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Treating endometriosis as an autoimmune disease

Warren B. Nothnick, Ph.D.

Departments of Obstetrics and Gynecology and Molecular and Integrative Physiology, University of Kansas Medical Center, Kansas City, Kansas

Objective: To review the literature on the role of autoimmunity in the etiology of endometriosis, compare the similarities in the pathophysiologies between endometriosis and autoimmune diseases, and discuss the use of immunomodulators currently used to treat autoimmune diseases as potential therapies for endometriosis.

Design: The literature on endometriosis and other autoimmune diseases was reviewed, and summary data are presented.

Results: Endometriosis shares many similarities with autoimmune diseases such as rheumatoid arthritis, Crohn's disease, and psoriasis. These similarities include elevated levels of cytokines, decreased cell apoptosis, and T- and B-cell abnormalities. Because the use of immunomodulators and inflammatory modulators has proven to be an effective means of medical management for these autoimmune diseases, similar therapies may prove useful in treating endometriosis.

Conclusion(s): Although substantial evidence indicates that endometriosis at least shares many similarities with autoimmune diseases, endometriosis is primarily treated by using compounds that induce a hypoestrogenic environment. A review of the literature combined with the shortcomings of current means of medical management for endometriosis support the postulate that treatment of endometriosis with immunomodulators and inflammatory modulators is warranted. (Fertil Steril® 2001;76:223–31. ©2001 by American Society for Reproductive Medicine.)

Key Words: Endometriosis, autoimmune disease, inflammation, cytokines, immunomodulators, medical therapy

THE PATHOPHYSIOLOGY OF ENDOMETRIOSIS

Immunological Factors

Endometriosis is a disease that occurs in menstruating females and is characterized by such symptoms as pelvic pain, dysmenorrhea, and infertility. Although nearly three quarters of a century have passed since the initial description of endometriosis (1), our current understanding of the etiology and pathophysiology of the disease still remains unclear. Since its first description in 1927, several different hypotheses have been put forth that attempt to explain the mechanisms for the development of this disease. Of these, it is Sampson's (1) theory of retrograde menstruation that has gained the most supportive evidence. The presence in the peritoneal fluid of viable endometrial tissue (2–4) that is capable of growth (5–7) and the anatomical distribution of endometriotic implants (8) support this theory.

However, this theory has one major shortcoming: the fact that nearly all women of reproductive age exhibit some degree of retrograde menstruation (9, 10). As such, one would postulate that there must be some other factor or factors that allow for this retrogradely displaced endometrial tissue to develop into ectopic lesions in only a portion of these women. Alternatively, it has been suggested that all women actually develop "endometriosis" to some extent, even be it microscopic (11). Some women exhibit symptoms associated with the disease, such as pelvic pain, which eventually, through diagnostic laparoscopy, lead to the diagnosis of the disease. In contrast, other women who display no symptoms may go undiagnosed until endometriosis is detected only due to pelvic surgery for other medical reasons.

Regardless, if all women have endometriosis to some degree (microscopic, minimal, severe) or if only a portion of all women who have reverse menstruation actually develop the disease, the question still remains as to what

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Reprint requests: Warren B. Nothnick, Ph.D., University of Kansas Medical Center, Department of Obstetrics and Gynecology, 3901 Rainbow Boulevard, Kansas City, Kansas 66160 (FAX: 913-588-6271; E-mail: wnothnic@kumc.edu).

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allows for the development (in the latter case) or progression (in the former case) of the disease. Numerous investigators have suggested that there is an association between the presence of endometriosis and an altered immune system. The theory of an altered immune system and endometriosis suggests that changes in cell-mediated immunity and/or humoral immunity may contribute to the development of the disease.

Cell-mediated immunity and endometriosis

Changes in cell-mediated immunity have been reported in women with endometriosis, but data are inconsistent. In women with endometriosis, a reduction in proliferation of peripheral blood lymphocytes in response to recognition of endometrial antigen and cells has been reported (12, 13). Similarly, a decreased destruction of endometrial cells has been suggested because the cytotoxic effect of peripheral blood lymphocytes against autologous endometrial cells is decreased in women with endometriosis (13). Badaway and colleagues (14) reported that the ratio of T-helper to T-suppressor cells is increased in the peripheral blood of women with endometriosis, but conflicting studies (13, 15) have also indicated no marked differences in peripheral lymphocyte profiles. In addition, increases in the T-helper to T-suppressor ratio and concentration of both cells respectively have been reported in peritoneal fluid (16, 17) and endometriotic tissue (compared with eutopic endometrium) (18) in women with endometriosis. Yet, in a separate study, no significant differences in the T-helper to T-suppressor ratio could be detected between eutopic endometrium from women with and without endometriosis. In all, there appear to be inconsistencies in the alterations in T-cells and their role in the pathophysiology of endometriosis (19). Many of these inconsistencies may stem from differences among studies with respect to the methodology used for T-cell assessment and patient population variability.

Natural killer (NK) cells have also been shown to be altered in women with endometriosis, and those data as a whole appear to be more consistent. Natural killer cells, as their name implies, are large granular lymphocytes that kill cells bearing undefined target molecules and opsonized cells coated with antibody. Both peripheral and peritoneal NK cells from women with endometriosis display decreased cytotoxicity to autologous and heterologous endometrium compared with those of controls (20, 21), and peritoneal NK cell cytotoxicity has been to be shown to be inversely correlated with the more severe stages of the disease (22). Thus, it is postulated that the decrease in NK cytotoxicity to retrogradely shed endometrial tissue may allow for the establishment of this tissue within the peritoneal cavity.

Although the general consensus is that there is a decrease in NK cytotoxicity, there are discrepancies in the percentage or number of these cells in women with endometriosis. Separate studies have suggested that the number or percentage of NK cells in women with the disease may be decreased (23), increased (16), or unchanged (20). It is interesting to note that the reduced number of NK cells in women with endometriosis can be restored after GnRH agonist (GnRH-a) treatment (23), which may be due to a direct effect of GnRH-a or which may be indirect, via the lowering of serum estradiol levels. The direct effect may be manifested at the level of control of soluble peritoneal factors. Both peritoneal fluid and sera from women with endometriosis have been shown to reduce NK cell activity (24, 25). The constituents of the fluids responsible for this suppression have been hypothesized to be products of monocytes or macrophages. Monocyte and macrophage products are known modulators of both immune and nonimmune cells, and these cells and their products have been an intense area of investigation in the pathophysiology of endometriosis.

For example, both peripheral monocytes (26) and peritoneal macrophages (27–30) are altered in women with endometriosis. Zeller and colleagues (26) noted that although the percentage of peripheral monocytes in women with endometriosis does not differ from that in women without the disease, the activational status of these monocytes is increased in women with the disease. Peritoneal macrophages, on the other hand, have been shown to be increased in total number, concentration, and activational status (27–31).

Associated with the increase in the activational status of the macrophage is an increase in the release of their products, such as growth factors and cytokines. Indeed, the role of peritoneal fluid cytokines and growth factors in the pathophysiology of endometriosis has been an intense area of investigation over the past 20 years. The postulated role of these macrophage products include stimulation of endometrial cell proliferation (32, 33) and implantation of endometrial cells or tissue (34), increased tissue remodeling through regulation of matrix metalloproteinases (35), and increased angiogenesis of the ectopic endometrial tissue (36). As such, activated macrophages, through the liberation of cytokines and growth factors, could potentially contribute to the early establishment as well as the progression of the disease at several foci.

Humoral-mediated immunity and endometriosis

In addition to alterations in cell-mediated immunity, considerable evidence has been gathered that indicates that there are alterations in B-cell activity and an increased incidence of autoantibodies in women with endometriosis. In 1980, two independent reports suggested that an abnormal antigen-antibody reaction might occur in women with endometriosis: Startseva (37) detected increased B-cell function in women with endometriosis (as well as with adenomyosis), whereas Weed and Arquembourg (38) found that women with endometriosis had decreased C3 and G deposition in the endometrium and a corresponding reduction in the serum total complement levels. Immunoglobin G and IgA autoantibod-

TABLE 1

Common characteristics between autoimmune diseases and endometriosis.

Tissue damage
Polyclonal B lymphocyte activation
T-lymphocyte immunological abnormalities
B-lymphocyte immunological abnormalities
Associated autoimmune diseases
Preponderance of females
Multiorgan involvement
Familial occurrence
Possible environmental factors
Possible genetic basis
Altered apoptosis

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ies against endometrial and ovarian tissue in sera, vaginal, and cervical secretions in women with endometriosis have also been detected (39). In addition, autoantibodies of IgG, IgM, and IgA isotypes directed against cell-derived antigens such as phospholipids and histones has been reported (40). More recent studies (41, 42), however, question the presence of these autoantibodies and have even found varying degrees of correlation between the severity of endometriosis and autoantibodies, ranging from positive (43) to negative (44) to no relationship at all (45–47).

The Role of Autoimmunity

If there is an increase in the frequency of autoantibodies, one might suspect that endometriosis may be an autoimmune disease. This theory was first introduced by Gleicher and colleagues (40), and strong evidence would indicate that endometriosis fulfills most of the classification criteria of an autoimmune disease (Table 1). Like classical autoimmune diseases, endometriosis has been associated with polyclonal B-cell activation, immunological abnormalities in T- and B-cell functions, increased apoptosis, tissue damage, multiorgan involvement, familial occurrence, possible genetic basis, involvement of environmental cofactors, female preponderance, and association with other autoimmune diseases. Endometriosis is associated with recurrent pregnancy loss and infertility (48), which might be explained by the presence of autoantibody abnormalities. Further evidence for a role of autoimmunity or autoantibodies in the infertility associated with endometriosis comes from studies that have shown that treatment with Danazol (49) or GnRH analogues (50) suppresses the levels of autoantibodies associated with endometriosis. These data can be interpreted to suggest that abnormal autoantibodies may play a role in the infertility associated with endometriosis.

However, for a disease to be truly autoimmune in nature, it should be manifested in normal animals after adaptive transfer of immunoglobulin from the blood or affected tissues of subjects with the autoimmune disease. To date, no such studies have been performed. Although it remains uncertain whether endometriosis is an autoimmune disease, this disease does share characteristics with common autoimmune diseases such as rheumatoid arthritis (RA), Crohn's disease, and psoriasis. Some of the recurring pathophysiologies with endometriosis and these autoimmune diseases are increased inflammation, elevated levels of tissue-remodeling components, altered apoptosis, and increased local and/or systemic levels of cytokines.

An increase in peritoneal inflammation, as evidenced by elevated peritoneal fluid cytokine levels, is well established in women with endometriosis (reviewed in Oral et al. [(51)]). Although it is uncertain whether the elevated cytokines levels or inflammation are causes for or results of the disease, it is clear that these cytokines may have profound effects that can lead to the establishment and further progression of the disease. For example, cytokines can stimulate endometrial cell adhesion to peritoneal mesothelial cell monolayers in vitro (34), as well as to specific extracellular matrix proteins (52).

The role of adhesion molecules in the establishment of the early developmental stages of endometriosis is just becoming apparent (53, 54) and appears to play a very important role in the initial stages of endometriosis development. Once the endometrial cells and tissue adhere to the peritoneum or other pelvic surfaces with which they come into contact, the next proposed step in the development/progression of endometriosis involves the invasion of the underlying tissue-cell layer. Both in vitro (55) and in vivo (56, 57) studies indicate that endometriosis is an invasive disease.

One of the major groups of proteases postulated to play an important role in these invasive processes and hence the etiology and pathophysiology of endometriosis is the matrix metalloproteinases (MMPs). Matrix metalloproteinases are produced by endometriotic tissue (58, 59), and eutopic endometrial tissue from women with endometriosis expresses elevated MMP activity compared with endometrium from women free of the disease (Nothnick WB, unpublished observations). Further evidence supporting the role of MMPs in the establishment of endometriosis comes from the work of Bruner and colleagues (60), who injected human endometrial tissue into nude mice. In that study, it was demonstrated that MMP expression by the human endometrial tissue paralleled the ability of this tissue to develop into ectopic lesions. If MMP expression and activity was blocked by either progesterone treatment of the tissue before injection or if the tissue was injected into the mice concurrently with the MMP inhibitor, TIMP-1, the development of endometriosis was blocked.

Another important similarity between endometriosis and autoimmune diseases is the deregulation of the apoptotic process. Abnormalities in apoptosis have been postulated to play a role in autoimmune diseases by allowing either for the persistence of auto-reactive lymphocytes and/or the involvement of so-called death receptors and/or their ligands in tissue destruction (reviewed in O'Reilly and Strasser [(61)]). In short, the inability of cells to transmit a "death" signal or the ability of cells to avoid cell death is associated with

several autoimmune diseases. It is intriguing that with respect to endometriosis, several studies have demonstrated that endometrial cells from eutopic (and ectopic) endometrium from women with endometriosis have altered apoptotic mechanisms (62–64).

It is further postulated that this alteration in apoptosis then allows the retrogradely shed endometrial tissue to "escape" cell death. As such, in women with endometriosis, that portion of the menstrual effluent that is transported retrogradely through the fallopian tubes and into the peritoneal cavity contains a population of cells that are more viable (i.e., less likely to undergo apoptosis) and more apt to develop into ectopic lesions.

The exact mechanisms responsible for this altered apoptosis in endometrial tissue from women with endometriosis are unknown. It has been demonstrated that this altered pattern of cell death can be "reversed" by GnRH analogue therapy, suggesting that this peptide may play a role in the normal turnover of endometrial cells during the course of the menstrual cycle (62).

One cytokine that has gained recent attention in the pathophysiology of autoimmune diseases is tumor necrosis factor-α (TNF-α). Tumor necrosis factor-α is a major product of activated macrophages. It activates inflammatory leukocytes as well as leads to the production of other proinflammatory cytokines, such as interleukin-1 (IL-1), IL-6, and additional TNF-α (65). Because TNF-α is proposed to play an important role in autoimmune diseases, it is not surprising that new treatments for diseases such as RA, Crohn's disease, and psoriasis have become available (reviewed in Infante and Lahita [(66)]). It is also evident that TNF-α may play an important role in the pathophysiology of endometriosis. Numerous studies have demonstrated that TNF-α levels are elevated in the peritoneal fluid of patients with endometriosis [reviewed in Oral et al. (51)].

Tumor necrosis factor-α can stimulate endometrial cell adhesion as well as induce MMP expression (67–69), both of which are necessary events in the initial development of the disease. As such, would the targeting of TNF-α be a logical starting point in the development of novel treatments for endometriosis? The following paragraphs discuss the current treatments for endometriosis and how the application of autoimmune disease treatment modalities, specifically anti-TNF-α therapies, might be beneficial for the treatment of endometriosis.

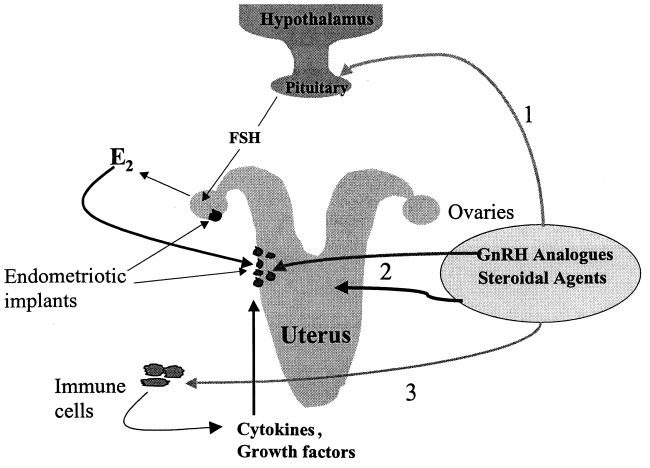
TREATING ENDOMETRIOSIS AS AN AUTOIMMUNE DISEASE

Current Treatments for Endometriosis

Current treatment modalities of endometriosis may be classified as either medical or surgical and, in some cases, both medical and surgical treatment may be combined. Surgical treatment is usually used in cases of severe disease,

FIGURE 1

Potential sites of inhibitory action of medical therapies on endometriotic lesion growth. Current medical therapies for endometriosis include steroidal analogues (such as Danazol) and GnRH analogues, both of which suppress pituitary gonadotropin release. These agents induce a hypoestrogenic state by suppressing gonadotropin (FSH) release, which in turn leads to suppressed production of the female steroid estradiol. Because estradiol stimulates endometriotic implant growth, reduced estradiol production leads to a suppression of endometriotic tissue proliferation (depicted in *pathway 1*). More recent evidence suggests that these agents may also effect endometriotic lesion growth by directly suppressing proliferation of both eutopic and ectopic endometrial tissue (*pathway 2*) as well as by suppressing cytokine/growth factor release from cells of the immune system (*pathway 3*) that are also postulated to stimulate endometriotic implant growth. As such, the mechanisms by which current therapies suppress endometriotic implant growth may operate at the level of the hypothalamic-pituitary axis, the eutopic and ectopic endometrial tissue, the cells of the immune system, or a combination of these three pathways.



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large endometriomas (>2–3 cm), anatomic distortion of the pelvic structures, or adhesive disease. Methods of surgery involve excision, laser ablation, or fulguration with low-voltage electrocautery. However, as endometriosis may be a microscopic disease, not all endometriotic implants may be visible and therefore may not be removed by surgery. Thus, medical treatment that can induce a generalized suppression of the disease may be necessary.

Current medical modalities for endometriosis include estrogen-progesterone combinations, progestogens, anti-progestigens, danazol, and GnRH-a (reviewed in Moghissi [(70)]). Although primarily considered to act at the level of the hypothalamic-pituitary axis, the mechanisms by which these agents prevent endometriotic implant growth action may actually be at multiple levels (Fig. 1). Both steroidal and GnRH-a therapies induce a decrease in gonadotropin release

and subsequent suppression of ovarian steroidogenesis. The suppression of ovarian steroids and the induction of a hypoestrogenic state is then thought to be the primary mechanism by which these modalities suppress endometriotic implant growth. However, progesterone is a potent immunosuppressive agent capable of blocking both cytokine release and action (35, 71, 72).

Similarly, both danazol (73–75) and GnRH analogues (76, 77) have been postulated to perhaps act at the level of immune cells which may in turn lead to the suppression of cytokine levels. Cytokines and growth factors derived from peritoneal immune cells have been implicated as factors that may regulate endometriotic implant growth (78). GnRH peptide and receptors in both eutopic endometrium (79, 80) and endometriotic tissue (81) suggest that GnRH-a may exert antiproliferative affects on endometrial tissue as well. Borroni and colleagues (81) have demonstrated that the GnRH agonist leuprolide acetate can inhibit endometriotic cell proliferation. Thus, it is uncertain whether current therapies elicit their inhibitory effect on endometriotic implant growth at the level of the pituitary-ovarian axis by inhibiting the subsequent production of estradiol, on the cells of the immune system by inhibiting cytokine release, on the endometrial tissue itself, or at all three levels.

Although GnRH-a are one of the most common medical therapies to treat endometriosis, their use is associated with some disadvantages and side effects. Treatment of endometriosis with GnRH agonists is limited to 6 months because of possible adverse effects on bone metabolism (reviewed in Adashi [(82)]), as well as other consequences of the hypoestrogenic state which ensues from their use. This adverse effect on bone density can be overcome by hormone add-back therapy (83), but add-back therapy may reduce the efficacy of GnRH-a (83). In addition, it is very well established that use of GnRH-a is associated with physical side effects such as vasomotor instability, headache, and hot flashes (84, 85), as well as psychiatric side effects, which include depressive mood symptoms (86, 87).

In all, it may seem counterproductive on one hand to use GnRH-a to suppress steroid production to reduce the disease and the disease-associated symptoms while on the other hand adding back the steroids to overcome the detrimental effects of their absence. Additionally, one must consider the variability in therapeutic efficacy of the different brands of GnRH-a (for instance, Leuprolide, Nafarelin, Buserelin, Goserelin), coupled with the different doses, durations, and forms of the added-back steroids. For the most part, current data suggest that GnRH-a therapy coupled with steroid add-back provides effective suppression of endometriosis and endometriosis-associated symptoms (such as pelvic pain) while protecting against detrimental effects of GnRH-a such as bone loss. One can only wonder, though, in this day and age of medicine, why more specific treatments for endometriosis have not been developed. Ideally, such a treatment would induce a regression of the disease and its associated symptoms but would not have the detrimental effects associated with a hypoestrogenic state (such as bone loss). Because it is well established that endometriosis is a disease associated with immune or inflammatory disorders, immunomodulatory drugs may be viable candidates for use in the treatment of endometriosis.

The Use of Anti-TNF Therapies

Although the management of endometriosis today is almost exclusively accomplished through the use of GnRH-a and steroidogenic compounds, the use of immunomodulatory agents to treat endometriosis-associated infertility and suppress endometriotic implant growth has been suggested (88–92). In 1991, Steinleitner and colleagues (88) were the first to demonstrate that treatment of endometriosis-associated infertility could be reversed in hamsters with surgically induced endometriosis. Furthermore, the use of pentoxifylline to treat endometriosis-associated infertility in humans has been reported (89). Although their results were not statistically significant, Balasch and colleagues (89) reported nearly a doubling (31% vs. 18.5%) of pregnancy rates in women with endometriosis treated with pentoxifylline versus placebo. However, these findings were obtained using a small study population that the authors recognized as limiting their study (89). The authors also stressed that larger, multicenter trials may be warranted to adequately address the effect of pentoxifylline on endometriosis-associated infertility in humans (89).

Lastly, pentoxifylline has been successfully used to reduce endometriotic implant growth in an animal model (90). This regression occurred independently of steroid action; circulating estradiol and progesterone levels were unaffected by pentoxifylline administration. This is a potentially exciting finding because these data may be interpreted to suggest that pentoxifylline is capable of inducing regression of endometriotic tissue without inducing a hypoestrogenic state. Lastly, both Evers (91) and Gleicher (92) stressed that because current treatment modalities may not be ideal and may be ineffective to some extent, the design of new experimental treatment approaches toward endometriosis that are based on immunomodulation may be warranted.

In summary, both animal and human studies indicate that pentoxifylline may be beneficial in the treatment of endometriosis-associated infertility or in treatment of the disease itself (i.e., growth of the endometriotic implants). As it appears that pentoxifylline can induce regression of ectopic endometrial tissue without inducing an estrogen deficiency, pentoxifylline may be an exciting alternative to conventional, steroid-suppressing GnRH analogues.

Pentoxifylline

Pentoxifylline (also know as Trental [Aventis Pharmaceuticals; Bridgewater, NJ]) is a methylxanthine that acts as a phosphodiesterase inhibitor. Pentoxifylline has been used for

over 20 years in the treatment of peripheral vasculature disease, cerebrovascular disease, and many other conditions that involve defective regional microcirculation (reviewed in Ward and Clissold [(93)]). By increasing intracellular cyclic adenosine monophosphate levels in platelets, polymorphonuclear leukocytes, and monocytes, pentoxifylline increases red blood cell deformity, reduces blood viscosity, and decreases the potential for platelet aggregation and thrombus formation. The drug is generally well tolerated and shows no significantly greater complaints or adverse effects compared with placebo. Pentoxifylline does not appear to have any significant interactions with other drugs, such as antihypertensive drugs, beta blockers, digitalis, diuretics, antidiabetic agents, and antiarrhythmics.

There have been reports of bleeding and/or prolonged prothrombin time in patients treated with pentoxifylline with and without anticoagulants or platelet aggregation inhibitors, but no causal relationship has been established. Patients who are currently taking warfarin, as well as theophylline-containing drugs, should be closely monitored for prothrombin times and theophylline toxicity, respectively. Pentoxifylline is commonly administered orally in sustained-release form (400-mg capsule) two to three times daily. It is interesting to note that in the studies in which pentoxifylline was used to treat endometriosis or endometriosis-associated infertility, the administered doses were below those commonly prescribed for circulatory disorders, yet substantial effects were noted.

In addition to being used to treat vascular disease, pentoxifylline use has also been suggested for the treatment of RA (94) and irritable bowel disease (95), both of which are diseases associated with elevated levels of cytokines such as TNF-α. Pentoxifylline reduces both the production and action of cytokines such as TNF-α through the elevation of intracellular cyclic AMP levels and subsequent down-regulation of cytokine production and action (96, 97).

Leflunomide

In the last 3 years, additional "anti-TNF-α" compounds have gained Food and Drug Administration approval for the use in treatment of autoimmune diseases such as RA [reviewed in Fox et al. (98)]. Leflunomide (Arava; Aventis Pharmaceuticals) is an isoxazole derivative that was initially derived from anti-inflammatory drug development based on its ability to prevent proliferation of mitogen-stimulated lymphocytes, control autoimmune diseases in rodent models, and prolong tissue graft survival. Leflunomide elicits its antiproliferative effect by inhibiting de novo pyrimidine synthesis and elicits its antiproliferative effects through its major active metabolite, A77-1726. Nearly 100% of the drug is absorbed from the intestinal tract, with rapid nonenzymatic conversion to A77-1726 in both the intestinal mucosal and plasma. The plasma half-life of leflunomide (A77-1726) is 15 days and may be attributed to strong serum protein binding (99.5% is bound to serum albumin). A77-1726 shows no drug interactions with oral contraceptives or oral hypoglycemic medications but may weakly displace certain nonsteroidal anti-inflammatory agents.

Leflunomide is administered as a loading dose of a single 100-mg tablet for 3 days, followed by daily doses of 20 mg. Side effects or adverse reactions are minimal and include diarrhea, alopecia, rash, and elevated liver enzymes, primarily alanine amino transferase and aspartate amino transferase. The elevation of liver enzymes is reversible upon discontinuation of the therapy.

Etanercept

More specific anticytokine therapies have also been developed to treat rheumatoid arthritis. Etanercept (Enbrel; manufactured by Immunex, Seattle, WA and marketed by Immunex and Wyeth-Ayerst Laboratories, St. Davids, PA) is a protein that competes with the endogenous TNF receptor for TNF ligand [reviewed in Jarvis and Faulds (99)]. Composed of the extracellular ligand-binding portion of the TNF receptor linked to the Fc portion of the human IgG1, etanercept binds specifically with TNF, therefore inhibiting the binding of ligand and receptor and the biological effects of this ligand-receptor coupling (such as inflammation and cytokine production). The maximum plasma concentration achieved in patients receiving 25 mg of etanercept is approximately 1.2 mg/L, with the average time to achieve this level being about 3 days.

Etanercept is suspected to be eliminated from the body by the reticuloendothelial system of the liver or spleen, with an estimated terminal half-life of 20 hours in rodent studies. The recommended dose of etanercept is 25 mg, given twice weekly as a subcutaneous injection. The most commonly reported side effects include allergic reactions or injection site reactions, non–upper and upper respiratory tract infections, and headaches, all of which were mild to moderate and not severe enough to discontinue therapy. No interactions between etanercept and other drugs have been reported. Because TNF-α modulates cellular immune responses, the possibility exists that anti-TNF-α therapies may affect host defense against infections and malignancies, and necessary precautions should be taken into account.

Infliximab

Infliximab (Remicade; Centocor, Inc., Malvern, PA), which is currently approved for the treatment of both RA and Crohn's disease, is a chimeric monoclonal antibody that binds to both soluble and membrane forms of TNF-α and neutralizes its biological effects [reviewed in Markham and Lamb (100)]. Additionally, infliximab has also been shown to reduce serum levels of inflammatory mediators, decrease synovial tissue chemokine expression, and decrease lymphocyte migration into the joints of patients with RA. Pharmacokinetic data on infliximab are limited, but data pertaining

to patients with RA who also received methotrexate are available. In these studies, the median serum concentrations in patients receiving the standard dosage regime of infliximab at 3 mg/kg at weeks 0, 2, and 6 and every 8 weeks thereafter was 1.5 mg/L at week 30 (which was 8 weeks after the last dose). It has been reported that serum levels of $\geq$1 mg/dL of infliximab are required to elicit clinical responses.

Adverse events associated with infliximab include upper respiratory tract infections, headache, nausea, sinusitis, rash, and cough. Because infliximab is administered intravenously, it is not surprising that infusion-related reactions have been reported as well. Additionally, the development of antibodies to infliximab as well as the development of antinuclear antibodies and antibodies to double-stranded DNA have been reported. The frequency of the development of anti-infliximab antibodies appears to be inversely related to the dosage of infliximab administered. Concurrent treatment with methotrexate appears to reduce the development of these antibodies and is often prescribed in conjunction with infliximab therapy. The estimated proportion of those patients who receive infliximab who may develop antibodies to the drug is 10%, and the clinical significance of these antibodies is presently unknown.

CONCLUSIONS

Endometriosis is an intriguing disease whose pathophysiology still remains unclear. Great strides have been made in recent years to better understand the potential factors and mechanisms that contribute to the development and progression of the disease. There is overwhelming evidence that immune factors such as cytokines may be involved in the pathophysiology and etiology of the disease. We are now just beginning to understand how these immune factors may participate in the development of the disease at the level of the endometrium, peritoneum, and peritoneal cavity.

Although the debate will continue as to whether endometriosis is truly an autoimmune disease, currently available data strongly suggest that there are at least some very important similarities between endometriosis and such autoimmune diseases as RA, Crohn's disease, and psoriasis. The most central of these similarities is the elevated levels of cytokines such as TNF-α and the pathologic events manifested by their enhanced production. Although the current medical treatment regimens appear to be sufficient, there is definitely room for improvement with respect to the desire to avoid those adverse and unpleasant side effects associated with the hypoestrogenic environment generated by the current GnRH-a therapies. The use of anti-TNF-α therapies for treatment of autoimmune diseases appears to hold promise. As such, it may be warranted for similar therapies to be extended to the treatment of endometriosis.

References

1. Sampson JA. Peritoneal endometriosis due to menstrual dissemination of endometrial tissue into the peritoneal cavity. Am J Obstet Gynecol 1927;14:422–69.
2. Beyth Y, Jaffe H, Jevij ISH, Sadovsky E. Retrograde seeding of endometrium: sequelia of tubal flushing. Fertil Steril 1975;26:1094–7.
3. Bartosik D, Jacob S, Kelly LJ. Endometrial tissue in peritoneal fluid. Fertil Steril 1986;46:796–800.
4. Keettel WC, Stein RJ. The viability of cast-off menstrual endometrium. Am J Obstet Gynecol 1951;61:440–2.
5. Munagyer G, Willemsen WNP, Rolland R, Vermer HM. Cell with mucous membrane of the female genital tract in culture: a comparative study with regard to the histogenesis of endometriosis. In Vitro Cell Dev Biol 1987;23:111–7.
6. Telinde RW, Scott RB. Experimental endometriosis. Am J Obstet Gynecol 1950;60:1147–73.
7. Ridley JH, Edwards IK. Experimental endometriosis in the human. Am J Obstet Gynecol 1958;76:783–90.
8. Jenkins S, Olive DL, Haney AF. Endometriosis: pathogenic implications of the anatomic distribution. Obstet Gynecol 1986;67:335–8.
9. Halme J, Hammond MG, Hulka JF, Raj SG, Talbert L. Retrograde menstruation in healthy women and in patients with endometriosis. Obstet Gynecol 1984;64:151–4.
10. Liu DTY, Hitchcock A. Endometriosis: its association with retrograde menstruation, dysmenorrhea, and tubal pathology. Br J Obstet Gynaecol 1986;93:859–86.
11. Koninckx PR, Oosterlynck D, D'Hooghe T, Meuleman C. Deeply infiltrating endometriosis is a disease whereas mild endometriosis could be considered a nondisease. Ann NY Acad Sci 1994;734:333–41.
12. Dmowski WP, Steele RW, Gaker GF. Deficient cellular immunity in endometriosis. Am J Obstet Gynecol 1981;141:377–83.
13. Steele RW, Dmoswki WP, Marmer DJ. Immunologic aspects of human endometriosis. Am J Reprod Immunol 1984;6:33–6.
14. Badaway SZ, Cuenca V, Stitzel A, Tice D. Immune rosettes of T and B lymphocytes in infertile women with endometriosis. J Reprod Med 1987;32:194–7.
15. Gleicher N, Dmowski WP, Siegel I, Liu TL, Friberg J, Radwansea E, et al. Lymphocyte subsets in endometriosis. Obstet Gynecol 1984;63:463–6.
16. Hill JA, Faris HM, Schiff I, Anderson DJ. Characterization of leukocyte subpopulations in the peritoneal fluid of women with endometriosis. Fertil Steril 1988;50:216–22.
17. Dmowski WP, Gebel HM, Braun DP. The role of cell-mediated immunity in pathogenesis of endometriosis. Acta Obstet Gynecol Scand Suppl 1994;159:7–14.
18. Witz CA, Montoya IA, Dey TD, Schenken RS. Characterization of lymphocyte subpopulations and T cell activation ind endometriosis. Am J Reprod Immunol 1994;32:173–9.
19. Mettler L, Volkov NI, Kulakov VI, Jurgenson A, Parwaresch MR. Lymphocyte subsets in the endometrium of patients with endometriosis throughout the menstrual cycle. Am J Reprod Immunol 1996;36:342–9.
20. Oosterlynck DJ, Cornillie FJ, Waer M, Vandeputte M, Koninckx PR. Women with endometriosis show a defect in natural killer activity resulting in a decreased cytotoxicity to autologous endometrium. Fertil Steril 1991;56:45–51.
21. Wilson TJ, Hertzog PJ, Angus D, Munnery L, Wood EC, Kola I. Decreased natural killer cell activity in endometriosis patients: relationship to disease pathogenesis. Fertil Steril 1994;62:1086–8.
22. Ho HN, Chao KH, Chen HF, Wu MY, Yang YS, Lee TY. Peritoneal natural killer cytotoxicity and CD25+CD3+ lymphocyte subpopulation are decreased in women with stage III-IV endometriosis. Hum Reprod 1995;10:2671–5.
23. Hsu C, Lin Y, Wang S, Huang K. Immunomodulation in women with endometriosis receiving GnRH agonist. Obstet Gynecol 1997;89:993–8.
24. Kanzaki H, Sheng-Wang HS, Kariya M, Mori T. Suppression of natural killer cell activity by sera from patients with endometriosis. Am J Obstet Gynecol 1992;167:257–61.
25. Oosterlynck KD, Meuleman C, Waer M, Koninckx PR, Vandeputte M. Immunosuppressive activity of peritoneal fluid in women with endometriosis. Obstet Gynecol 1993;82:206–12.
26. Zeller JM, Henig I, Radwanska E, Dmowski WP. Enhancement of human monocyte and peritoneal macrophage chemiluminescence activities in women with endometriosis. Am J Reprod Immunol Microbiol 1987;13:78–82.
27. Haney AF, Muscato JJ, Weinberg JB. Peritoneal fluid cell populations in infertility patients. Fertil Steril 1981;35:696–8.
28. Halme J, Becker S, Hammond MG, Raj S. Pelvic macrophages in normal and infertile women: the role of patent tubes. Am J Obstet Gynecol 1982;142:890–5.
29. Halme J, Becker S, Wing R. Accentuated cyclic activation of peritoneal macrophages in patients with endometriosis. Am J Obstet Gynecol 1984;148:85–90.
30. Halme J, Becker S, Haskill S. Altered maturation and function of

peritoneal macrophages: possible role in the pathogenesis of endometriosis. Am J Obstet Gynecol 1987;156:783–9.
31. Dunselman GAJ, Hendrix MGR, Bouckaert PXJ, Evers JLH. Functional aspects of peritoneal macrophages in endometriosis of women. J Reprod Fertil 1988;82:707–10.
32. Hammond MG, Oh ST, Anners J, Surrey ES, Halme J. The effect of growth factors on the proliferation of human endometrial stromal cells in culture. Am J Obstet Gynecol 1993;168:1131–6.
33. Iwabe T, Harada T, Tsudo T, Nagano Y, Yoshida S, Tanikawa M, et al. Tumor necrosis factor-α promotes proliferation of endometriotic stromal cells by inducing interleukin-8 gene and protein expression. J Clin Endocrinol Metab 2000;85:824–9.
34. Zhang RJ, Wild RA, Ojago JM. Effect of TNF-α on adhesion of human endometrial stromal cells to peritoneal mesothelial cells: an in vitro system. Fertil Steril 1993;59:1196–201.
35. Osteen KG, Keller NR, Feltus FA, Melner MH. Paracrine regulation of matrix metalloproteinase expression in the normal endometrium. Gynecol Obstet Invest 1999;48(suppl 1):2–13.
36. Taylor RN, Ryan IP, Moore ES, Hornung D, Shifren JL, Tseng JF. Angiogenesis and macrophage activation in endometriosis. Ann NY Acad Sci 1997;828:194–207.
37. Startseva NV. Clinical immunological aspects of genital endometriosis. Akush Ginekol (Mosk) 1980;3:23–6.
38. Weed JC, Arquembourg PC. Endometriosis: can it produce an autoimmune response resulting in infertility? Clin Obstet Gynecol 1980; 23:885–93.
39. Mathur S, Peress MR, Williamson HO, Youmans CD, Manye SA, Garvin AJ, et al. Autoimmunity to endometrium and ovary in endometriosis. Clin Exp Immunol 1982;50:259–66.
40. Gleicher N, el-Roeiy A, Confino E, Friberg J. Is endometriosis an autoimmune disease? Obstet Gynecol 1987;70:115–22.
41. Meek SC, Hodge DD, Musich JR. Autoimmunity in infertility patients with endometriosis. Am J Obstet Gynecol 1988;158:1365–73.
42. Switchenko AC, Kauffman RS, Becker M. Are there antiendometrial antibodies in sera of women with endometriosis? Fertil Steril 1991; 56:235–41.
43. Taylor PV, Maloney MD, Campbell JM, Skerrow SM, Nip MM, Parmar R, et al. Autoreactivity in women with endometriosis. Br J Obstet Gynecol 1991;98:680–4.
44. Evers JL, Dunselman GA, Van der Linden PJ. Markers for endometriosis. Baillieres Clin Obstet Gynecol 1993;7:715–39.
45. Badaway SZ, Cuenca V, Freliech H, Stefanu C. Endometrial antibodies in serum and peritoneal fluid of infertile patients with and without endometriosis. Fertil Steril 1990;53:930–2.
46. Kennedy SH, Sargent IL, Starkey PM, Hicks BR, Barlow DH. Localization of anti-endometrial antibody binding in women with endometriosis using a double-labelling immunohistochemical method. Br J Obstet Gynaecol 1990;97:671–4.
47. Wild RA, Shivers CA. Antiendometrial antibodies in patients with endometriosis. Am J Reprod Immunol Microbiol 1985:8:84–6.
48. Muse K. Endometriosis and infertility. In: Wilson E, ed. Endometriosis. New York: Alan R. Liss, Inc., 1987:91–110.
49. el-Roeiy A, Dmowski WP, Gleicher N, Radwanska E, Harlow L, Binor Z, et al. Danazol but not gonadotropin-releasing hormone agonists suppress autoantibodies in endometriosis. Fertil Steril 1988;50: 864–71.
50. Kennedy SH, Starkey PM, Sargent IL, Hicks BR, Barlow DH. Antiendometrial antibodies in endometriosis measured by an enzyme-linked, immunosorbent assay before and after treatment with danazaol and nafarelin. Obstet Gynecol 1990;75:914–8.
51. Oral E, Olive DL, Arici A. The peritoneal environmental in endometriosis. Hum Reprod Update 1997;2:385–98.
52. Sillem M, Prifti S, Monga B, Arslic T, Runnebau B. Integrin-mediated adhesion of uterine endometrial cells from endometriosis patients to extracellular matrix proteins is enhanced by tumor necrosis factor alpha (TNFα) and interleukin-1 (IL-1). Eur J Obstet Gynecol Reprod Biol 1999;87:123–7.
53. Witz CA, Montoya-Rodriquez IA, Schenken RS. Whole explants of peritoneum and endometrium: a novel model of the early endometriosis lesion. Fertil Steril 1999;71:56–60.
54. Witz CA, Takahashi A, Montoya-Rodriguez IA, Cho S, Schenken RS. Expression of the $\alpha 2\beta 1$ and $\alpha 3\beta 1$ integrins at the surface of mesothelial cells: a potential attachment site of endometrial cells. Fertil Steril 2000;74:579–84.
55. Gaetje R, Kotzian S, Herrmann G, Baumann R, Starzinski-Powitz A. Invasiveness of endometriotic cells in vitro. Lancet 1995;346:1463–4.
56. Spuijbroek MDEH, Dunselman GAJ, Menheere PPCA, Evers JL. Early endometriosis invades the extracellular matrix. Fertil Steril 1992;58:929–33.
57. Cornillie FJ, Oosterlynck D, Lauweryns JM, Koninckx PR. Deeply infiltrating pelvic endometriosis: histology and clinical significance. Fertil Steril 1990;53:978–83.
58. Saito T, Mizumoto H, Kuroki K, Fujii M, Moir S, Kudo R. Expression of MMP-3 and TIMP-1 in endometriosis and the influence of danazol. Acta Obstet Gynecol Jpn 1995;47:495–6.
59. Osteen KG, Bruner KL, Sharpe-Timms KL. Steroid and growth factor regulation of matrix metalloproteinase expression and endometriosis. Semin Reprod Endocrinol 1996;14:247–55.
60. Bruner KL, Matrisian LM, Rodgers WH, Gorstein F, Osteen KG. Suppression of matrix metalloproteinases inhibits establishment of ectopic lesions by human tissue in nude mice. J Clin Invest 1997;99: 2851–7.
61. O'Reilly LA, Strasser A. Apoptosis and autoimmune disease. Inflamm Res 1999;48:5–21.
62. Imai A, Takagai A, Tamaya T. Gonadotropin-releasing hormone analog repairs reduced endometrial cell apoptosis in endometriosis in vitro. Am J Obstet Gynecol 2000;182:1142–6.
63. Gebel HM, Braun DP, Tambur A, Frame D, Rana N, Dmowski WP. Spontaneous apoptosis of endometrial tissue is impaired in women with endometriosis. Fertil Steril 1998;69:1042–7.
64. Meresman GF, Vighi S, Buquet RA, Contreras-Ortiz O, Tesone M, Rumi LS. Apoptosis and expression of Bcl-2 and Bax in eutopic endometrium from women with endometriosis. Fertil Steril 2000;74: 760–6.
65. Eisen HN. General immunology. 2nd rev. ed. Philadelphia: JB Lippincott Co., 1980.
66. Infante R, Lahita RG. Rheumatoid arthritis: new disease-modifying and anti-inflammatory drugs. Geriatrics 2000;55:30–40.
67. Singer CF, Marbaix E, Lemoine P, Courtoy PJ, Eeckhout Y. Local cytokines induce differential expression of matrix metalloproteinases but not their tissue inhibitors in human endometrial fibroblasts. Eur J Biochem 1999;259:40–5.
68. Rawdanowicz TJ, Hampton AL, Nagase H, Woolley DE, Salamonsen LA. Matrix metalloproteinase production by cultured human endometrial stromal cells: identification of interstitial collagenase, gelatinase-A, gelatinase-B, and stromelysin-1 and their differential regulation by interleukin-1 alpha and tumor necrosis factor-alpha. J Clin Endocrinol Metab 1994;79:530–6.
69. Zhang J, Nie G, Jian W, Woolley DE, Salamonsen LA. Mast cell regulation of human endometrial matrix metalloproteinases: a mechanism underlying menstruation. Biol Reprod 1998;59:693–703.
70. Moghissi KS. Medical treatment of endometriosis. Clin Obstet Gynecol 1999;42:620–32.
71. Clemens LE, Sitter PK, Stites DP. Mechanism of immunosuppression of progesterone on maternal lymphocyte activation during pregnancy. J Immunol 1979;122:1978–85.
72. Schust DJ, Anderson DJ, Hill JA. Progesterone-induced immunosuppression is not mediated through the progesterone receptor. Hum Reprod 1996;11:980–5.
73. Hill JA, Barbieri RL, Anderson DJ. Immunosuppressive effects of danazol in vitro. Fertil Steril 1987;48:414–8.
74. Mori H, Nakagawa M, Itoh N, Wada K, Tamaya T. Danazol suppresses the production of interleukin-1β and tumor necrosis factor by human monocytes. Am J Reprod Immunol 1990:24:45–50.
75. Ota H, Igarashi S, Hayakawa M, Matsui T, Tanaka H, Tanaka T. Effect of danazol on the immunocompetent cells in the eutopic endometrium in patients with endometriosis: a multicenter cooperative study. Fertil Steril 1996;65:545–51.
76. Wu M-Y, Chao K-H, Chen S-U, Chen H-F, Yang Y-S, Huang S-C, et al. The suppression of peritoneal cellular immunity in women with endometriosis could be restored after gonadotropin releasing hormone agonist treatment. Am J Reprod Immunol 1996;35:510–6.
77. Chen HF, Jeung EB, Stephenson M, Leung PC. Human peripheral blood mononuclear cells express gonadotropin-releasing hormone (GnRH), GnRH receptor and interleukin-2 receptor gamma-chain messenger ribonucleic acids that are regulated by GnRH in vitro. J Clin Endocrinol Metab 1999;84:743–50.
78. Harada T, Enatsu A, Mitsunari M, Nagano Y, Ito M, Tsudo T, et al. Role of cytokines in progression of endometriosis. Gynecol Obstet Invest 1999;47(suppl 1):34–40.
79. Imai A, Ohno T, Iida K, Fuseya T, Furui T, Tamaya T. Presence of gonadotropin-releasing hormone receptor and its messenger ribonucleic acid in endometrial carcinoma and endometrium. Gynecol Oncol 1994;55:144–8.
80. Ikeda M, Taga M, Kurogi K, Minaguchi G. Gene expression of gonadotropin-releasing hormone, but not its receptor in human endometrium and decidua. Mol Cell Endocrinol 1997;135:165–8.
81. Borroni R, Di Blasio AM, Gaffuri B, Santorsola R, Busacca M, Vigano P, et al. Expression of GnRH receptor gene in human ectopic endometrial cells and inhibition of their proliferation by leuprolide acetate. Mol Cell Endocrinol 2000;159:37–43.
82. Adashi EY. Long-term gonadotropin-releasing hormone agonist therapy: the evolving issue of steroidal 'add-back' paradigms. Hum Reprod Update 1994;7:1380–97.

83. Kiesel L, Schweppe KW, Sillem M, Siebzehnrubl E. Should add-back therapy for endometriosis be deferred for optimal results? Br J Obstet Gynaecol 1996;103(suppl 14):15–7.
84. Watanabe Y, Nakamura G, Matsuguchi H, Nozaki M, Sano M, Nakano H. Efficacy of a low-dose leuprolide acetate depot in the treatment of uterine leiomyomata in Japanese women. Fertil Steril 1992; 58:66–71.
85. Wheeler JM, Knittle JD, Miller JD. Depot leuprolide acetate versus danazol in the treatment of women with symptomatic endometriosis: a multicenter, double-blind randomized clinical trial. II. Assessment of safety. The Lupron Endometriosis Study Group. Am J Obstet Gynecol 1993;169:26–33.
86. Warnock JK, Bundren JC, Morris DW. Depressive symptoms associated with gonadotropin-releasing hormone agonists. Depression Anxiety 1998;7:171–7.
87. Rachman M, Garfield DA, Rachman I, Cohen R. Lupron-induced mania. Biol Psychiatry 1999;45:243–4.
88. Steinleitner A, Lambert H, Roy S. Immunomodulation with pentoxifylline abrogates macrophage-mediated infertility in an in vivo model: a paradigm for a novel approach to the treatment of endometriosis-associated subfertility. Fertil Steril 1991;55:26–31.
89. Balasch J, Creus M, Fabregues F, Carmona F, Martinez-Roman S, Manau D, et al. Pentoxifylline versus placebo in the treatment of infertility associated with minimal or mild endometriosis: a pilot randomized clinical trial. Hum Reprod 1997;12:2046–50.
90. Nothnick WB, Curry TE, Vernon MW. Immunomodulation of rat endometriotic implant growth and protein production. Am J Reprod Immunol 1994;31:151–62.
91. Evers JLH. How to treat endometriosis-associated subfertility. Or should we? Focus Reprod 1993;III:9–15.
92. Gleicher N. Immune dysfunction—a potential target for treatment in endometriosis. Br J Obstet Gynecol 1995;102(suppl 12):4–7.
93. Ward A, Clissold SP. Pentoxifylline: a review of its pharmacodynamic and pharmacokinetic properties, and its therapeutic efficacy. Drugs 1987;34:50–97.
94. Maksymowych WP, Avina-Zubieta A, Luong MH, Russel A-S. An open study of pentoxifylline in the treatment of severe refractory rheumatoid arthritis. J Rheumatol 1995;22:625–9.
95. Reimund JM, Bumont S, Muller CD, Kenney JS, Kedinger M, Baumann R, et al. In vitro effects of pentoxifylline on inflammatory cytokine release in patients with inflammatory bowel disease. Gut 1997;40:475–80.
96. Strieter RM, Remick DG, Ward PA, Spengler RN, Lynch JP III, Larrick J, et al. Cellular and molecular regulation of tumor necrosis factor-alpha by pentoxifylline. Biochem Biophys Res Commun 1988; 155:1230–6.
97. Sullivan GW, Carper HT, Novick WJ Jr, Mandell GL. Inhibition of the inflammatory action of interleukin-1 and tumor necrosis factor (alpha) on neutrophil function by pentoxifylline. Infect Immun 1988; 56:1722–9.
98. Fox RI, Herrmann ML, Frangou CG, Wahl GM, Morris RE, Strand V, et al. Mechanism of action for leflunomide in rheumatoid arthritis. Clin Immunol 1999;93:198–208.
99. Jarvis B, Faulds D. Etanercept: a review of its use in rheumatoid arthritis. Drugs 1999;57:945–66.
100. Markham A, Lamb HM. Infliximab: a review of its use in the management of rheumatoid arthritis. Drugs 2000;59:1341–59.