EFFECTS OF TIBOLONE ON THE BREAST OF POSTMENOPAUSAL WOMEN

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SUMMARY

For decades, hormone therapy (HT) has been the mainstay for managing menopausal symptoms. However, the prolonged use of either single estrogen therapy (ET) or a combination therapy of estrogen and progestogen (EPT) might be associated with a slightly increased risk of breast cancer. Alternative therapies that are effective in the prevention and/or treatment of menopause, having associated morbidities but no unwanted effects, are of primary interest in clinical practice. Tibolone (Livial; NV Organon, Oss, The Netherlands) is structurally related to 19-nortestosterone derivatives and is a new postmenopausal regimen with a unique pharmacological profile, licensed for the relief of climacteric symptoms and the prevention of osteoporosis in postmenopausal women. Tibolone exhibits weak estrogenic, progestogenic, and androgenic activities, which in theory might influence the breast. The effect of tibolone on breast tissue, however, is obscure. The purpose of this study was to assess the effects of tibolone on breast safety, and the collected data include preclinical models, clinical observation, and epidemiologic study. Although *in vitro* studies showed conflicting results (with the majority being favorable effects) regarding the effects of tibolone on breast cells, *in vivo* studies showed favorable effects of tibolone on the breast in animal models. Similarly, an epidemiologic study indicated an increased risk of breast cancer when tibolone was used to manage climacteric symptoms of postmenopausal women, but accumulated data obtained from radiologic studies (mammography) showed a possible protective effect of tibolone on the breast. Taken together, we conclude that tibolone, if not superior to conventional HT, may be more acceptable to clinicians as a therapeutic drug option for use with symptomatic menopausal women. Only time will tell whether tibolone will be the preferred option. [*Taiwan J Obstet Gynecol* 2007;46(2):121–126]

Key Words: breast cancer, estrogen, hormone therapy, progestogen, tibolone

Hormone Therapy and Breast Cancer

Menopause occurs naturally when the ovarian follicular pool is functionally exhausted, or it can also be induced by surgical removal of both ovaries. The resulting hypoestrogenic state may adversely affect the estrogen target tissues, which include the brain, skeleton, skin, and the cardiovascular and genitourinary systems [1]. The reaction of target tissues to estrogen deficiency, and the resultant frequency and severity of the climacteric symptoms vary significantly among women. These climacteric symptoms sometimes bother peri- and/or postmenopausal women, resulting in severe interference in their quality of life [2]. There are two broad categories of menopausal hormone therapy (HT): estrogen alone therapy (ET), and estrogen combined with progestogen therapy (EPT) [1]. The goals of menopausal hormone therapies are to (1) reduce symptoms resulting from estrogen depletion, including hot flushes, sleeplessness, lethargy, and depressed mood; (2) treat urogenital atrophy and vaginal dryness; and (3) minimize the risk of disorders that may be more frequent during HT [1]. Although ET and EPT may improve a woman's quality of life, each woman has a unique risk profile which might lead to more, or less, benefit from HT [1].

Most importantly, concerns have arisen regarding the possible association of breast cancer and HT. An association between breast cancer and hormone use would

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be plausible because breast cancer incidence is increased by hormonal factors such as early menarche and late menopause [3]. In a 1997 reanalysis of 51 epidemiologic studies, which included more than 90% of the world's literature on breast cancer and hormone use, breast cancer risk increased by 2.3% per year of hormone use (mostly estrogen use) compared with an increased risk of 2.8% per year for natural delay in the onset of menopause [4], suggesting that HT does increase the risk of developing breast carcinoma and that this risk increases with the increasing duration of HT use. A review of the 19 epidemiologic studies published after 1997 estimated the average breast cancer risks to be 1.18 (95% confidence interval, CI, 1.01–1.38) with current use of ET and 1.70 (95% CI, 1.36–2.17) with current use of EPT [5].

Data have accumulated in randomized clinical trials involving more than 30,000 women and in epidemiologic studies involving more than 1.8 million women [1,5]. With the use of ET, the average risk of invasive breast cancer was 0.81 (95% CI, 0.63–1.03) in four randomized trials involving 12,643 women [6–9]. With the use of EPT, the average breast cancer risk was 1.24 (95% CI, 1.03–1.50) in four randomized trials involving 19,756 women [10–13]. The absolute effect of EPT in the Women's Health Initiative, and Heart and Estrogen/Progestin Replacement Study trials added 8 and 17 cases per 10,000 women per year, respectively, to the natural risk [12,14]. Breast cancer risk does not vary significantly with different types of estrogen or progestin preparations, with use of lower dosages or with different routes of administration [5]. In six epidemiologic studies, including the Million Women Study [15], the average relative risks with sequential and continuous progestin regimens were 1.85 (95% CI, 1.72–1.99) and 1.94 (95% CI, 1.78–2.11), respectively, a difference that was not significant [5].

Although in the epidemiologic studies, the increased breast cancer risk diminished soon after discontinuing hormones and largely disappeared by 5 years after cessation [5], the use of HT as a standard treatment applied to all menopausal women will not meet the needs of many individual women [1]. Health care providers should therefore consider the relative balance between the benefits and risks of treatment for each patient before drawing conclusions or recommending HT. Therefore, an alternative, such as tibolone that delivers the beneficial but not the adverse side effects of steroid hormones, would be clinically advantageous [16]. However, data from the Million Women Study suggested that tibolone increases the risk of breast cancer [15]; but when compared with conventional HT, tibolone seems to be a relatively safe medication in terms of this risk [2]. Even with the increased risk to current users of 1.30 (1.21–1.40, $p < 0.0001$), 2.00 (1.88–2.12, $p < 0.0001$), and 1.45 (1.25–1.68, $p < 0.0001$) for ET, EPT, and tibolone, respectively, the magnitude of the associated risk was substantially greater for EP than for other types of HT ($p < 0.0001$) [15]. In order to understand the role of tibolone therapy on the breast in postmenopausal women, we reviewed recent studies addressing the tibolone effect on the breast, including preclinical models, clinical observation, and epidemiologic study.

Tibolone

Tibolone (Livial®), which has been on the market since 1988, is a new postmenopausal regimen with a unique pharmacologic profile, licensed for the relief of climacteric symptoms and the prevention of osteoporosis in postmenopausal women [17–19]. Although tibolone is frequently described as a HT product, recent research has provided a clearer insight into the mechanism and action of tibolone, which is significantly different from those of conventional HT, such as ET and EPT [17]. The interesting clinical profile of tibolone was already apparent in the first study into its effects on bone [20]. At that time, the estrogenic, progestogenic, and androgenic properties were known from classical bioassays, but they could not fully explain the observed tissue-selective effects, such as the estrogenic-like activity on bone, vagina, and brain, and the non-stimulating effects on the endometrium [20].

Tibolone lacks an aromatic A-ring and the 3-hydroxyl substituent, normally required for agonist activity at estrogen receptors (ERs), and yet its effects on brain, bone, and vagina are estrogen-like. This indicated that the estrogenic activity is due to metabolism into estrogenic metabolites [17]. It was indeed subsequently found that tibolone can be converted into two 3-hydroxy metabolites (i.e. 3-α and 3-β hydroxy metabolites) with estrogenic activity. Although the binding affinity of these two estrogenic metabolites is low, the high levels found in the circulation mean that a full biologic response can be obtained [17], and the estrogenic activity is exerted in a tissue-selective manner [21–30]. In addition, these two 3-hydroxy metabolites of tibolone show selectivity for the classical ERα over the ERβ isoform [17]. Both hydroxy-metabolites have a half-life of approximately 7 hours, but circulatory levels of the 3α-hydroxy metabolite are about four times higher than those of the 3β-hydroxy metabolite.

Tibolone has a 3-keto-Δ5-10 steroid structure with 17α-ethinyl and 7α-methyl groups; it is very rapidly metabolized to 3α- and 3β-hydroxy tibolone by hepatic and intestinal 3α- and 3β-hydroxysteroid dehydrogenase

(HSD) [17]. Tibolone is not a substrate for 3β-HSD type I or type II, and the conversion is therefore most likely due to 3β-HSD activity residing in 17β-HSD type II [17]. A third metabolite, the Δ^4-isomer of tibolone, is formed directly from tibolone by 3β-HSD-isomerase, for which the 3β-hydroxy metabolite is also a potential substrate. The Δ^4-isomer of tibolone activates the progesterone and androgen receptors, but not the ER [31], and has a higher affinity for progesterone receptor type B than for type A.

The majority of the metabolites of tibolone (approximately 80%) are in the inactive mono- and disulfated forms, from which active estrogenic 3-hydroxy metabolites may be continuously formed via the sulfatase enzyme [32], the extent depending on the local enzyme activity level. The 3α-hydroxy sulfated tibolone is, as expected, the main sulfated metabolite [17]. Like the parent compound, the Δ^4-isomer is rapidly removed from the circulation [17].

In vitro Studies of Tibolone on Breast Epithelial Cells and Breast Cancer Cells

Tibolone and its metabolites have been shown to be very potent inhibitors in the conversion of estrone sulfate to estradiol in the hormone-dependent breast cancer cell [27–29]. Studies have shown that the metabolites of tibolone regulate the activity of local enzymes normally involved in the production of active estrogens in the breast [30]. Tibolone has a different effect than conventional HT at the level of the breast. It does not seem to stimulate breast tissue and might have an inhibitory effect on the growth of human breast tumor cells *in vitro* [31], in addition to slowing down the proliferation rate and increasing differentiation and apoptosis [27]. In normal breast cells, tibolone and its Δ^4-isomer significantly increased apoptosis, as indicated by flow cytometry or morphologic analysis [32], because the authors found that the proportion of cells showing apoptotic features, such as blebbing, DNA fragmentation, and nuclear destruction, was increased by tibolone and its metabolites. To elucidate the possible mechanism, the authors found that tibolone and its Δ^4-isomer induced 17β-HSD activity and strongly inhibited expression of Bcl-2 in normal breast cells [33]. The decrease in Bcl-2 and slight decrease in Bcl-xl expression support the effect of tibolone and the Δ^4-metabolite on apoptosis, but these parameters were not influenced by the two 3-hydroxy metabolites, thus suggesting that another pathway may be involved [29]. The antiproliferative effect of tibolone and its Δ^4-metabolite, together with their proapoptotic effect in breast tissue, suggests that these substances may have beneficial effects on normal and breast cancer cells [34].

In contrast, some studies showed conflicting results [35]. In one study, tibolone was examined alone and in the presence of 0.1 nM estradiol in the concentration range of 0.001 μM to 1 μM. Tibolone led to significant cell growth in the concentration range of 0.01 μM to 1 μM and was not able to inhibit estradiol-induced proliferation at the concentrations of 0.01 μM and 0.1 μM in the breast cancer cell experiments [35]. The authors concluded that drawing a clinical consequence from their experiments would result in not recommending the use of tibolone in postmenopausal women at high risk for breast cancer development until long-term controlled clinical studies have been performed on the effect of tibolone administration and breast cancer risk [35].

In vivo Studies of Tibolone on Breast Epithelial Cells and Breast Cancer Cells

The well-known 7,12-dimethylbenz(a)anthracene (DMBA)-rat model was used to test the effect of tibolone, both in treatment and prophylactic protocols, on tumor growth and initiation in rats. Tibolone inhibited tumor growth at least as effectively as tamoxifen [30, 36]. In the prophylactic protocol, tibolone was extremely efficacious in preventing tumor development. It was a surprising observation that tibolone is converted mainly to estrogenic metabolites. In ovariectomized monkeys, 2 years of treatment with tibolone did not stimulate the breast, whilst a combination of conjugated equine estrogens (CEE) plus medroxyprogesterone acetate (MPA) resulted in a clear stimulation. The metabolism of tibolone in this model monkey model was comparable to that in postmenopausal women. Similarly, normal breast tissue in nude mice was not stimulated by tibolone as shown by testing its effect on estrogenic-sensitive parameters [36]. In the same model, transplanted MCF7 cells were also not stimulated by tibolone [36]. Receptor studies using antagonistic transactivation assays ruled out the possibility of an antagonistic effect of tibolone, or one of its metabolites, on any of the steroid receptors [37]. Furthermore, the antitumor effect of tibolone in the DMBA model was not sensitive to flutamide, thus suggesting that it was not an androgenic effect [36]. In ovariectomized monkeys, it has been shown that tibolone did not increase the proliferation marker Ki67, whilst an increase was seen with CEE plus MPA [38]. Valdivia et al even observed a significant decrease in Ki67 and a concomitant increase in apoptosis in the biopsy specimens of postmenopausal women treated with tibolone [39].

Breast Tenderness in Women Treated with Tibolone

In women, the lack of stimulating effects of tibolone on the breast is illustrated by the absence of an increase in breast tenderness, as seen with other HT [2,40]. Numerous studies have shown that breast pain is increased in a small percentage of women treated with tibolone; combined published data showed that 6% of women experience an increase and the same percentage showed a decrease [36]. In a recent placebo-controlled trial conducted in 64 women who had reported breast symptoms with a range of hormone replacement regimens, switching to tibolone or placebo resulted in a significant reduction in breast tenderness and mastalgia [41].

Mammographic Change in Women Treated with Tibolone

An increase in mammographic density should be regarded as an unwanted side effect of HT [42], because increased breast density can impair the interpretation of mammograms, thus increasing the failure rate of breast cancer screening programs [36]. Tibolone does not increase breast density and therefore does not negatively affect mammographic screening for breast cancer. Women on tibolone therefore require fewer repeat mammograms. Valdivia et al [43] reported a decrease in mammographic density compared with baseline after 1 year of tibolone treatment, whereas treatment with EPT induced an increase in breast density. In a randomized, double-blind study comparing the effects of tibolone and placebo in 20 women with mastopathy, a reduction in breast density was observed in 40% of the women receiving tibolone and 10% of those given placebo [44]. A recently published article on the long-term effects of tibolone on mammographic density elaborated on tibolone's unique effects on the breast and suggested that tibolone seems to have a minimal effect on mammographic density [21]. All of the above suggest that tibolone has an advantage over conventional HT in terms of breast cancer risk. Moreover, tamoxifen is often used as adjuvant therapy and/or preventive therapy for breast cancer. Unfortunately, many women with breast cancer suffer vasomotor symptoms rather than risk recurrence with conventional HT. In a small randomized controlled trial in women with early breast cancer undergoing adjuvant tamoxifen treatment, tibolone reduced hot flushes and night sweats and improved quality of life compared with placebo [45]. Dimitrakakis' group designed an observational, prospective, open, non-randomized study to assess the safety and efficacy of tibolone for the treatment of climacteric symptoms in women who had a history of breast cancer which was treated with a complete (5-year) course of tamoxifen therapy [46]. The women were followed up for a mean duration of 61 months follow-up, and the authors found that tibolone was effective in the treatment of climacteric symptoms and was well tolerated in the tibolone group of 52 women. The cancer recurrence rate in the tibolone group was comparable to that of the untreated controls [46]. Therefore, the authors concluded that the overall safety and tolerance were similar to those of the general population of postmenopausal women treated with tibolone [46]. Moreover, tibolone has been shown to prevent the increase in hot flushes in postmenopausal women given tamoxifen following surgery for breast cancer without untoward effects on the endometrium [47]. Clinically, tibolone does not act like an estrogen on the endometrium (i.e. no hyperplasia), thus indicating that sufficient progestogenic activity is exerted by tibolone treatment. Progestogenic activity of tibolone is less expressed, if at all, in the breast.

Potential Benefits of Tibolone in Treating Menopausal Syndrome

Although menopause is a natural course in women, some women suffer from symptoms which significantly affect their lives. In addition, circulating levels of estrogen are reduced in postmenopausal women, but the breast tissue of postmenopausal women is able to synthesize estrogens locally, which explains why the risk of breast cancer is not minimized after menopause. In fact, estradiol is the main factor supporting the growth and evolution of breast cancers, and one of the pathways involved in the transformation of estradiol is the sulfatase pathway, which transforms estrone sulfate to estradiol [27], especially after menopause.

Numerous clinical studies have shown that tibolone has beneficial effects in relation to climacteric symptoms and vaginal atrophy in postmenopausal women; relief from climacteric symptoms develops within 3–5 weeks, and the maximum effect is usually seen by 3 months [48]. In addition, tibolone shows beneficial effects on mood and sexual well-being [49]. The results of this observation demonstrate a similar positive effect in improving insomnia, libido, and mood instability, and most importantly, there was a high rate of continuous use of tibolone. Moreover, the low dosage (1.25 mg/day) seemed to be effective for women with climacteric symptoms. In this observation study, seven women were

reported to be satisfied with this low dosage treatment, which is in agreement with a previous report showing that tibolone induced a decrease in the frequency and intensity of climacteric symptoms, leading to statistically significant differences compared to placebo, for dose levels of 1.25 mg and higher [50].

Concerning irregular vaginal spotting and/or possible unwanted pathologic changes to the endometrium, tibolone therapy is reported to have high rates of amenorrhea after 10 years, with minimal evidence of adverse effects on endometrial pathology [51]. In our previous study, we also found that irregular bleeding episodes were markedly decreased during and at the end of treatment [2].

In terms of biochemical changes in the blood, tibolone is reported to lower lipoprotein(a), fibrinogen, and plasminogen activator inhibitor-1 levels and to improve glucose tolerance, insulin sensitivity, and endothelial function. However, it also lowers high-density lipoprotein cholesterol by more than 20% [52]. Therefore, the long-term impact of tibolone on the risk of coronary heart disease is not known and needs to be studied [52].

With the beneficial effect of preventing and/or treating osteoporosis, tibolone prevents bone loss and has been shown to increase the bone mineral density in early and late postmenopausal women [53].

Conclusion

Although *in vitro* studies showed conflicting results regarding the effects of tibolone on breast cells, *in vivo* studies showed a protective effect of tibolone on the breast in animal models. Although epidemiologic studies show an increase in the risk of breast cancer among women treated with tibolone, accumulation of data obtained from radiologic studies presents promising results. Since tibolone, as a selective tissue estrogenic activity regulator [50], is easy to use, only a single tablet containing 2.5 mg of tibolone is needed every day [2]. Tibolone, if not superior to conventional HT, may be more acceptable to clinicians as a therapeutic drug option for symptomatic menopausal women. Time will tell.

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