

SHORT THESIS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY (PhD)

Phagocytic function of neutrophil granulocytes and monocytes in
endometriosis

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INTRODUCTION

Endometriosis is a benign, chronic, systemic, estrogen-dependent, inflammatory disease in which endometrial glandular and stromal tissue, which is originally eutopic (i.e., within the uterine cavity), appears outside the uterine cavity (in an ectopic) localization. The ectopically appearing tissues are functioning synchronously with the menstrual cycle in a similar way to the ectopic endometrium. The leading symptoms of the disease are pain (chronic pelvic pain, dysmenorrhea, dyspareunia, and dysuria), and infertility, which together or separately can lead to a deterioration in the quality of life of affected women. These make the disease important not only from a medical, but also from a social and economic point of view. Determining the exact background of the origin and pathogenesis of the disease is still a challenge for medical science, because despite decades of research, the details of its pathophysiology, the possibility of early diagnosis, a correct indication and technique of surgical treatment, an effective reduction of the chance of recurrence and an internationally validated, complete protocol for a therapy without massive side effects are still to be found.

LITERATURE REVIEW

Endometriosis affects at least 10% of women of reproductive age, but the concrete prevalence may be underestimated. The first clinical symptoms usually appear between 25-45 years of age. At this age, endometriosis may be at the background of 30-60% of chronic pelvic pain and infertility may be diagnosed in 30-50% of affected patients. In endometriosis, the development of infertility is created by a very complex group of factors that may affect the vagina, uterus, fallopian tubes, and ovaries, but may also have a negative impact on oocyte function, motility of the sperm and the embryo's implantation, alter oocyte quality and response to the hormonal environment due to altered immune function and may also impair the chances of implantation of the pregnancy.

Endometriosis may affect around 200,000 women in Hungary and 190 million women of reproductive age worldwide, and its prevalence is rising year by year, partly due to modern diagnostic tools. The cost of the medicine used to treat the symptoms, the longtime of a correct diagnosis and the surgical treatment itself also place an extremely high economic burden on our society.

The anatomic structures most commonly affected by endometriotic lesions in the pelvis are the ovaries (as endometriomas and adhesions), the ovarian fossa, the apex of the bladder, the Douglas pouch and the sacrouterine ligaments. These can be affected superficially, but in many cases, lesions may extend more than 5 mm below the peritoneum.

The diagnosis should be based primarily on appropriate anamnestic data, a physical examination performed according to professional standards and imaging studies (pelvic transvaginal ultrasound and MR imaging) performed by an appropriate specialist.

In conclusion, endometriosis is a multifactorial disease with a complex etiopathogenesis, and the exact clarification of its pathogenesis is a major challenge for gynecologists, reproductive specialists, endocrinologists, and researchers. The theory of retrograde menstruation, described by Sampson in 1927, when menstrual blood is flows back through the fallopian tubes into the pelvic cavity, is one of the well-known basic of the pathogenesis of endometriosis, but it is not the only factor involved in the development of endometriosis, as a large proportion of healthy women have developed endometriosis without it. Recently, several studies have been conducted to understand the development and progression of endometriosis, and many other factors have been described in its pathogenesis, such as lymphovascular (benign) spreading, mesothelial, epithelial, stem cell or Müllerian duct transformation and genetic factors that explain the appearance of endometriotic lesions outside the pelvic cavity. In endometriosis, the balance between proliferation and apoptosis is upset,

endocrine functions are altered, epigenetic changes occur, and the role of the altered microbiome has been brought to the forefront of research.

However, recently, research on the pathophysiology of endometriosis has focused on the changes of the immune system. The irregular function of some cells of innate and acquired immunity has been observed in patients with endometriosis. However, the question of whether the immune dysregulation is the cause or the consequence of the development of endometriosis is still equivocal.

There is a large amount of literature on cells of acquired immunity, among which the variation of T lymphocytes in peritoneal fluid is a prominent finding.

The shift in the balance between type 1 and type 2 T-helper lymphocytes towards type 2 is a definitive change, may play a role in the pathogenesis of endometriosis. Type 2 T-helper lymphocytes influences the differentiation of B lymphocytes to suppress both cellular and humoral responses, due to the suppression of Th1 lymphocytes by increased levels of IL-10, IL-12 in endometriosis, the Th1/Th2 ratio is shifted in the way of Th2 lymphocytes in the peritoneal fluid, contributing to the elimination of potentially harmful cells checked by the immune system. These kinds of cells can be, for example, the endometrial debris flowing backward through the Fallopian tube to the ectopic site. In endometriosis, number of T-regulatory lymphocytes, which also play a role in maintaining immune tolerance against self-antigens, are increased in the peritoneal fluid but not in the peripheral blood, which may lead to autoimmune-type reactions and a reduced cellular immune response in the peritoneal fluid. The increased incidence of malignant and autoimmune disease in endometriosis may be due to the changes in T lymphocytes mentioned above.

In endometriosis, the balance between NK cell subtypes is broken and the function of the highly cytotoxic CD56dimCD16+ NK cells is inhibited in the peritoneal fluid and peripheral blood of affected women by increased cytokines such as IL-6, IL-15, TGF β . Reduced

cytotoxicity might lead to impaired elimination of ectopically appearing endometrial tissue.

In order to maintain homeostasis, human body has a mechanism which is called phagocytosis. This mechanism found in many cells, but only a few cell types can perform this function with high efficiency, for example: neutrophil granulocytes, monocytes (derived macrophages), dendritic cells, osteoclasts. The function of this mechanism is to recognize pathogens, entered our body, such as foreign and damaged autologous tissues. It has been suggested that endometrial debris that has appeared ectopically may also be considered as damaged autologous tissue.

The researches of innate immunity cells focused mainly on macrophages and neutrophil granulocytes in the peritoneal space.

The number of macrophages is increased in the peritoneal fluid of endometriotic women and also in the environment of ectopically located endometrial layer. Macrophages produce proinflammatory cytokines such as $\text{TNF}\alpha$, IL-6, IL-8, IL-1 β , contributing to the maintenance of the local inflammation that has developed, which attract neutrophilic granulocytes close to the endometriotic lesions, further enhancing the inflammatory milieu, which favors to the increase of pain. They also produce proangiogenic substances crucial for neoangiogenesis, e.g., VEGF. The results of these studies suggest that the phagocytic function of macrophages accumulating in the peritoneal space due to endometriosis is reduced, which may be due to reduced expression of CD-36 scavenger and Annexin A2 receptors, which are essential for the process of engraftment. However, there is no scientific data on the quality and quantity of macrophages, and on their peripheral blood precursors (monocytes) in the in peripheral blood samples, and their functional changes in endometriosis are not fully clarified.

However, these monocytes circulating in the peripheral blood are also capable of phagocytosis, antigen presentation and production of certain cytokines before they exit into the

tissues and transform into macrophages. Under physiological conditions, monocytes participate in the defenses of the human body by phagocytosis and are also capable of antigen presentation and production of chemotactic factors.

Even though neutrophil granulocytes, which are also capable of phagocytosis, are natural immune cells with a major role in the defense against "foreign" cells and tissues, a few is known about their role in the pathophysiology of endometriosis. In endometriosis, the number of neutrophil granulocytes is increased in the peritoneal fluid due to factors produced by epithelial cells such as ENA-78, HNP1-3, which attract these neutrophil granulocytes there. The neutrophil granulocytes, attracted, undergo a functional change, they produce additional neutrophil granulocyte-attracting inflammatory cytokines (VEGF, IL-8, CXCL-10) in the peritoneal cavity, which, due to the locally established inflammation, contribute significantly to the initial progression of endometriosis mainly by creating oxidative stress and inflammatory response.

OBJECTIVES

In our research we sought to answer the following questions:

1. Assessment of phagocytic function in patients with endometriosis

In our studies, we observed whether the phagocytic function of phagocytic cells of the natural immune system capable of phagocytosis is altered in patients with endometriosis, thus possibly leading to reduced elimination of ectopic endometrial tissue, and whether the function studied changes after removal of endometriosis.

1.1 How does the phagocytic function of peripheral neutrophil granulocytes change in women with endometriosis compared to the healthy ones?

1.2 How does the phagocytic function of peripheral monocytes change in women with endometriosis compared to the healthy population?

1.3 How does the phagocytic function of the cells studied change after complete surgical removal of visible endometriosis?

1.4 Does the surgical intervention influence the phagocytic function of these cells?

1.5 Is the change in phagocytic function observed in patients with endometriosis reversible?

2. Plasma tests in patients with endometriosis

We also wanted to see whether the phase 1 studies could be reproduced by plasma tests. Whether incubation in different plasma samples changes the phagocytic function of the cells. Whether plasma from healthy women improves the phagocytic function of cells from endometriotic women, and whether plasma from endometriotic women's blood samples can impair the function of cells from healthy women.

2.1 Does incubation in the blood plasma of women with endometriosis influence the phagocytic function of cells from healthy women?

2.2 Is there an influence on the phagocytic function of cells from healthy women when incubated in the plasma of women who have undergone complete surgical removal of endometriosis?

2.3 Is there an effect on the phagocytic function of cells from women with endometriosis when incubated in their own plasma?

2.4 Is there an effect on the phagocytic function of cells from women with endometriosis when incubated in the plasma of other endometriotic women?

2.5 Is there any influence on the phagocytic function of cells isolated before the surgical removal of endometriosis incubated in the plasma of healthy women?

2.6 Is there any effect on the phagocytic function of cells isolated after surgical removal of endometriosis incubated in the blood plasma of the same woman?

2.7 Is there any effect on the phagocytic function of cells isolated after surgical removal of endometriosis incubated in the plasma of another woman who has also undergone surgery?

2.8 Does incubating cells isolated after surgical removal of endometriosis in the plasma of a healthy woman influence the phagocytic function of the cells?

2.9 Is there an effect of performing the above tests on heat inactivated plasma samples?

MATERIALS AND METHODS

Women involved in the study:

In the first phase of the study, 26 women with possible endometriosis based on symptoms, ultrasound examination and palpation findings were sampled preoperatively, and 13 patients were sampled postoperatively (7th postoperative day) after informed consent from the Regional and Institutional Committee of Science and Research Ethics.

The effect of surgical intervention and anesthesia was investigated by selecting a "surgical control" group of 14 women who were scheduled for laparoscopic surgery for benign gynecological disease. The 14 women in the surgical control group were also sampled pre- and postoperatively. The control group consisted of 23 healthy women.

Inclusion criteria for female patients with endometriosis were laparoscopically removed endometriosis confirmed by histopathological examination, and reproductive age (18-45 years). Since some hypotheses suggest that there may be significant differences in the pathogenesis of early and advanced stage of endometriosis, we took blood samples from patients with surgically confirmed early-stage endometriosis.

Exclusion criteria included smoking, major internal or immunological disease, regular use of hormonal drugs within six months, body mass index over 30 kg/m² and vaccination within six months.

In the surgical control group, inclusion also required minimally invasive laparoscopic surgery, a history of benign gynecological disease confirmed by histopathological examination

after surgery, and childbearing age (18-45 years). The exclusion criteria were the same as those used in the endometriosis group.

In healthy women, the inclusion criteria were reproductive age (18-45 years) and regular menstrual cycle. Exclusion criteria were the same as for the other groups, with the addition of any gynecological pathology.

In the second phase of our study, also with the permission of the Regional and Institutional Committee of Science and Research Ethics and after informed consent of the patients, blood samples were taken from 8 patients with endometriosis on the morning before surgery (preoperative sample) and on the 7th postoperative day (postoperative sample). The control group consisted of 16 healthy women.

The inclusion and exclusion criteria for each group were the same as in the groups studied in the first phase of our studies.

In each of the different groups, blood sampling was performed during the follicular phase of the menstruation cycle.

Laboratory test

Separation of plasma and white blood cells from peripheral blood samples

Blood was taken into EDTA-containing, vacuum-assisted, closed blood collection tubes in each group.

Plasma fractions, which were separated in the second phase of the study from each group, were obtained after centrifugation of the blood samples at 800 G for 10 minutes at 20°C. After the collection of the plasma, it was divided into two equal parts and one half was inactivated in 56°C warm water for 30 minutes to determine the heat resistance of the possible immunomodulatory factors and to exclude the effect of the complement system on the phagocytosis.

After separation of the plasma fractions, Ficoll solutions of different density gradients (1.119 g/cm³ followed by 1.077 g/cm³) were layered and the white blood cells were separated by centrifugation. Layers of monocyte-lymphocyte and neutrophil granulocytes were collected in different tubes. The viability of the cells was 96-98%, checked with the help of trypan blue stain. In the absence of erythrocyte contamination, lysis of red blood cells was not necessary.

In the second phase of the study, all plasma experiments were performed with both inactivated and noninactivated plasma. Separated monocytes and neutrophil granulocytes from women with endometriosis before surgery were incubated in plasma collected from healthy women. Likewise, we incubated separated monocytes and neutrophil granulocytes from women with endometriosis after surgery in plasma from healthy women, and monocytes and neutrophil granulocytes from healthy women were incubated in separated plasma from women with endometriosis before and after surgery. To observe the effect of "foreign" plasma on phagocytic function of monocytes and neutrophil granulocytes, these cells divided from each group were treated with their own plasma (autologous) and with plasma from the same group but from another individual (heterologous). The incubation at 37°C for 60 minutes was followed by plasma depletion to assess phagocytic function.

Cells were stuck onto a cell culture chamber and incubated with the cells after fluorescent labelling and opsonization of Zymosan A particles in buffer containing fluorescein isothiocyanate. The fluorescence of non-phagocytosed particles was quenched by trypan blue staining. Fixation was performed in 4% paraformaldehyde solution. This was followed by indirect immunofluorescence labeling of monocytes with mouse CD14⁺ monocyte specific antibodies followed by fluorescently labeled (Dylight 594) mouse IgG antibody secondary antibodies. The labeling marked the cell membrane of monocytes under a fluorescence microscope by red color, in order to identify monocytes. In the last step, fluorescent labeling of

nuclei of both neutrophil granulocytes and monocyte was performed with a cover medium containing 4',6-diamino-2-phenylindole.

Samples were evaluated in randomly selected fields of view using an Axioplan fluorescence microscope (Zeiss, Oberkochen, Germany). In each field of view 100 cells were examined and particles were counted. The particles phagocytosed by 100 cells were added together and divided by 100 to calculate the phagocytosis index, which was the average number of Zymosan particles phagocytized by a cell. In the fields of neutrophil granulocytes, cells were identified by their morphological characteristics, and monocytes was marked by the previously described way.

Statistical analysis

Results are presented as mean (\pm standard deviation). The distribution of the sample was checked by the Shapiro-Wilk test and was found to be normal. In both research phases, one-way ANOVA was used to compare the clinical data of the participants. The pre- and postoperative values of the same group were tested by using two-sample t-test. Repeated measures of ANOVA were used to compare the phagocytosis index values obtained by treating monocytes and granulocytes of the same women (endometriosis patients, healthy) with different plasma (autologous, heterologous, pre-, and postoperative). Two-sample t-test was used to compare treatment with inactivated and noninactivated plasma for each combination. A value of $p < 0.05$ was considered statistically significant.

RESULTS

The age, body mass index, gravidity and parity of the women studied did not differ significantly between the different groups at any stage of the research.

Results of the first phase of the study

The values of phagocytosis indices of monocytes and neutrophil granulocytes isolated from peripheral blood samples collected before surgery from endometriotic patients were

significantly ($p < 0.001$) decreased compared to the values of phagocytosis indices of monocytes and neutrophil granulocytes from blood samples of healthy women. Compared to the preoperative results in patients with endometriosis, the phagocytosis indices of monocytes and neutrophil granulocytes separated from samples taken on 7th postoperative showed significant ($p < 0.001$) increase compared to the preoperative values. The phagocytosis indices of healthy women and values from postoperative endometriosis patients did not differ ($p = 0.060$) significantly. In the control group with surgery, there was no significant difference ($p = 0.731$) between the phagocytosis indices of monocytes and neutrophil granulocytes from blood samples taken preoperatively and postoperatively ($FI = 3.35 \pm 0.52$).

Results of the plasma experiments

Results of phagocytosis indices of neutrophil granulocytes and monocytes from healthy women

When neutrophil granulocytes from healthy women were incubated in inactivated and noninactivated plasma isolated before the surgery from women with endometriosis, a significant decrease in phagocytosis indices of the cells was observed ($p < 0.001$) compared to phagocytosis indices measured after incubation in autologous and heterologous plasma samples from healthy women. The phagocytosis indices of the examined cells did not differ significantly when neutrophil granulocytes from healthy women were incubated in autologous and heterologous plasma and compared to the effect of incubation in isolated plasma samples on the 7th postoperative day of women with endometriosis.

A review of the results of the monocyte phagocytosis indices showed that treatment with plasma samples isolated from endometriosis patients before surgery significantly reduced (inactivated, non-inactivated: $p < 0.001$) the phagocytosis indices of monocytes from healthy women compared to the results after incubation in autologous and heterologous plasma samples. The phagocytosis indices of monocytes from healthy women were not significantly

different when cells were incubated in plasma samples isolated from autologous, heterologous or endometriotic patients after surgery.

Phagocytosis index of neutrophil granulocytes and monocytes isolated before surgery in women with endometriosis

After treating preoperatively isolated neutrophil granulocytes in plasma samples from healthy women with endometriosis, we observed a significant increase ($p < 0.001$) compared to treatments in inactivated and non-inactivated, autologous, and heterologous plasma samples from women with endometriosis after surgery.

After treatment in autologous and heterologous plasma samples isolated preoperatively from endometriosis patients, phagocytosis indices of monocytes were significantly decreased ($p < 0.001$) compared to the incubation in inactivated and non-inactivated plasma samples from healthy women.

Phagocytosis indices of neutrophil granulocytes and monocytes collected after surgery from women with endometriosis

There was no significant difference in the phagocytosis indices of neutrophil granulocytes isolated on the seventh day after surgery from women with endometriosis in autologous and plasma samples from the same population (inactivated: $p=0.655$ and 0.050 ; non-inactivated: $p=0.504$ and 0.845) compared to post-treatment results in plasma samples from healthy women.

Similarly, there was no significant difference in the results of phagocytosis of monocytes isolated from endometriotic women after surgery (inactivated: $p=0.517$ and 0.300 ; non-inactivated: $p=0.869$ and 0.072) compared to the results after incubation in plasma samples from autologous, heterologous, and healthy women.

For the plasma results, there was no significant difference in phagocytosis indices following incubation in inactivated and noninactivated plasma samples, similarly no significant difference was found following incubation in autologous and heterologous plasma samples.

DISCUSSION

The prevalence of endometriosis in women of reproductive age is around 10%, and half of women with chronic pelvic pain and/or infertility is diagnosed with endometriosis. The significant deterioration in quality of life caused by the symptoms and the public health impact of the disease are now unquestionable.

Current knowledge suggests that changes in innate and acquired immunity and immunosurveillance may play a pivotal role in the pathogenesis of endometriosis. Alterations of the immune function may contribute to the ability of ectopic endometrial tissue to adhere to, grow and damage the affected areas through retrograde menstruation or other kinds of dissemination. Despite of intensive research, the mechanism that prevents the removal of endometrial glands and stroma by immune cells outside the uterine cavity is still poorly understood.

There is great amount of data on alterations of the immune cells in peritoneal fluid in women affected by endometriosis, in particular regarding the functions of T lymphocytes. An acute inflammatory response is generated in the peritoneal fluid and in the area affected by endometriosis, which attracts and activates T-helper and T-regulatory cells. Following a cyclical decrease in acute inflammation, a dysfunctional role of monocytes/macrophages dominates the affected area by inducing and maintaining chronic inflammation, facilitating vascular remodeling, with consequent adhesions and scarring, which in the end cause clinical signs (chronic pelvic pain and infertility). Impaired cytotoxicity of NK and T cells in both peritoneal fluid and peripheral blood, which may be influenced by the immunomodulatory factors observed in the blood of women with endometriosis. It has been observed that the

number of innate immune cells, neutrophil granulocytes, is increased in the peritoneal cavity in the early stages of endometriosis, but their number decreases intraperitoneally in advanced stages of the disease. In addition to the change in the number of cells, their function is also altered, so the pro- and anti-inflammatory cytokines IL-1, IL-4, IL-6, IL-8, IL-10 and TNF α are produced in the serum and peritoneal fluid of women with endometriosis, which is more pronounced in advanced stages. Regarding immunomodulatory factors, plasma concentrations of some growth and angiogenesis-promoting factors (TGF- β , IGF-1, HGF and VEGF) are also increased compared to healthy individuals. However, only a few studies mention that the elimination of endometriosis by immune cells is likely to be impaired, which may contribute to the persistence and progression of endometriosis.

In macrophages, appearing in the peritoneal fluid of endometriotic women, the expression of CD-36 scavenger receptors, crucial for phagocytosis, and thus the activation of matrix metalloproteases is reduced, which may be due to increased levels of prostaglandin E2, circulating in the serum of endometriotic women. There is also an increase in the number of macrophages and the amount of proinflammatory cytokines they secrete in endometriosis, but the differentiation of macrophages is towards the M2 subtype, which favors cell proliferation, rather than the opposite M1 subtype. It is hypothesized that monocytes and their differentiating macrophages, as well as neutrophil granulocytes, which are important for phagocytosis, may play a role in the elimination of ectopic endometrium. The study of immune cell function in the peripheral blood of patients with endometriosis is a less researched area, and thus relatively just a few literatures are available.

In line with our hypothesis, in the first phase of our study we investigated the phagocytic function of neutrophil granulocytes and monocytes in peripheral blood of women affected by endometriosis before and after surgery, comparing to healthy women and surgical control group. The changes observed in the first phase of our studies provided the basis for their

continuation, i.e., plasma experiments in which cells from each study group were treated with plasma from their own and other groups.

Our results indicate that the phagocytic function of neutrophil granulocytes and monocytes in the peripheral blood of women with endometriosis is significantly reduced compared to the phagocytic function observed in healthy women. This reduction is reversible, as it returns to normal following surgical removal of endometriosis. It can be concluded that laparoscopic surgery has no effect on the phagocytic function of the cells, in these cases the function tests are similar to the results of the healthy control group. Thus, the decrease in phagocytic function of peripheral neutrophil granulocytes and monocytes is likely to be caused by the factor(s) produced by endometriosis.

To confirm our hypothesis that these endometriosis-derived factors affecting phagocytic function are also present and present their effects in the peripheral blood, we performed plasma experiments. These studies confirmed the presence of immunomodulatory factor(s) circulating in the peripheral blood of endometriotic patients, as the phagocytic function of cells from all groups is reduced when they were incubated in the plasma of women with endometriosis. Further evidence of reversible impairment of function is that the reduced phagocytic function of cells from endometriotic patients is normalized when cells are incubated in the plasma of any of the other groups. To ensure the reliability of the data, the studies were also performed with autologous plasma, but in none of the cases did it alter the function tested, i.e., the conditions of the experimental work did not affect the phagocytic function. For a similar purpose, the studies were also performed with heat-inactivated plasma samples, to exclude any possible effect of the complement system on immune function. Although our results suggest that the complement system has no effect on phagocytic function, as there is no difference in the results of experiments with inactivated and noninactivated samples, our hypothesized factors were found to be heat stable.

Our results support our hypothesis that ectopic endometrium and/or their microenvironment may produce immunosuppressive factors that reduce the phagocytic function of monocytes and neutrophil granulocytes circulating in peripheral blood.

The identification of immunosuppressive factors in the plasma of women with endometriosis may be the subject of further investigations. It can be hypothesized that in addition to elevated pro- and anti-inflammatory cytokines (e.g., IL-10) present in the blood of endometriosis patients may influence phagocytic function. Based on our results, we hypothesize that the elimination of endometrial cells from the abdominal cavity is disturbed due to impaired immune function, and the phenomenon described may be part of the progression of endometriosis.

Our research may also highlight that not only immune dysfunction may cause the development of endometriosis, but endometriosis may also affect immunological function.

SUMMARY

Endometriosis is a benign, chronic condition that negatively affects the quality of life of women affected and also responsible for infertility in many women. The pathophysiology of the disease is not fully understood, but an impaired immune response may play a key role in it. Phagocytic function of the innate immune system may play a role in ectopic endometrial elimination. The aim of our studies was to investigate the phagocytic function of neutrophil granulocytes and monocytes. In the first phase of our studies, we examined and compared the phagocytic function of cells from healthy women and women undergoing surgery for other benign gynecological conditions before and after surgery. In the second phase of our work, we performed plasma experiments with inactivated and noninactivated plasma samples. A total of 34 preoperative and 22 postoperative blood samples were taken from women with endometriosis, 32 healthy women served as controls, and 14 women who had undergone surgery for benign gynecological diseases had pre- and postoperative blood samples taken as a

surgical control group. The cells were isolated and incubated in different plasma samples, after which their phagocytosis indices were determined under fluorescence microscopy.

In patients with endometriosis, neutrophil granulocyte and monocyte phagocytic function was significantly reduced compared to healthy women. When the cells with reduced phagocytic function were incubated in plasma samples from healthy women, the function tested normalized. In addition, when cells from healthy women were treated with plasma samples from endometriotic women, phagocytic function was significantly reduced. There was no difference between the results of heat-incubated and nonincubated plasma samples.

Our results suggest that active endometriotic lesions may produce thermostable, immunomodulatory factors that reduce the phagocytic function of neutrophilic granulocytes and monocytes found peripherally. This function normalizes after complete surgical removal of endometriosis and returns to levels measured in healthy women.

PUBLICATIONS



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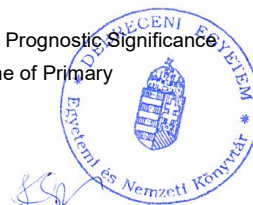
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List of publications related to the dissertation

1. **Lukács, L.**, Kovács, A. R., Pál, L., Szűcs, S., Lampé, R.: Evaluating the Phagocytic Index of Peripheral Leukocytes in Endometriosis by Plasma Experiments. *Medicina (Kaunas)*. 58 (7), 1-12, 2022.
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IF: 2.156

List of other publications

3. Bélteki, B., Boldogh, B. Z., Lipták, M. G., **Lukács, L.**, Lampé, R., Török, P.: A meddőség és a méh strukturális rendellenességeinek lehetséges kapcsolata. *86*, 254-259, 2023.
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9. Kovács, A. R., Pál, L., Szűcs, S., **Lukács, L.**, Kövér, Á., Krasznai, Z. T., Lampé, R.: Petefészekrákos betegek perifériás monocitáinak és neutrofil granulocitáinak fagocita-funkciója. *Nőgyógy. Onkol.* 23, 54-58, 2018.

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