Endometriosis: Interaction of Immune and Endocrine Systems

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ABSTRACT

Endometriosis is a common gynecologic disorder characterized by the presence of endometrial tissue outside the uterine cavity. Although no single theory can explain all cases of endometriosis, the most commonly accepted theory is Sampson's theory of retrograde menstruation. Retrograde menstruation occurs in 76 to 90% of women. The much lower prevalence of endometriosis suggests that additional factors determine susceptibility to endometriosis. Endometriosis is associated with changes in both cell-mediated and humoral immunity. Impaired natural killer cell activity resulting in inadequate removal of refluxed menstrual debris may play a role in the development of endometriotic implants. Moreover, although the peritoneal fluid of women with endometriosis contains increased numbers of immune cells, these seem to facilitate rather than inhibit the development of endometriosis. Macrophages that would be expected to clear endometrial cells from the peritoneal cavity appear to enhance their proliferation by secreting growth factors and cytokines. Although it is unclear whether these immunologic alterations induce endometriosis or are a consequence of its presence, they appear to play an important role in allowing endometriosis implants to persist and progress and contribute to the development of associated infertility and pelvic pain. Danazol and gonadotropin-releasing hormone (GnRH) agonists are commonly used for the medical treatment of endometriosis. These medications seem to down-regulate cellular and humoral immune responses concomitant with their effect on endometriotic implants. Immunomodulatory effects of danazol and GnRH agonists are likely to contribute to the observed clinical improvement associated with their use.

KEYWORDS: Endometriosis, immune response, cytokines, treatment of endometriosis

Endometriosis is a common gynecologic disorder characterized by the presence of endometrial tissue outside the uterine cavity. Various theories have been put forth to explain the mechanisms for the development of this disease. Although no single theory can explain all cases of endometriosis, the most commonly accepted theory is Sampson's theory of retrograde menstruation.[1] This theory suggests that endometriotic implants reach their most common site of implantation, the peritoneal cavity, by traveling through the fallopian tubes during menstrual shedding. Sampson's theory is supported by the finding of viable endometrial tissue in the peritoneal fluid that is capable of growth[2–5] and the dependent anatomical distribution of endometriotic implants.[6]

Retrograde menstruation occurs in 76 to 90% of women.[7,8] The much lower prevalence of endometriosis,

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6.2 to 8.2%,[9,10] suggests that additional factors are involved in determining susceptibility to endometriosis. Impaired immune response resulting in inadequate removal of refluxed menstrual debris has been proposed as a possible causative factor in the development of endometriosis.

The immune system consists of cells and molecules organized primarily to protect the body from infections. There are two fundamentally different types of immune responses: innate (natural) and acquired (adaptive). The innate immune response consists of all the immune defenses that lack immunologic memory. Thus, innate responses occur to the same extent independent of how many times the infectious agent is encountered. Acquired immune response differs from innate immune response by its ability to improve upon repeated exposure to a given infectious agent.

Endometriosis is associated with changes in both cell-mediated and humoral components of innate and acquired immunity. Although the peritoneal fluid of women with endometriosis contains increased numbers of immune cells, these seem to facilitate rather than inhibit the development of endometriosis. Leukocytes that would be expected to clear endometrial cells from the peritoneal cavity appear to enhance their proliferation by secreting growth factors and cytokines. Although it is unclear whether these immunologic alterations induce endometriosis or are a consequence of its presence, they appear to play an important role in allowing endometriosis implants to persist and progress.

The pelvic inflammation in women with endometriosis also seems to contribute to the development of their most common complaints: pain and infertility. Secretory products of immune cells in the peritoneal fluid such as cytokines and prostaglandins contribute to dysmenorrhea that may progress to dyspareunia and chronic pelvic pain. Pelvic inflammation may also lead to adhesion formation and scarring and disrupt fallopian tube patency. Similarly, the inflammatory environment may impair folliculogenesis, fertilization, and embryo implantation, resulting in infertility.

In this review we summarize the alterations in the immune parameters of women with endometriosis. We also discuss how they may play a role in the pathogenesis of endometriosis and their response to hormonal treatment.

CELLULAR IMMUNE RESPONSE

Macrophages

Macrophages are a key component of the innate immune system. They originate in the bone marrow, circulate as monocytes, and then migrate to tissues and body cavities where they function primarily as phagocytes. They defend the host by recognition, phagocytosis, and destruction of offending microorganisms. They also act as scavenger cells, eliminating cellular debris and apoptotic cells. Macrophages are capable of secreting various growth factors, cytokines, complement components, prostanoids, and hydrolytic enzymes. These secretory products help mediate the protective and scavenger function of macrophages in addition to promoting growth and proliferation of other cell types.

Macrophages are the most abundant nucleated cells found in peritoneal fluid[11] and their association with the development of endometriosis has been investigated intensively. Haney et al[12] first reported an increase in the number of peritoneal macrophages of infertile women with endometriosis. Subsequent studies confirmed an increased number and activity of peritoneal fluid macrophages in women with endometriosis.[13–17] The extent of endometriosis does not appear to correlate with the macrophage count, although a tendency toward higher numbers has been observed in women with minimal and mild stages of disease.[13]

Whereas the increased number and activity of peritoneal fluid macrophages in women with endometriosis would be expected to facilitate the clearance of ectopic endometrial cells and slow down or inhibit the development of endometriosis, they seem to promote growth of ectopic endometrium. This effect may be due to an increase in the release of growth-promoting cytokines and growth factors[18] combined with an impaired scavenger function.

Scavenger function is one of the vital functions of macrophages. It involves phagocytosis and allows macrophages to eliminate invading foreign material as well as cellular debris and apoptotic cells.[19,20] A variety of surface receptors mediate this activity.[19–22] These receptors are regulated by a number of factors, including cytokines and growth factors.[23–25] Abnormal levels of these cytokines and hormones present in the peritoneal fluid of women with endometriosis may impair the scavenger function of macrophages.[26] Another determinant of macrophage scavenger receptor function is the interaction between macrophages and extracellular matrix components. When macrophages are not adherent to the extracellular matrix, they do not express type A scavenger receptors.[27] Hence, in contrast to tissue macrophages, peritoneal fluid macrophages in women with endometriosis that are not in contact with extracellular matrix may not be competent scavengers despite their differentiated status.

Secretory products of peritoneal macrophages and circulating monocytes of women with endometriosis seem to mediate growth and maintenance of ectopic endometrium.[18] Peritoneal fluid from women with endometriosis stimulates proliferation of cultured endometrial stromal cells.[28] Peripheral blood monocytes obtained from women with endometriosis enhance proliferation of cocultured autologous endometrial cells, whereas monocytes from fertile women show the opposite effect and sup-

press endometrial cell proliferation.[29] In addition to their growth-stimulatory effect on endometriotic implants, macrophage products are implicated in the pathophysiology of endometriosis-associated pain and infertility.

Natural Killer (NK) Cells

NK cells are an important component of the innate immune system. They participate in host defenses against infections[30] and have antitumor effects.[31] NK cells recognize their targets in one of two ways. Like many other immune cells, they have receptors that bind immunoglobulin G (IgG) and they kill IgG-coated target cells by a process called *antibody-dependent cellular cytotoxicity*. The second recognition system is characteristic of NK cells and involves *killer-activating receptors* and *killer-inhibitory receptors* (KIRs).[32] If the killer-activating receptors are occupied, NK cells show cytotoxic activity. Conversely, when stimulated, KIRs send inhibitory signals that override the *kill* signal and suppress cytotoxic activity.

It has been suggested that a decrease in NK cell activity may lead to impaired clearance of regurgitated endometrial cells from the peritoneal cavity and facilitate development of endometriosis. Initial studies investigating NK cell numbers in peritoneal fluid of women with endometriosis reported conflicting results. Whereas some found a decrease in peritoneal NK cells,[33] others reported no change[34] or an increase.[17] On the other hand, studies investigating NK cell activity in women with endometriosis consistently showed a decrease in cytotoxic activity. NK cells from both the peritoneal fluid and the peripheral blood of women with endometriosis were found to have decreased cytotoxic activity against autologous and heterologous endometrium.[34,35] The decrease in NK cell cytotoxicity in the peritoneal fluid was more pronounced in the moderate and severe stages of endometriosis.[36] These findings suggest that the alterations in NK cell activity in women with endometriosis are due to qualitative rather than quantitative changes.

Multiple mechanisms seem to be involved in the suppression of NK cell activity in women with endometriosis. Both sera[37] and peritoneal fluid[38,39] from women with endometriosis suppress NK cell cytotoxicity,[37,38] suggesting that soluble factors are also involved. Wu et al[40] found that peritoneal NK cells of women with endometriosis have higher KIR expression. More recently, Maeda et al[41] identified KIR2DL1 as the subtype of KIR overexpressed in peripheral and peritoneal NK cells of women with endometriosis. Because expression of KIR appears to be related to NK cell activity, increased KIR expression in NK cells in women with endometriosis may contribute to decrease peritoneal NK cell activity.

Gonadotropin-releasing hormone (GnRH) agonist treatment causes a progressive increase in both NK cell activity[42] and number[43] in women with endometriosis. This might be a direct effect of the GnRH agonist or a consequence of estradiol levels decreased by the GnRH agonist. Conversely, danazol suppresses spontaneous and activated NK cell cytotoxicity in vitro at doses slightly higher than would be achieved in women taking 600 mg daily.[44]

Lymphocytes

Lymphocytes constitute the main cell type that mediates the acquired immune response. B and T lymphocytes account for humoral and cellular acquired immune responses, respectively, but there is significant cross talk between the two systems of cells. B cells secrete immunoglobulins, the antigen-specific antibodies responsible for eliminating extracellular microorganisms. T cells help B cells to make antibodies. T cells can also eradicate intracellular pathogens by activating macrophages and by killing virally infected cells. Both B and T cells develop from pluripotent stem cells in the fetal liver and in bone marrow. B cells reach maturity in the bone marrow, but T cells must travel to the thymus to complete their development. Mature T and B lymphocytes are then transported to the lymph nodes, spleen, and mucosa-associated lymphoid tissue, where the adaptive immune responses are generated.

In the thymus, T cells are differentiated into two major subclasses distinguished by the presence of the respective glycoproteins CD8 and CD4. The CD4 phenotype is the mature helper T cell, and the $CD8^+$ phenotype constitutes the suppressor/cytotoxic T cells. CD8 and CD4 serve as coreceptor molecules for major histocompatibility complex (MHC) class I and class II molecules, respectively. The activation of T cells depends not only on the mere presence of their cognate antigen but also on their binding, their mean T cells, to MHC proteins on antigen-presenting cells.

More than 20 years ago, Dmowski et al[45] showed that T cell–mediated immunity to autologous endometrium is suppressed in rhesus monkeys with spontaneous endometriosis. Similarly, cytotoxic activity of peripheral blood lymphocytes against autologous endometrial cells is decreased in women with endometriosis.[46] These observations led to the speculation that endometriosis develops as a result of an impaired cell-mediated immune response that is believed to be critical in clearing ectopic endometrial cells from the peritoneal cavity.[47]

Quantitative analysis of T cells and their subgroups in the peripheral blood and peritoneal fluid of women with endometriosis followed these studies. Total lymphocyte numbers and helper/suppressor ratio in the peripheral blood are not markedly affected in women with endometriosis,[17,48] although one study reported an increase in helper/suppressor ratio.[49] Similarly, there is no change in total lymphocyte content or helper/suppres-

sor ratios in the eutopic endometrium of women with endometriosis compared with eutopic endometrium from normal controls.[50]

On the other hand, an increase in peritoneal fluid T-lymphocyte numbers is observed in women with endometriosis.[17,47] Both helper and suppressor subtypes seem to be increased in numbers.[17,47] The number of T lymphocytes scattered in the stroma of ectopic endometrium as detected by immunohistochemistry is also elevated compared with proliferative and secretory eutopic endometrium from women without endometriosis.[51] An increase in both helper and suppressor subtypes contributes to the observed increase in T-cell content of implants while their relative ratio seems to be unchanged.[51]

The effect of danazol on lymphocyte activation has been measured using peripheral blood mononuclear cell cultures obtained from healthy men and women. Concentrations of danazol comparable to those attained in women taking 600 mg/day significantly inhibit macrophage-dependent T-lymphocyte proliferation in the phytohemagglutinin, concanavalin A, and mixed lymphocyte culture systems.[52] Danazol at the same concentration does not significantly affect macrophage-dependent T-lymphocyte activation of B lymphocytes as assessed in the pokeweed mitogen assay.[52] Dexamethasone and progesterone had similar effects. Danazol treatment in vitro and in vivo also suppresses peripheral blood monocytes (PBM)-mediated enhancement of endometrial cell proliferation.[53] The effects are against both PBM and endometrial cells, suggesting that danazol affects monocyte-derived growth-stimulating factors and endometrial cell response to growth-stimulating factors.[53]

Similar to the effects of danazol, GnRH agonist treatment for 2 to 4 months causes a significant increase in T-lymphocyte mitogenic activity of peripheral blood lymphocytes but does not have a significant effect on T-cell subsets or B-cell numbers.[43]

HUMORAL IMMUNE RESPONSE

Autoantibodies

In addition to alterations in cell-mediated immunity, considerable evidence indicates that endometriosis is associated with polyclonal B-cell activation and an increased incidence of autoantibodies.

An increase in B-cell activity in women with endometriosis was recognized two decades ago. Weed and Arquembourg[54] demonstrated IgG and complement deposits in the eutopic endometrium of women with endometriosis associated with a reduction in the serum total complement level. Mathur et al[55] were the first to report an abnormal incidence of autoantibodies in women with endometriosis, and they presented the first evidence that patients with endometriosis and patients with established autoimmune diseases may exhibit similar autoantibody profiles. They detected IgG and IgA autoantibodies in sera and in cervical and vaginal secretions of women with endometriosis to ovarian and endometrial cells.[55] Gleicher et al[56] reported the presence of IgG, IgM, and IgA autoantibodies directed against cell-derived antigens such as phospholipids, histones, and polynucleotides in women with endometriosis and argued that ectopic endometrium might induce an autoimmune response and contribute to infertility associated with endometriosis. Antibodies to serum transferrin and α_2-Heremans Schmidt glycoprotein have also been described and proposed as diagnostic markers.[57] However, they did not have adequate specificity. Recent evidence suggests that a common carbohydrate epitope, the Thomsen-Friedenreich–like (T) antigen, found on many of these antigens may be involved in the autoantibody response.[58] The anti-T–like response may be indicative of an underlying genetic defect.[59]

The association between autoantibody abnormalities and endometriosis could explain endometriosis-related infertility and recurrent pregnancy loss. An increased risk of pregnancy loss has been associated with the presence of abnormal non–organ-specific[60] as well as organ-specific autoantibodies.[61]

Treatment with danazol[62] or GnRH analogs[63] suppresses the levels of autoantibodies associated with endometriosis. Danazol treatment (400 mg ID) also results in down-regulation of T cells, macrophages, and expressed human leukocyte antigens in eutopic endometrium of women with endometriosis.[64] In women with endometriosis undergoing in vitro fertilization, the presence of autoantibodies is associated with significantly lower pregnancy rates. Treatment of women with endometriosis who have autoantibodies with corticosteroids results in a significant increase in pregnancy rates.[65] These data can be interpreted to suggest that autoantibodies may play a role in the infertility associated with endometriosis. Some authors take this further and argue that endometriosis acts like an autoimmune disease.

Like classical autoimmune diseases, endometriosis is characterized by polyclonal B-cell activation, immunological abnormalities in T- and B-cell functions, increased apoptosis, tissue damage, and multiorgan involvement. Moreover, there is familial occurrence, possible genetic basis, female preponderance, and increased likelihood of other autoimmune diseases.[66–68] However, according to Rose and Bona,[69] in order to classify a disorder as "autoimmune," there should be "direct" or "indirect" evidence of autoimmune etiology. Direct proof of autoimmune etiology is "disease reproducibility through direct transfer of autoantibody or autoantigen-specific T cells to animals or into in vitro systems." Indirect evidence of autoimmune etiology is defined as "disease reproducibility

in animals through experimental immunization with the implicated autoantigen or through cell transfer from animals with genetically determined models of disease." Furthermore, isolation of autoantibodies or identification of self-reactive T cells from the major target tissues of those affected with the disease was also considered indirect evidence of autoimmune etiology. These properties have not been investigated properly in women with endometriosis.[66]

Currently, although it seems that autoantibodies may be involved in certain cases of endometriosis-associated infertility, the relative importance of autoimmunity in the pathogenesis and pathophysiology of this disease is still controversial.

Cytokines and Growth Factors

Cytokines are a large family of low-molecular-weight soluble proteins involved in regulating cellular activity. They act as paracrine and autocrine messengers both within the immune system and between the immune system and other systems of the body. Their action is mediated by specific cytokine receptors.

Cytokines and growth factors play an important role in regulating chemotaxis, mitosis, angiogenesis, and differentiation. Whereas an impaired cellular immune response has been implicated as a permissive factor in survival of endometrial cells in the peritoneal cavity, cytokines and growth factors seem to promote implantation and growth of ectopic endometrium by inducing proliferation and angiogenesis. Moreover, certain cytokines are also implicated in the attachment of endometrial cells to the peritoneal surface and invasion of these cells into the mesothelium. In this section we review some of the cytokines and growth factors that are elevated in women with endometriosis and are likely to be involved in the pathogenesis of the disease.

Interleukin-1

Interleukin-1 (IL-1) is a cytokine that plays an important role in inflammation and immune response. It is secreted mainly by activated monocytes and macrophages as well as T and B cells and NK cells. It affects the activation of T cells and the differentiation of B cells. There are two distinct molecular forms of IL-1 (IL-1α and IL-1β) derived from two different genes. An IL-1 receptor antagonist (IL-1ra) has also been defined and acts as an endogenous receptor inhibitor.

IL-1 has been isolated from the peritoneal fluid of women with endometriosis. Most researchers found increased levels in such women,[70–72] although others found no difference.[73] Endometrial stromal cells isolated from endometriotic lesions show a two- to threefold increase in IL-1 receptor expression compared with those isolated from normal endometrium. Hence, up-regulation of IL-1 receptor expression may be another mechanism by which endometriotic tissue becomes more responsive to IL-1.

IL-1 promotes the development of endometriosis by up-regulating the expression of other cytokines and growth factors. For example, IL-1β induces the expression of angiogenic factors vascular endothelial growth factor and interleukin-6 in endometriotic stromal but not in normal endometrial stromal cells.[74] Hence, IL-1β may promote angiogenesis in endometriotic lesions. IL-1β also increases soluble intercellular adhesion molecule-1 (sICAM-1) shedding from endometrial cells and may interfere with peritoneal immune surveillance.[75]

There are conflicting observations concerning the in vitro effects of IL-1 on early reproductive events.[71,76,77] IL-1 inhibits mouse embryo development in vitro but only at very high concentrations. IL-1 also impairs the oocyte-penetrating capacity of the sperm in both the hamster and the human.[78] However, IL-1 does not appear to affect sperm motion parameters significantly.[76]

Treatment of women with endometriosis with danazol results in a decrease in soluble serum IL-1β levels.[79] Danazol suppresses the production of IL-1β by human monocytes in vitro.[80] Treatment of women with endometriosis with danazol or the GnRH agonist buserelin results in down-regulation of elevated IL-1 in the peritoneal fluid.[81] Moreover, the embryotoxicity of peritoneal fluid from women with endometriosis becomes undetectable after medical treatment.[81]

Interleukin-8

Interleukin-8 (IL-8) is a chemokine that induces chemotaxis of neutrophils and is a potent angiogenic factor. Besides mesothelial cells that form the majority of the peritoneal cells, macrophages, endometrial cells, endothelial cells, and fibroblasts are potential sources of this chemokine.[82,83]

Normal eutopic endometrium expresses IL-8. Endometrial IL-8 expression is highest in the late secretory and early proliferative phase, around the time of retrograde menstruation.[84] IL-8 is elevated in the peritoneal fluid of women with endometriosis, and peritoneal fluid IL-8 levels correlate with the severity of the disease.[83,85] IL-8 is expressed in ectopic endometrium in both stromal and epithelial compartments independent of menstrual cycle phase.[86] Moreover, IL-8 is expressed in cultured mesothelial cells and its expression is up-regulated by IL-1β and tumor necrosis factor-α (TNF-α), suggesting that under inflammatory conditions mesothelium may be an important source of IL-8.[83]

IL-8 seems to have multiple effects that would promote the implantation and growth of ectopic endometrium in the peritoneal cavity. IL-8 stimulates the

adhesion of endometrial stromal cells to fibronectin in a concentration-dependent manner.[87] Thus, IL-8 may facilitate the initial attachment of endometrial cells to the peritoneal surface. Interestingly, in vitro attachment of endometrial stromal cells to extracellular matrix up-regulates IL-8 gene expression.[88]

After the initial attachment to the mesothelium, endometriotic cells invade the extracellular matrix of the peritoneum.[89] Secretion of metalloproteinases plays a key role in endometrial cell invasion of the extracellular matrix. In vitro metalloproteinases activity is up-regulated when endometrial cells are exposed to IL-8, especially when those cells are also grown in the presence of extracellular matrix proteins.[90] Finally, IL-8 also increases endometrial stromal cell proliferation in vitro in a concentration-dependent manner and anti–IL-8 antibody inhibits endometrial stromal proliferation.[91]

Monocyte Chemotactic Protein-1 (MCP-1)

Endometriosis is associated with elevated numbers of activated macrophages in the peritoneal cavity. The products of these macrophages are thought to affect the growth of endometriotic implants. A possible mediator of macrophage recruitment into peritoneal cavity in women with endometriosis is MCP-1.

MCP-1 is a monocyte chemoattractant and activating cytokine. It is produced by monocytes, T lymphocytes, fibroblasts, and vascular smooth muscle and endothelial cells as well as endometrial cells. Its production is up-regulated by other cytokines such as interferon-γ, IL-1, and TNF-α.

The concentration of MCP-1 is elevated in the peritoneal fluid of women with endometriosis compared with women without endometriosis, and its level also correlates with the severity of the disease and tends to decrease with medical treatment.[81,92] Elevated MCP-1 levels in the peritoneal environment of women with endometriosis seem to be due to an increase in MCP-1 expression in eutopic and ectopic endometrial cells. MCP-1 is expressed in eutopic endometrium from normal women and women with endometriosis[93] as well as in endometriotic implants.[94] MCP-1 expression is increased in eutopic endometrium of women with endometriosis[93] and in ectopic endometrium.[94]

IL-1β stimulation up-regulates MCP-1 expression in eutopic endometrial epithelial cells from women with endometriosis[95] as well as ectopic endometrial cells in culture.[96] Estrogen pretreatment markedly increases IL-1β–induced MCP-1 expression in both cell types.[94,97] On the other hand, estrogen pretreatment does not cause a significant increase in MCP-1 expression in epithelial cell cultures from normal women or in stromal cell cultures.[97]

In cultured eutopic endometrial epithelial cells from women with endometriosis, treatment with buserelin acetate (a GnRH agonist) has no significant effect on MCP-1 expression, whereas danazol (a testosterone analog) and dexamethasone (an anti-inflammatory glucocorticoid hormone) show a direct dose-dependent inhibitory effect on MCP-1 expression.[98] These medications have a similar effect on MCP-1 expression by cultured endometriotic cells.[99]

RANTES

RANTES (regulated on activation, normal T-cell expressed and secreted) is another cytokine chemoattractant for monocytes as well as memory T cells and eosinophils. It is secreted by T cells, some epithelial cells, and mesenchymal cells.

The concentration of RANTES is increased in the peritoneal fluid of women with endometriosis and its level correlates with the severity of the disease.[100] Ectopic endometrial implants are the most likely source of elevated peritoneal fluid RANTES in women with endometriosis.

In normal endometrium, RANTES is expressed in the stromal component.[101] Cultured endometrial stromal cells synthesize RANTES messenger RNA (mRNA) and secrete protein when stimulated by inflammatory proteins, whereas epithelial cells synthesize neither protein nor mRNA.[101] The discrepancy between basal RANTES expression in vivo and in vitro, with a requirement for exogenous cytokine stimulation for RANTES expression in cultured endometrial stromal cells, suggests that RANTES secretion is induced by other cytokines in the peritoneal cavity.

In endometriotic implants, the pattern of RANTES protein distribution is similar to that found in the normal endometrium.[101] However, endometriotic cell cultures produce significantly higher amounts of RANTES when stimulated with cytokines compared with cultures derived from normal endometrium. Increased RANTES production by ectopic endometrial implants in response to chemokines may explain its elevated levels in peritoneal fluid and contributes to the increased recruitment of macrophages into the peritoneal cavity in women with endometriosis.

Tumor Necrosis Factor-α

TNF-α is a cytokine that plays a key role in a multitude of inflammatory processes. Initially identified for its ability to kill certain cell lines, TNF-α was later found to be able to initiate an inflammatory response by activating a cascade of cytokines. It is produced by neutrophils, activated lymphocytes, macrophages, NK cells, and several other nonhematopoietic cells.

TNF-α has been implicated in the pathogenesis and pathophysiology of endometriosis. TNF-α is expressed in eutopic human endometrium, predominantly

in epithelial cells and during the secretory phase of the menstrual cycle.[102] Like some of the other cytokines previously discussed, TNF-α is expressed in cultured endometrial epithelial cells and its expression is up-regulated by IL-1.[103] TNF-α concentrations are increased in the peritoneal fluid of patients with endometriosis, and its level correlates with the stage of the disease.[104] In addition to ectopic endometrial cells, peritoneal macrophages have been suggested as a possible source of elevated peritoneal fluid TNF-α in women with endometriosis.[73]

TNF-α causes an increase in the adherence of cultured stromal cells to mesothelial cells,[105] suggesting that it may facilitate adherence of ectopic endometrial tissue to the peritoneum and allowing implants to develop. TNF-α may also be involved in endometriosis-associated infertility. It affects sperm motility in vitro, but only at very high concentrations.[77] Moreover, TNF-α shows embryotoxic effects.[76]

As with its effects on IL-1 expression, danazol treatment results in suppression of TNF production by human monocytes in vitro.[80] Treatment of women with endometriosis with danazol or the GnRH agonist buserelin results in down-regulation of elevated TNF in the peritoneal fluid.[81] Moreover, the embryotoxicity of peritoneal fluid from women with endometriosis becomes undetectable after medical treatment.[81]

Vascular Endothelial Growth Factor

A key condition for the survival and growth of ectopic endometrial tissue following successful adhesion is the establishment of a new blood supply. Active endometriotic implants are markedly vascularized. An important mediator of local angiogenesis is vascular endothelial growth factor (VEGF). VEGF is a heparin-binding glycoprotein and a potent angiogenic produced by monocytes, macrophages, and smooth muscle cells. It is a very potent mitogen for endothelial cells, induces vascular permeability, and acts as a chemoattractant for monocytes.

VEGF protein is localized predominantly in endometrial glands. Stromal staining is less abundant and more diffuse.[106,107] Estradiol up-regulates VEGF expression in cultured endometrial stromal cells[106] and peritoneal macrophages.[108] Hypoxia, IL-1, platelet-derived growth factor and transforming growth factor-β, epidermal growth factor, and prostaglandin E_2 are other factors that up-regulate VEGF expression.[74,109,110]

VEGF is elevated in the peritoneal fluid of women with endometriosis, and peritoneal fluid VEGF concentrations are significantly higher in women with moderate to severe endometriosis than in women with minimal to mild endometriosis.[108] VEGF is expressed in endometriotic lesions. Its expression is more pronounced around red endometriotic lesions as compared with the more inactive black powder-burn implants.[108,111] As with TNF-α, the cellular sources of VEGF in peritoneal fluid include activated peritoneal macrophages[108] in addition to endometriotic lesions.[106]

CONCLUSION

Endometriosis is a common gynecologic disorder characterized by the presence of endometrial tissue outside the uterine cavity. Although no single theory can explain all cases of endometriosis, the most commonly accepted theory is Sampson's theory of retrograde menstruation. Retrograde menstruation occurs in 76 to 90% of women. The much lower prevalence of endometriosis suggests that additional factors determine susceptibility to endometriosis.

Endometriosis is associated with changes in both cell-mediated and humoral immunity. Impaired NK cell activity resulting in inadequate removal of refluxed menstrual debris may play a role in the development of endometriotic implants. Moreover, although the peritoneal fluid of women with endometriosis contains increased numbers of immune cells, these seem to facilitate rather than inhibit the development of endometriosis. Macrophages that would be expected to clear endometrial cells from the peritoneal cavity appear to enhance their proliferation by secreting growth factors and cytokines. Several cytokines and growth factors display increased levels in the peritoneal environment of women with endometriosis. These immune mediators seem to promote implantation and growth of ectopic endometrium by inducing proliferation and angiogenesis. Although it is unclear whether these immunologic alterations induce endometriosis or are a consequence of its presence, they appear to play an important role in allowing endometriosis implants to persist and progress and contribute to the development of associated infertility and pelvic pain.

Danazol and GnRH agonists are commonly used for the medical treatment of endometriosis. These medications seem to down-regulate the cellular and humoral immune response concomitant with their effect on endometriotic implants. Immunomodulatory effects of danazol and GnRH agonists are likely to contribute to the observed clinical improvement associated with their use.

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