание ученои степени кандидата биологических наук.* Сигулда, 194 стр.
38. Зверева, Г.В, Хомин, С.П. (1976) *Гинекологические болезни коров*. Киев: Урожай, 152 с.

Anotācija

Pētījuma mērķis bija noskaidrot dažus augšanas un degradācijas procesus, iekaisuma faktoru un neiropeptīdus saturošo pozitīvo komponentu daudzumu un mijiedarbību govs endometrijā saistībā ar dzemdes audu pārmaiņām specifisku, endogēnu bioloģiski aktīvu vielu ietekmē pēcdzemdību periodā govīm. Biopsijas paraugi ņemti no deviņām Latvijas brūnās šķirnes govīm LLU MPS „Vecauce" 2004./2005. gada ziemas perioda pirmajā nedēļā un atkārtoti četras nedēļas pēc atnešanās. Iegūtie materiāli izmeklēti RSU Anatomijas un antropoloģijas institūta Morfoloģijas laboratorijās. Noteikts apoptotisko šūnu daudzums ar TUNEL diagnostikas metodi. Imūnhistoķīmiski noteiktas matrices metalloproteināzes 2 un 9 (MMP-2 un MMP-9, darba atšķaidījums 1:100,

R&D, Anglija), audzēju nekrotiskais faktors-α (TNF-α, darba atšķaidījums 1:100, *Abcam*, Anglija), interleikīns-10 (IL-10, darba atšķaidījums 1:400, *Abcam*, Anglija) un proteīngēna peptīds 9.5 (PGP 9.5, darba atšķaidījums 1:1600, *DakoCytomation*, Dānija). Veikta eozīna/hematoksilīna audu krāsošana. Pozitīvo struktūru daudzums vērtēts semikvantitatīvi trīs reprezentatīvos redzes laukos. Datu statistiskajā apstrādē lietoti Stjudenta t-tests, Vilkinsona tests, divfaktoru un daudzfaktoru korelāciju tests. Laikā no pirmās līdz piektajai nedēļai pēc dzemdībām novērots statistiski nozīmīgs iekaisuma šūnu infiltrācijas palielinājums un TNF-α, VEGF un NGFR p75 pozitīvo struktūru daudzuma pieaugums govs endometrijā ($p<0.05$). Vidēja, statistiski nozīmīga ($p<0.05$) pozitīva korelācija novērota starp iekaisuma šūnu daudzumu un TNF-α ($r=0.52$), apoptotisko šūnu daudzumu un TNF-α ($r=0.58$), TNF-α un NGFR p75 ($r=0.51$), NGFR p75 un PGP 9.5 ($r=0.49$), VEGF un apoptotisko šūnu daudzumu ($r=0.59$). Pētījums parādīja minēto faktoru un iekaisuma marķieru līdzsvarotu mijiedarbību govs endometrijā fizioloģiskā pēcdzemdību periodā, kā arī minēto komponentu un faktoru darbības intensitātes atšķirības atsevišķu indivīdu starpā, iespējams, atkarībā no imūnsistēmas kvalitātes.