Future Targets in Endometriosis Treatment: Targeting the Endometriotic Implant

Warren B. Nothnick[1,2*] and Xuan Zhang[1]

[1]*Departments of Obstetrics and Gynecology and* [2]*Molecular and Integrative Physiology, School of Medicine, University of Kansas Medical Center, Kansas City, KS, USA*

Abstract: Endometriosis is an enigmatic, debilitating disease which affects as many as 15% of all women of reproductive age and is characterized by pelvic pain and infertility. Current treatment regimes used to manage the disease do so by inducing a hypoestrogenic state. While the absence of circulating estrogen levels lead to a regression of the disease, this hypoestrogenism also induces many unpleasant side-effects. As such, these and other shortcomings of current drug therapies emphasize their limitations and the necessity for the development of novel endometriosis treatments. In this review, current therapies which target the biochemistry of the implant are discussed with emphasis on aromatase inhibitors, anti-angiogenic compounds and vascular-disrupting agents.

Key Words: Endometriosis, treatment, aromatase inhibitors, angiogenesis.

1. INTRODUCTION

Endometriosis is a disease which affects approximately 10 to 15 % of all women of reproductive age and as many as 40 to 50% of all women with infertility [1]. In the disease, endometrial tissue grows in ectopic locations, predominantly in the pelvic cavity. Endometriosis is characterized by the cheif complaints of pelvic pain, dysmenorrhea and infertility. The disease is thought to develop *via* reverse menstruation of viable endometrial tissue into the peritoneal cavity. However, because almost all women of reproductive age exhibit some degree of retrograde menstruation [2], it is postulated that some other factors must contribute to the development and progression of the disease. As evident from the literature, there is a strong assoication between the presence of endometriosis and an altered immune system/inflammatory reaction [3].

2. CURRENT TREATMENTS FOR ENDOMETRIOSIS

The primary goals of endometriosis treatment are to remove the endometriotic tissue, restore normal anatomy, prevent or at least delay progression of the disease and relieve the symptoms associated with the disease. Current treatments can be surgical, chemical or a combination of both with further targets within the context of chemical suppression. In this review, we will focus on current means of chemical suppression of the disease and its related symptoms.

The goals of the current medical modalities for endometriosis are to reduce endometriotic implant mass and to manage the pelvic pain associated with the disease. Current treatment regimes include estrogen and progesterone combinations, progestogens, anti-progestogens, danazol, and gonadotropin-releasing hormone agonists (GnRH-a; [4]). These drugs are equally effective in reducing the endometriotic implant mass (the severity of the disease) as well as reducing pelvic pain associated with endometriosis [4, 5]. A major advantage associated with the use of progestins is the more tolerable side effects and limited cost [4, 6], while early reports indicate that GnRH antagonist treatment is not associated with the "flare-up" effects so common with GnRHa therapies [7]. The mechanism by which these compounds suppress endometriotic implant mass has long been assumed to act primarily at the level of the hypothalamic-pituitary axis. Suppression of gonadotropin release and subsequent depletion of systemic estrogen levels is postulated to lead to a regression of the endometriotic mass and concurrently a reduction in pelvic pain due to the lack of estrogen [3]. The concept that estrogen is essential for endometriotic implant growth is well supported in both the human context of the disease and studies utilizing animal models of endometriosis.

Despite the fact that GnRH agonists are one of the most common medical therapies used to treat endometriosis, their use is associated with disadvantages and undesirable side effects, most of which stem from the induction of a hypoestrogenic state. One of the major limitations in using GnRH agonists in treating endometriosis is that their use is limited to 6 months because of possible adverse effects on bone metabolism [8]. This adverse effect on bone density can be overcome by hormone add-back therapy but add-back therapy may reduce the efficacy of GnRHa [9]. However, more recent data indicates that add-back therapy does not reduce the efficacy of GnRH agonists. [10]. None the less, additional drawbacks to GnRHa usage are evident and include physical side effects such as vasomotor instability, headache, and hot flashes [11, 12] as well as psychiatric side effects which include depressive mood symptoms [13, 14]. Lastly, the combination of estrogen add-back and Lupron therapy must be balanced to prevent disease progression and provide sufficient estradiol for maintenance of bone density [8].

*Address correspondence to this author at the Department of Obstetrics and Gynecology, University of Kansas Medical Center, 3901 Rainbow Boulevard, Kansas City, Kansas 66160, USA; Tel: 913 588 6277; Fax 913 588 7180; E-mail: wnothnic@kumc.edu

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One must consider the variability in therapeutic efficacy of the different types of GnRHa (Leuprolide, Nafarelin, Buserelin, Gosereln, etc.) coupled with the different doses, durations and forms of the "added back" steroids. For the most part, current data suggests that GnRHa 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 GnRHa such as bone loss. However, there are limitations and shortcomings of the current drug therapies. Development towards a more ideal therapy would focus on a drug that could induce regression of the disease and its' associated symptoms, but not have the detrimental effects associated with a hypoestrogenic state (such as bone loss). Further, these compounds may be directed at targeting the endometriotic implant as opposed to targeting systemic production or action of estradiol.

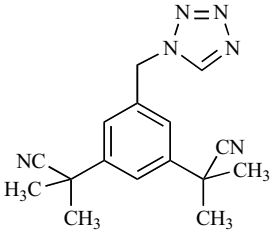
Currently, aromatase inhibitors have emerged as a major means of treating endometriosis. Primarily used in the treatment of breast cancer, these agents are hypothesized to induce regression of the disease and its associated symptoms by disrupting local estradiol production by the endometriotic implant (see Fig. **1**).

3. TREATING ENDOMETRIOSIS WITH AROMATASE INHIBITORS

The use of aromatase inhibitors to treat endometriosis was first proposed based upon the observation that endometriotic tissue expresses elevated levels of aromatase activity compared to eutopic endometrial tissue [15-18]. To date, anastrozole or letrozole combined with standard regimes have been used in both postmenopausal and premenopausal endometriosis patients successfully to alleviate chronic pelvic pain and reduce the lesion size [reviewed in 19] with mild side-effects. However, in some cases aromatase inhibitors led to ovarian cysts and in other cases recurrence happened during follow-ups after the treatment [19]. It appears that the use of aromatase inhibitors has to be individualized by carefully choosing the combined medical treatments to minimize side-effects and prevent recurrences.

3.1. Anastrozole

Anastrozole (chemical formula = 2,2'[5-(1H-1,2,4-triazol-l-ylmethyl)-1,3-phenylene]bis (2-methylpropiononitrile); trade name Arimidex; Fig. **2**) was first described for endometriosis treatment in a case report [20]. In this report, a post-menopausal patient was successfully treated with anastrozole reporting regression of the disease and absence of pelvic pain associated with the disease.



2,2'[5-(1H-1,2-triazol-1-ylmothyl) -1,3-phenylene]bis(2-methylpropiononitrile)

Fig. (2). Chemical structure and formula of Anastrozole.

Similar findings were reported in which anastrozole combined with progesterone, calcitriol and rofecoxib was given to two premenopausal endometriosis patients for six repeated 28-day cycles [21]. The treatments resulted in a rapid, progressive elimination of symptoms over 3 months with the maintenance of remission of symptoms for over a year after treatment in both cases.

In addition to these case reports, larger randomized controlled trials have been reported [22-24]. In a study by Soysal and colleagues [22], 80 premenopausal women were treated with either anastrozole and the GnRH agonist goserelin or goserelin alone following surgery for severe endometriosis. Co-treatment with anastrozole significantly increased the pain-free interval and decreased symptom recurrence rates. Although patients who received anastrozole co-treat-ment had higher spinal bone loss compared with the goserelin-only group at the completion of the treatment, menopausal quality of life and bone mineral density at 2 years after medical therapy remained unaffected.

In a prospective open-label phase II trial [23], 15 premenopausal patients with refractory endometriosis and chronic pelvic pain were treated with 1 mg anastrazole and 20 ug ethinyl estradiol/0.1 mg levonorgestrel daily for 6 months. This treatment led to significant pain relief in fourteen out of the 15 patients. Estradiol levels were suppressed

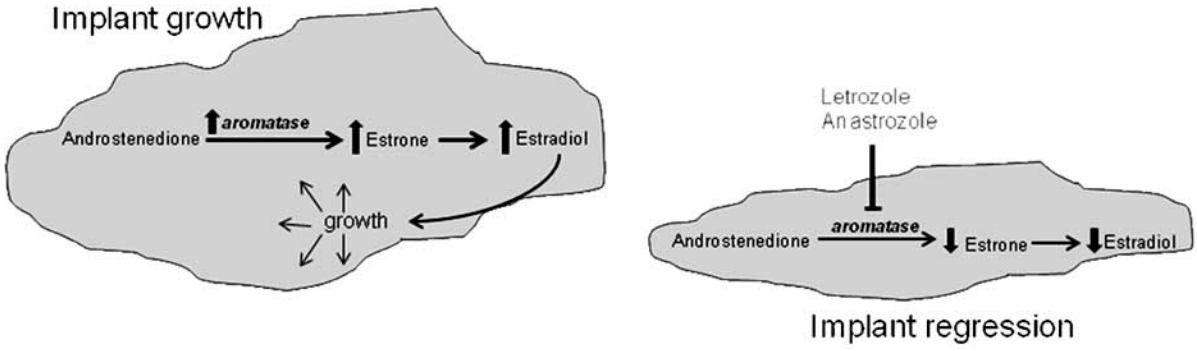


Fig. (1). Aromatase and edometriotic implant growth. Elevated production of the enzyme, aromatase, by endometriotic tissue drives local production of estradiol which in turn stimulates implant growth (figure on the left). Aromatase inhibitors such as letrozole and anastrozole inhibit production of the enzyme which reduces local production of estradiol by the implant. Reduction of endometriotic implant production of estradiol in turn leads to a regression of the endometriotic implant (figure on the right).

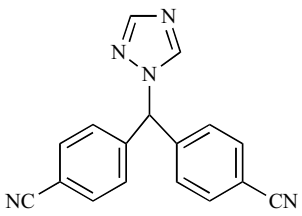
during the treatment, and side effects were mild and improved over time.

In a nonrandomized pilot study [24], anastrozole (0.25 mg anastrozole/d for 6 months) was vaginally administered to patients with rectovaginal endometriosis. This treatment resulted in an improvement in dysmenorrhea, physical and social functioning, but not in chronic pelvic pain and dyspareunia in a series of 10 patients during the therapy.

In addition to trails using anastrozole, studies have also been conducted using the third generation aromatase inhibitor, letrozole.

3.2. Letrozole

Letrozole (chemical formula = 4-[(4-cyanophenyl)-(1,2, 4-triazol-l-yl) methyl] benzonitrile; trade name Femara; Fig. (**3**) has also been used in the treatment of endometriosis.



4-[(4-cyanophenyl)-(1,2,4-triazol-1-yl)methyl]benzonitrile

Fig. (3). Chemical structure and formula of Letrozole.

In an open-label, non-randomized proof of concept study [25], 10 patients with endometriosis were evaluated. Letrozole was administered (2.5 mg) daily along with the progestin, norethindrone acetate (2.5 mg), calcium citrate (1,250 mg), and vitamin D (800 IU) for 6 months. After completion of the treatment, a second-look laparoscopy verified that histologically demonstrable endometriosis was not present in any patient. Also, pelvic pain scores decreased significantly in response to treatment and no significant change in bone density was detected. Perhaps more important was the finding that systemic estradiol levels were not different before and after treatment suggesting that letrozole did not induce regression of the disease by affecting systemic steroid levels.

Letrozole has also been used in case reports in which a 31 year old woman with recurrent endometriosis was given 2.5 mg letrozole, 1000 mg calcium and 400 IU vitamin D daily for 6 months [26]. The patient's pelvic pain and dyspareunia was significantly decreased during the treatment without a decrease in the bone density. Similarly, letrozole was also successfully used in a post-menopausal patient with recurrent endometrioma, who had undergone abdominal hysterectomy and salpingo-oophorectomy a decade earlier [27]. After eighteen months of treatment, the endometrioma was almost completely regressed and the patient was free of symptoms. Further, an additional case report [28], in which letrozole was administered to a middle-aged woman with endometriosis and severe pelvic pain after hysterectomy and bilateral salpingo-oopherectomy demonstrated that letrozole successfully relieved pain associated with the disease.

While these findings are promising, it should be noted that not all the clinical trials using letrozole in endometriosis were successful. In an open-label prospective study in 2007 [29], letrozole (2.5 mg/day) combined with norethisterone acetate (2.5 mg/day) was given to patients with pain symptoms related to the presence of rectovaginal endometriosis. Although the intensity of symptoms was reduced during the treatment, pain recurred at 3-month follow-up. Five women underwent surgery during the follow-up, and histological examination of rectovaginal nodules revealed the presence of active endometriotic lesions. In another open-label prospective study [30], 12 women with stage IV refractory endometriosis were treated with daily oral administration of letrozole (2.5 mg), desogestrel (75 ug), elemental calcium (1000 mg) and vitamin D (880 I.U.). None of the patients completed the scheduled six-month treatment because they all developed ovarian cysts. At interruption of treatment, all women reported significant improvements in dysmenorrhea and dyspareunia, but pain symptoms quickly recurred at three-month follow up.

In addition to human trials animal studies have also evaluated other aromatase inhibitors. In two separate studies, rats with surgically-induced endometriosis were treated with the non-steroidal aromatase inhibitors fadrozole hydrochloride [31] or YM511 [32]. While treatment resulted in dose-dependent volume reduction of the endometriotic implants, the use of these products has not yet been reported in humans.

Overall, it appears that aromatase inhibitors are successful in treating endometriosis with respect to the disease and its associated symptoms. However, there may be limitations with respect to the stage of disease and the effectiveness of these compounds. Clearly, additional trials are needed which encompass a larger and more diverse population of patients with respect to disease stage.

While the use of aromatase inhibitors has become more common in human trials, additional agents which target the endometriotic implant have also been investigated. One potentially promising target area may be the disruption of endometriotic implant angiogenesis. Disrupting or severely reducing the blood supply to the implants would in turn lead to a regression of the disease and in turn the symptoms brought about by their presence.

4. FUTURE TARGETS

4.1. Angiogenesis Disruptors

There is considerable evidence that vascular development is important in the pathogenesis of endometriosis [33, 34-36] and agents which target the angiogenic process have begun to attract attention in endometriosis treatment [37]. It is well established that peritoneal fluid from women with endometriosis has greater angiogenic activity [34] and concentrations of the angiogenic agent, vascular endothelial growth factor (VEGF) [35, 36] compared to that of women without the disease. Further, early endometriotic lesions are characterized by high vascular density [38, 39]. As such, investigation into the potential use of anti-angiogenic or vascular-disrupting therapies in the treatment of endometriosis

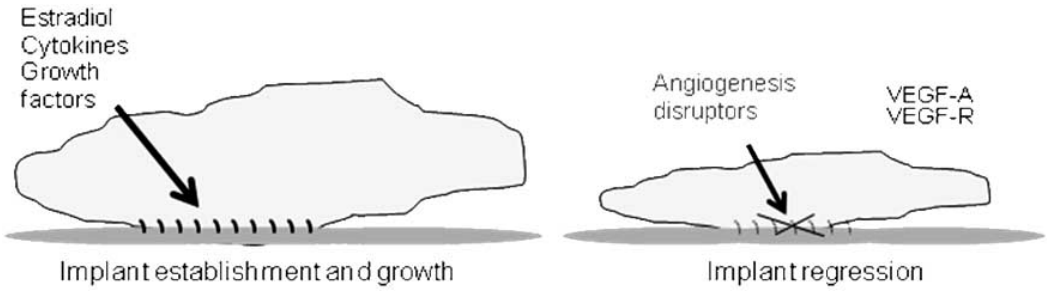


Fig. (4). Angiogenesis and endometriotic implant growth. Angiogenesis within the endometriotic tissue is essential for establishment and growth of the endometriotic implant (figure on the left). Angiogenesis disrupters may hibit the development and/or maintenance of blood vessel growth by targeting vascular endothelial growth factor A (VEGF-A) and or its receptor (VEGF-R). Inhibition of vascular development in turn leads to death of the implant and regression of the disease (figure on the right).

has just begun to receive attention. To date, this work has been limited to animal models but appears to hold great promise. One of the first studies was conducted by Hull and colleagues using the nude mouse model for endometriosis [40]. In this study, the investigators examined the effectiveness of anti-VEGF-A therapy (both a soluble truncated receptor (flt-1) and an affinity-purified antibody to human VEGF-A) in preventing the establishment of endometriosis in nude mice. The authors found that both anti-angiogenic agents inhibited the growth of endometriotic implants by disrupting the vascular supply. An additional study by Nap and colleagues [41] demonstrated that angiostatic compounds significantly decreased microvessel density and the number of endometriotic lesions suggesting that inhibitors of angiogenesis interfere with the maintenance and growth of ectopic endometrial tissue. Further work has been presented by Becker and coworkers [42-44] examining the effect of endostatin [42], endostatin peptides [43] and the angiogenesis inhibitor, 2-methoxyestradiol [44]. Collectively, these studies support the notion that disruption of angiogenesis and established vascular blood supplies may be promising targets for treatment of endometriosis (Fig. **4**). However, there are several factors which must be taken into consideration with respect to their possible application into endometriosis treatment. First, anti-angiogenic agents may only be useful for early stage disease as they may not effectively target pre-existing pericyte-protected vessels. The possible necessity for chronic administration to overcome this shortcoming coupled with the potential adverse affects on other processes (such as reproductive function) must be thoroughly evaluated in additional animal models. Further, while vascular-disrupting agents (which target the existing blood supply) theoretically may be useful to target established disease, the potential impact on the reproductive process also needs to be examined. With these additional studies and extension into the human, treatment of endometriosis with such agents may be in the not too distant future.

CONCLUSIONS

Endometriosis remains one of the most perplexing diseases for patients, physicians and researchers. The difficulty in coping with, treating and understanding this disease is derived from the poor understanding of how and why the disease develops. While the majority of current therapies are successful in treating the disease and its symptoms, these treatments are not ideal as they induce an early menopause. More ideal treatments would be able to suppress the disease and alleviate the pain associated with the disease while not preventing ovulation or menstruation. The theory behind such compounds as aromatase inhibitors, anti-angiogenic compounds and vascular-disrupting agents is an attractive one as it seeks to target processes which are critical to the establishment and maintenance of the disease tissue. While these agents hold promise, additional studies must be conducted to examine their potential long-term use and side effects. Future identification of endometriotic implant "specific" molecules which are critical for the establishment and maintenance of the disease tissue may provide pharmacological targets which will allow for the treatment of the disease independent of impacting other vital biological processes.

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