

# ENDOMETRIOSIS AND THE HUMAN MICROBIOME

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## Abstract

The dysbiosis of the intestinal microbiome is associated with various diseases. It is important to understand the factors that influence the intestinal microbiome and the microbiome regulation strategies to increase therapeutic responses.

Endometriosis affects about 10% of women of childbearing age. Among those affected by endometriosis, up to 50% of them suffer from chronic pelvic pain and / or infertility. Endometriosis is characterized by inflammation and oestrogen dependence.

Endometriosis is a condition that affects fertility, creates a state of "congestion" with an excessive growth of the endometrial tissue in many other areas of the body.

The microbiota plays a role in the occurrence of endometriosis by affecting the epigenetic, immunological and / or biochemical functions of the host. Intestinal bacteria are involved in the oestrogen metabolism. The oestrogen-microbiome link (or the stroboloma) helps explain where the excess oestrogen comes from.

Endometriosis appears to be associated with the increased presence of Proteobacteria, Enterobacteriaceae, Streptococcus spp. and Escherichia coli in various areas occupied by the microbiome.

Further studies are needed to analyse the association between endometriosis and microbiota. Endometriosis is influenced by diet.

## Keywords:

*endometriosis, microbiome, endometrial microbiota*

## Introduction

The study of the microbiome, a true universe, began two decades ago, so most studies are limited and the results are preliminary.

Researchers have observed the link between the microbiome and some diseases, but it is not yet possible to draw cause-effect conclusions about this vast and heterogeneous group of microorganisms and their influence on the body.

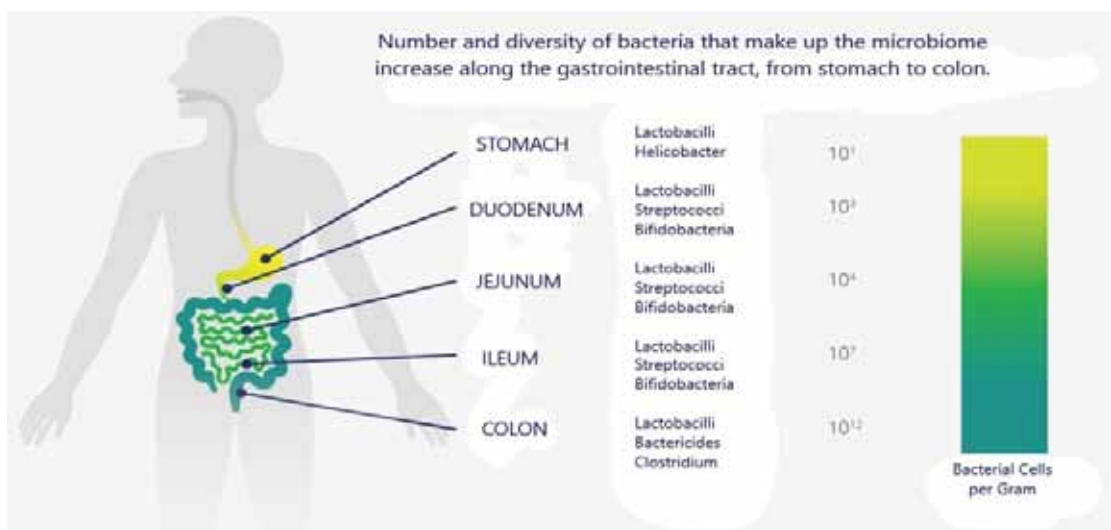
An imbalance in the microbiome (dysbiosis) can lead to a wide range of health problems, including endometriosis.

In the not too distant future, according to the most enthusiastic researchers, the administration of prebiotics (compounds that serve as a substrate for the growth of beneficial bacteria), probiotics (beneficial bacteria themselves) or faecal transplantation (stool rich in microorganisms from healthy donors) could

become a routine, so we could hope to operate at full capacity, from the inside out. Each individual has a unique community of bacteria. When we talk about the microbiome, we refer mainly to the digestive tract, where over 90% of the microorganisms in the human body live (Fig.1).

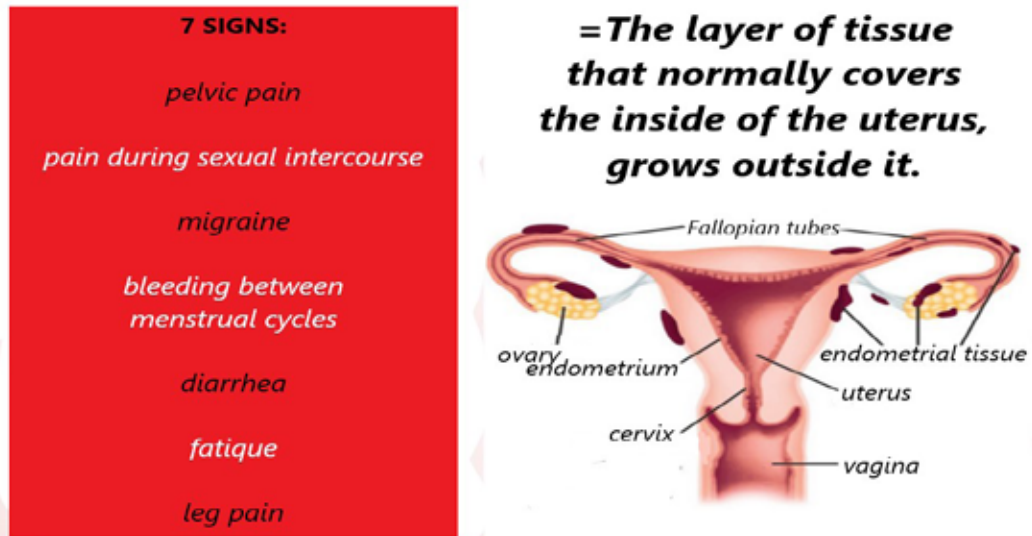
The most fascinating aspect is that each individual has a specific mixture of bacteria, different from that of the others. Endometriosis, like a growing number of other medical conditions, indicates the imbalance of the intestinal microbiome. Infertility is common, being obvious in about 50% of women with endometriosis.

Endometriosis is a common gynaecological condition, defined by the presence of endometrial-like tissue outside the uterus, most commonly on the pelvic and ovarian viscera, which is associated with pelvic pain and infertility (1-6).



**Fig. 1** Distribution of the intestinal microbiome  
( translated after: <https://www.smikactiv.ro/microbiomul-intestinal/>)

## ENDOMETRIOSIS



**Fig. 2** Factors that determine endometriosis

(adapted after: <https://ro.pinterest.com/pin/834925218404085358>)

Endometriosis is a multifactorial disease (Fig.2) The aetiology and pathophysiology of the disease are still insufficiently elucidated. The pathogenesis of endometriosis is complex.

Genetic and immunological factors play a key role in the pathophysiology of the disease.

There has been shown that an association between the presence of endometriosis and the altered immune system exists.

The theory of the association of an altered immune system and the endometriosis suggests that changes in cell-mediated immunity and / or humoral immunity may contribute to the onset of the disease.

Recently, endometriosis has been considered an autoimmune condition. An abnormal antigen-antibody reaction may occur in the case of endometriosis (increased incidence of

autoantibodies in women with endometriosis), the patient with endometriosis has autoimmunity to the endometrial autoantigens, ovarian and nuclear antigens.

Endometrial autoantigens to which patients with endometriosis have autoimmune reactions are endometrial transferrin and alpha 2-HS glycoprotein. Antibody titres to these proteins are specifically elevated in endometriosis. Transferrin and alpha 2-HS glycoprotein levels are significantly increased in the peritoneal fluid of the patient with endometriosis. Antibodies to transferrin and alpha 2-HS glycoprotein inhibit sperm motility in vitro.

These data suggest that autoantibodies to the endometrium play an important role in the infertility frequently associated with endometriosis.

The patient with endometriosis has in the

serum and in the vaginal and cervical discharge IgG and IgA autoantibodies against the endometrial and ovarian tissue; endometriosis is associated with other autoimmune diseases and with recurrent immune-mediated abortions.

The presence of IgG, IgM, IgA autoantibodies against phospholipids and histones has been reported. Autoantibodies have titres that correlate with the severity of endometriosis.

These data are apparently contradictory to the reduced natural cell-mediated immunity observed during the disease. Anti-endometrial antibodies especially the IgG and IgA have been detected in the serum, the vaginal and cervical discharges of patients with endometriosis (sensitivity 0.84; specificity 1.00).

The presence of antinuclear antibodies was observed in approximately 30% of patients with endometriosis. IgG autoantibodies to phospholipids have been frequently described, followed by antibodies to histones and nucleotides in the order of their frequency. Histone antibodies are in the IgG, IgM and IgA classes. The incidence of IgM autoantibodies was reversed, antinucleotides occur more frequently and antiphospholipids less frequently.

A close correlation was noted between the presence of the anticoagulant lupus and the antinuclear antibodies with IgG and IgM.

These observations suggest that endometriosis is associated with abnormal polyclonal activation of B cells, a classic feature of autoimmune diseases. IgG titres are elevated in patients with endometriosis.

The woman with endometriosis has a reduction in the haemolytic activity of total serum complement (CH50) and the C3 component of the reduced complement. C3 deposits

are low in the endometrium, corresponding to the reduction of CH50 titres.

Numerous changes in cell-mediated immunity have been reported in women with endometriosis. There is a reduction in the proliferation of lymphocytes in the peripheral blood in the response to endometrial antigen recognition. Numerous data suggest that systemic T cell activity influences the pathogenesis of endometriosis. The serum Th/Ts ratio is increased in the peripheral blood, peritoneal fluid, and the endometriosis tissue.

NK (high volume granular lymphocyte) cells have certain characteristics in endometriosis. The number or the percentage of NK cells in a woman with endometriosis may be reduced, increased, or unchanged.

There is a reduction in the activity of NK cells in both peritoneal fluid and serum. The cytotoxic activity of NK cells is reduced compared to the autologous and heterologous endometrium, probably determined by the secretions of monocytes / macrophages that modulate immune and non-immune cells. Both peripheral monocytes and peritoneal macrophages show increased activity.

An increase in the number and activity of peritoneal macrophages was observed. Associated with the increase of macrophages activity is the increase in the release of their products, such as the growth factors and cytokines.

With a role in stimulating endometrial cell proliferation and endometrial cell implantation, tissue remodelling is increased due to increased expression of matrix metalloproteins (MMPs) and increased angiogenesis of the ectopic endometrial tissue. Activated macrophages (cytokine release and growth factors) may contribute to the evolution of the disease.

The cytotoxicity of NK cells is inversely correlated with the stage of the disease. Altered NK cytotoxicity to endometrial tissue may be partly responsible for the onset, spread, and progression of pelvic endometriosis. The cytotoxicity of peritoneal NK cells correlates inversely with the severity of endometriosis. The low NK cytotoxicity allows the establishment of this tissue inside the peritoneal cavity.

**The role of cytokines** and growth factors is obvious. Cytokines are likely responsible for the endometrial cell proliferation and endometrial cell implantation.

Increased ectopic endometrial tissue angiogenesis and neovascularization are probably the most important effects of cytokines on the ectopic endometrial tissue.

Cytokines play a major role in initiating, propagating and regulating immune and inflammatory responses.

A cytokine with an important role in the pathophysiology of autoimmune diseases is the tumour necrosis factor (TNF- $\alpha$ ), which is a product of activated macrophages.

TNF- $\alpha$  induces the production of other pro-inflammatory cytokines such as IL-1, IL-6. It is obvious that TNF- $\alpha$  plays an important role in the pathophysiology of endometriosis.

Numerous studies have shown that the level of TNF- $\alpha$  is increased in the peritoneal fluid of patients with endometriosis. TNF- $\alpha$  stimulates the adhesion of endometrial cells and also induces the increase of MMP expression.

The activation of immune cells leads to the release of the inflammatory cytokine cascade.

The increase of the peritoneal inflammation is demonstrated by the increase in the level of cytokines in the peritoneal fluid.

It is uncertain whether the elevated level of cytokines or the inflammation is the cause or consequence of the disease.

It is clear that cytokines may have effects leading to the evolution of endometriosis (e.g. cytokines may stimulate the adhesion of endometrial cells to the peritoneal mesothelial cells monolayer *in vitro*).

Both *in vitro* and *in vivo* studies show that endometriosis is an invasive disease. MMPs play an important role in the pathophysiology of endometriosis, which are produced by the endometriotic tissue that expresses increased MMP activity compared to women without endometriosis.

In the culture, endometriosis cell lines exhibit epithelial-like morphology and immunoreactivity for cytokeratins 8, 18, 19 vimentin, and class I HLA antigens.

Another similarity between endometriosis and autoimmune diseases is the disorder of the apoptosis process (the exact mechanism is still unknown).

The immune dysfunction is either a cause or a consequence of the disease. Some of the immune disorders have been involved in impairing the reproductive system, resulting in infertility. The severity of the disease is variable, from moderate to severe (stage I-IV). There is a weak correlation between the severity of the disease and the patient's symptoms.

All microorganisms, i.e. bacteria, archaea, yeasts and viruses, which live inside and on the human body, make up the human microbiota. The collective genome of microorganisms is called the microbiome (27).

The detection of bacteria with molecular techniques allowed the study of the micro-

biome in tissues and organs previously considered sterile, such as the endometrium. The dysbiosis of the intestinal microbiome and in the genital tract may be associated with endometriosis.

Endometriosis is a common gynaecological condition with a complex, multifactorial aetiology. Subsequently, an abnormal endometrial microbiota has been associated with insufficient implantation, pregnancy loss and other gynaecological and obstetric conditions. The endometrial tissue is usually limited to the uterus. In endometriosis, the endometrial tissue may exist in other areas, including the ovaries and fallopian tubes, as well as in the nearby and more distant tissues (7-12).

While endometriosis is associated with dysmenorrhea, pelvic pain, dyspareunia, and / or infertility, some women with endometriosis are asymptomatic (13,14).

The presence of viable endometrial cells in the pelvis caused by retrograde menstruation is the most plausible theory that explains the pathogenesis of the disease (15).

However, the occurrence of endometriosis in 1-2% of women, despite all women having some degree of retrograde menstruation, suggests other mechanisms that play a role (16)

The new aspect of endometriosis is an inflammatory disease of the whole body. The influence of the microbiome on immunomodulation and the development of several inflammatory diseases is well established.

In contrast, little is known about the composition of the microbiome along the female reproductive tract and its role in the development of endometriosis or other gynaecological conditions (17).

The cause of this disease is not fully known, but it may be related to an imbalance of the immune system. There may be a genetic component too.

Endometriosis is associated with the dysbiosis of the intestinal microbiota. There are two main roles of the microbiome in the endometriosis: oestrogen detoxification and immune regulation.

The intestinal microbiota is involved in oestrogen metabolism. Endometriosis is characterized by both inflammation and oestrogen dependence, but is not caused by oestrogen. Interestingly, the microbiome influences oestrogen metabolism and oestrogens influence the intestinal microbiota (18).

Given that endometriosis is an oestrogen-dominated condition, intestinal dysbiosis leading to abnormal circulating oestrogen levels could contribute to the onset of this disease (12).

There is a complex two-way interaction between endometriosis and the microbiome. Research on dysbiosis in the microbiome in relation to the symptoms and severity of endometriosis has increased and it is clear they are linked.

There are a number of risk factors associated with endometriosis, including an increased risk in Asian women, excessive oestrogen exposure, and a low body mass index. Bacteria in the microbiome help digest food, regulate the immune system, protect against other disease-causing bacteria and produce vitamins, including B vitamins (B12, thiamine and riboflavin) and vitamin K, which is needed for blood to clot.

Endometriosis is a problem related to both an oestrogen-dominated and inflammatory fer-

tivity, often accompanied by deficient immunity, pain and irregular bleeding, digestive problems, constipation, nausea, bloating, etc.

The diagnosis of endometriosis is difficult to establish. Endometriosis is characterized by inflammation with elevated levels of immune mediators, reduced regulatory T cells in the periphery and endometrium, increased regulatory T cells in the ectopic endometrial tissue. Endometriosis appears to be associated with elevated levels of Proteobacteria, Enterobacteriaceae, Streptococcus and E. coli with different locations of the microbiome.

A dysfunctional immune response appears to play a significant role and there is some evidence to suggest that the microbiome may modulate the immune response in endometriosis (17).

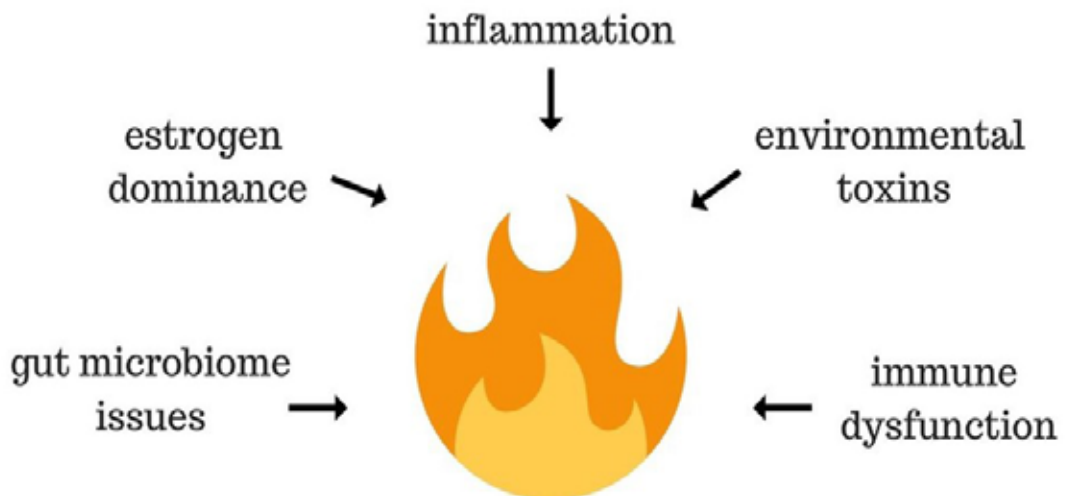
Theories state that dysbiosis in the microbiome could be a cause of inflammation.

This can trigger the symptoms of endometriosis (18, 19) (Fig.3).

However, it is suggested that the dysbiosis of the intestinal microbiome that promotes oestrogen deconjugation, which leads to increased circulatory levels, may contribute to a hyperestrogenic environment that underlies the evolution of endometriosis (18).

Recent studies suggest that the intestinal microbiota plays another crucial role in the human body by regulating the level of the circulating oestrogen. The stroboloma is the community of microbes capable of metabolizing the oestrogen. The stroboloma modulates the enterohepatic circulation of oestrogen and affects circulating and excreted oestrogen levels.

The stroboloma of women with endometriosis may have a higher number of bacteria that produce beta-glucuronidase, which leads



**Fig. 3** Factors that determine endometriosis

(after: <https://www.kaelyrd.com/blog/functional-nutrition-for-endometriosis>)

to increased levels of circulating oestrogen, which leads to endometriosis. Vaginal and endometrial dysbiosis, including decreased *Lactobacilli* and increased pathogenic gram-negative bacteria, has also been detected in women with endometriosis and may contribute to hormonal imbalance.

When the intestinal microbiome is balanced, the stroboloma produces the right amount of beta-glucuronidase to maintain oestrogenic homeostasis. However, when intestinal dysbiosis is present, beta-glucuronidase activity may be altered. It produces either a deficiency or an excess of free oestrogen, thus favouring the development of oestrogen-related pathologies. Intestinal dysbiosis may have the potential to alter the stroboloma, disrupt oestrogen homeostasis and affect these processes, causing the development of endometriosis (19-21).

The pathogenesis of this enigmatic disorder remains controversial, despite extensive research. Several studies have shown a relationship between endometriosis and autoimmune diseases. There is also evidence that, especially the intestinal microbiota, influences the pathogenesis of some autoimmune diseases.

Endometriosis has also been considered to be an autoimmune disease because it is often associated with the presence of autoantibodies, other autoimmune diseases, and possibly immune-mediated recurrent abortion (22-26).

The purpose of this systematic review was to understand the two-way interaction between the microbiome and endometriosis. While the microbiome could be a useful screening / diagnostic tool for endometriosis, it could become a therapeutic target. Whether the vaginal

and / or cervical microbiota varies throughout the menstrual cycle is controversial.

Further studies may clarify whether the association is causal and whether dysbiosis leads to endometriosis or endometriosis leads to dysbiosis. Investigating the microbiome of the reproductive tract will allow a better understanding of the role of bacterial communities in both physiology and pathophysiology, which in turn has an impact on the ability to achieve pregnancy and maintain a healthy pregnancy.

To alleviate this condition, we need to change diet by reducing the consumption of processed foods that contain artificial additives, avoiding the consumption of gluten and including more fruits and vegetables in our diet.

In general, the consumption of processed foods containing artificial additives should be reduced. Some nutritionists have found that the major part of women with endometriosis have a degree of intolerance to gluten-containing foods, and when this protein was removed from the diet, the disease was observed to improve after a strict, long-term diet.

Therefore, the consumption of flour, bread, pastries, pasta, couscous, fermented beverages made of these cereals, dough-based dishes, certain sausages, cheeses and pastries, sweets, etc. should be avoided or reduced.

## Conclusions

Endometriosis is a disease whose pathophysiology is still unclear.

The immune system plays a significant role in the pathogenesis of endometriosis, there are



similarities in the pathophysiology between endometriosis and autoimmune diseases. B cell function is increased in patients with endometriosis.

The increased frequency of autoantibodies supports the hypothesis that endometriosis may be an autoimmune disease. Endometriosis associates repeated abortions, infertility, which could be explained by the presence of autoantibodies.

Based on recent data (markers of immune reactivity), endometriosis has begun to be treated as an autoimmune disease. However, it is uncertain whether endometriosis is an autoimmune disease.

Endometriosis, an oestrogen-induced condition characterized by the growth of endometrial tissue outside the uterus, has been associated with intestinal dysbiosis. Endometriosis is a condition dominated by oestrogen. The intestinal microbiota is involved in the onset and evolution of endometriosis.

Laboratory and clinical studies show that there are indeed differences between the microbiome of patients with and without endometriosis. The microbiome may be involved in the pathogenesis of endometriosis. In the intestinal microbiome there is "the stroboloma", which includes the enteric microbial genes whose products have the ability to metabolize oestrogen in the intestine.

In the future, more in-depth studies should consider the evaluation of the human microbiome, especially in the follicular stage of the menstrual cycle, and its role in the pathogenesis of endometriosis.

## Conflict of interest

The author has no conflict of interest to make the declaration, had full access to all the data in the study and takes responsibility for the accuracy of the data analysis.

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