The Effect of Menopausal Hormone Therapies on Breast Cancer: Avoiding the Risk

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KEYWORDS

• Estrogens • Progestogen • Hormone therapy • Breast cancer • Menopause

KEY POINTS

- Menopause is often accompanied by significant symptoms that affect quality of life; concerns over breast cancer risk is the principle reason women may choose to avoid treatment.
- Data from prospective randomized trials confirm an increased risk of breast cancer associated with long term use of combined estrogen and progestin (P) hormone therapy.
- In contrast with the effects of combined estrogen and P, the risk of breast cancer was decreased after use of estrogen in the WHI estrogen alone trial.
- Progestogens, not E, seem to convey the risk of breast cancer; however, estrogen cannot be used alone in a woman with a uterus, given the known risks of long-term exposure.
- The recent addition of bazedoxifene combined with conjugated estrogen provides a progestin-free regimen that can be used in a woman with a uterus.

INTRODUCTION

Menopausal hormone therapy (MHT) is an effective treatment for menopausal symptoms, and based on observational studies demonstrating numerous beneficial effects, was popularized as a first-line approach to menopause management. MHT was found to be very effective in treating vasomotor symptoms and preventing osteoporosis.[1] It was also thought to reduce the risk of coronary heart disease.[1] These findings provided support for broadening the use of MHT in an effort to help prevent age-related deficits associated with loss of sex steroid hormones. Thus, MHT was heralded for use in the prevention of disease in postmenopausal women.[2] Although

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breast cancer has always been a risk associated with MHT, the effects of treatment on other life-threatening diseases, namely cardiovascular disease, were thought to outweigh the risk of breast cancer. However, randomized controlled trials (RCTs) demonstrated that MHT did not afford the positive benefits that had previously been predicted from observational studies.[1] On the contrary, in some instances, it was found to increase the risks for breast cancer, heart disease, and pulmonary embolism.[1] The Women's Health Initiative (WHI) Trial, in its landmark study findings in 2002, reversed many of the perceptions of positive health benefits of MHT that were seen in observational studies.[1,2] Briefly, the WHI hormone trials were RCTs of postmenopausal women aged 50 to 79 (average age, 63) designed to determine whether or not MHT (estrogen only, and estrogen plus progestin combination) prevented cardiovascular disease. A global index was designed to assess the risks and benefits of MHT with respect to coronary heart disease, breast cancer, stroke, pulmonary embolism, endometrial cancer, colorectal cancer, hip fracture, and death by other causes. The WHI trials looked at combination of conjugated equine estrogen (CEE) and medroxyprogesterone acetate (MPA; together referred to as EPT) use in postmenopausal women with an intact uterus, and conjugated estrogen therapy (ET) use in those with prior hysterectomy. In the WHI EPT arm, women were randomized to receive 0.625 mg/d of CEE plus 2.5 mg MPA or placebo, whereas CEE alone was compared with placebo in the ET trial.[1]

Although the WHI trials failed to demonstrate reduction in risk or coronary heart disease with use of MHT, with regard risk to the breast cancer, these trials yielded paradoxic and intriguing data. Long-term use of EPT was associated with an increased risk of breast cancer (hazard ratio [HR], 1.25; 95% CI, 1.07–1.46; $P = .004$)[3]; the risk, however, was reversed in women who had had a hysterectomy and were randomized to estrogen alone (HR, 0.82; 95% CI, 0.65–1.04).[4]

In this review, we specifically focus on the risk of breast cancer associated with MHT. Without the potential to extend life through reduction of cardiovascular disease, a risk/benefit analysis on the use of MHT is rendered substantially less favorable. The risk of breast cancer has become the greatest concern to women considering the use of MHT to avoid hot flashes. Breast cancer is the second leading cause of cancer death in women and the most commonly diagnosed cancer. The risk of a woman in the United States developing breast cancer over her lifetime is approximately 1 in 8. Most women have experienced breast cancer in their lives, either personally or through an afflicted relative or friend. It is thus important to address this prevalent concern and put patient perceived risks in perspective.

MENOPAUSAL HORMONE THERAPY IN CLINICAL TRIALS

Although there are many factors that influence a woman's risk of breast cancer, the role of MHT deserves special consideration. Although the absolute risk of breast cancer associated with use of MHT is quite small, a lack of appreciation of the distinction between absolute and relative risks has influenced both the public and prescribers.

In observational studies, inconsistent effects of estrogen alone or estrogen combined with a progestogen (P) were seen in postmenopausal women. In the largest observational study to date—the Million Women Study—it was found that estrogen plus P treatment increased postmenopausal women's risk of breast cancer; treatment with estrogen alone, as commonly undertaken in women who have had hysterectomies, increased this risk slightly, but far less than seen with combination therapy.[5] Current users of MHT were more likely than never users to develop breast cancer (adjusted relative risk [RR], 1.66; 95% CI, 1.58–1.75; $P<.0001$) and to die from it

(RR, 1.22; 95% CI, 1.00–1.48; $P = .05$). Past users of MHT were, however, not at an increased risk of disease.[5] The incidence of breast cancer was significantly increased with estrogen (RR, 1.30; 95% CI, 1.21–1.40; $P<.0001$) as well as with combination of estrogen plus P (RR, 2.00; 95% CI, 1.88–2.12; $P<.0001$).[5] In this study, the results did not vary significantly by the type of progestogen used or the route of administration. Time since menopause seemed to influence MHT-related breast cancer risk. Women starting MHT (whether estrogen alone or in combination with P) less than 5 years since menopause had a small increased risk of breast cancer (RR, 1.43 [95% CI, 1.36–1.49] in the estrogen alone group and 2.04 [95% CI, 1.97–2.12] in the EPT group) compared with women initiating MHT more than 5 years since menopause (RR, 1.05 [95% CI, 0.89–1.23] in the estrogen alone group and RR, 1.53 [95% CI, 1.38–1.69]). The risk of breast cancer declined to levels seen in never-users of MHT after cessation of treatment (RR, 1.00; 95% CI, 0.97–1.03).[6]

Similar to the Million Women Study, The Nurses' Health Study was a large, observational study conducted in the United States that linked long-term MHT use with breast cancer.[7] The risk of breast cancer was increased significantly among users of estrogen alone (RR, 1.32; 95% CI, 1.14–1.54) or of combination MHT (estrogen plus P; RR, 1.41; 95% CI, 1.15–1.74), compared with postmenopausal women who had never used MHT.[7]

The E3N French cohort, another observational study, found that risk of breast cancer was greatest among women using an estrogen in combination with synthetic progestogens, but not with use of natural progesterone.[8] Unlike the WHI ET arm, but similar to the Million Woman Study, the E3N French cohort study found an increased risk of breast cancer in estrogen alone users (the majority being estradiol (E2) rather than conjugated estrogen), although this risk was still lower compared with the substantially elevated risk seen with estrogen plus progestagen (ie, not including progesterone) treatment (RR, 1.29 [95% CI, 1.02–1.65] and RR, 1.69 [95% CI, 1.50- 1.91], respectively).[8]

Similarly, in the California Teachers Study, a prospective observational study, use of combination MHT with estrogen + P was associated with a greater increase in breast cancer risk (RR, 1.65; 95% CI, 1.48–1.84) when compared with estrogen use alone (RR, 1.17; 95% CI, 1.04–1.31), and when compared with those classified as never-hormone users (RR, 1). The type of estrogen and progestogen used among women were not specified.[9]

The Heart and Estrogen/Progestin Replacement Study (HERS) was a randomized, controlled study that assessed CEE plus MPA's effects on cardiovascular disease prevention.[10] In their follow-up observational study, HERS II, the effects of EPT on non-cardiovascular disease outcomes were also assessed. This follow-up study found no significantly increased risk of breast cancer, although the study was underpowered to evaluate this endpoint.[11]

Although observational studies assist in understanding relationships, a definitive conclusion as to cause and effect cannot be drawn from these studies. Thus, RCTs like the WHI Trials offer the best available insight into the role MHT plays in breast cancer risk.

Discrepancies in breast cancer outcomes, especially with ET use, between the WHI and observational trials may be owing to selection bias, confounding, or the type of hormones used. The use of CEE, a mixture of multiple estrogens—some with selective estrogen modulator (SERM)-like properties, in the WHI may have had distinct advantages to the breast. Last, because use of the combination of estrogen and P results in a greater risk of breast cancer than estrogen alone, further research has been aimed toward understanding the role that progestogens play in this risk. Data from RCTs are

deemed more reliable compared with those accrued from observational studies given a potential for bias introduced by measurable and nonmeasurable confounders inherent to any observational study design. Results of the large WHI hormone trials thus offer high-quality evidence of the effect of MHT on breast cancer risk.

MAMMARY GLAND BIOLOGY AND BREAST CANCER

During normal mammary gland development, both E2 and progesterone are responsible for enhancing cellular division and promoting lobular–alveolar breast development.[12] The mammary epithelium expresses both estrogen and progesterone receptors, and progesterone is needed for proper breast development and differentiation.[13] Similarly in the adult, during the luteal phase of the menstrual cycle and during pregnancy, progesterone facilitates breast cell proliferation, migration, and invasion.[14] In normal breast cells, proliferation occurs via paracrine interactions—because dividing epithelial cells do not contain estrogen and progesterone receptors—relying on growth factors secreted by adjacent stromal cells, which do contain sex hormone receptors.[14] In the progression from normal breast development to carcinoma, it is postulated that there is a transition from paracrine to autocrine signaling, because neoplastic cells express estrogen and progesterone receptors.[14]

Previous studies have demonstrated that estrogen plus progestogen results in increased breast proliferation compared with what is seen with estrogen treatment alone.[15] This suggests that the progestogen component itself may contribute to the carcinogenic effect of sex steroids.[16] Progestogens exert their intracellular effect by modulating the transcription of various target genes. As such, it is hypothesized that progestogens alter the normal signaling pathways facilitating normal proliferation; however, with increased proliferation and DNA replication comes an increased potential for new mutations and subsequent malignant transformation.[16] The exact pathway by which progestogens affect breast cancer cell proliferation/progression to breast cancer has not been characterized fully; however, studies on human breast cancer cells, as well as animal models, have contributed to a better understanding of the effects of progestogen in the mammary glands.

BREAST CANCER CELLS: STUDIES IN VITRO

Because both the WHI hormone trials, as well as most observational studies, have clearly demonstrated an increased risk of breast cancer with combined EPT, understanding the mechanism by which this may occur is critical. Studies using breast cancer cell culture systems have demonstrated that the effects of progestogens in vitro are influenced by progestogen type, dose, and duration of exposure, as well as cell culture conditions. Research using human breast cells has demonstrated that progestogens differentially affect breast cell proliferative activity.[17] A study by Courtin and colleagues[18] analyzed the effects of E2 alone, E2 plus progesterone (P4), and E2 plus MPA on cellular proliferation and apoptosis in breast cancer cells, as well as normal human breast cells. Treatment with E2 alone in all cell types resulted in increased cell proliferation. In normal human breast cells, the addition of P4 blocked the proliferative effect of E2, and also resulted in an increased number of apoptotic cells. When normal cells were treated with E2 plus MPA however, there was little effect on cellular proliferation, and the number of apoptotic cells was decreased. In MCF-7 and T47-D breast cancer cell lines, MPA did not induce cellular proliferation and neither MPA nor P4 affected apoptosis in these cells.[18] Microarray studies revealed induction of different sets of genes in hormonally treated cells, compared with control cells. E2 plus MPA modified genes in a distinctly different pattern from E2 plus P4.

Sweeney and colleagues[19] found that MPA combined with E2 stimulated proliferation in long-term estrogen-deprived MCF-7 (MCF-7:5C) cells, whereas E2 alone resulted in cell death.

The progesterone receptor exists as 2 isoforms (PR-A and PR-B) and is present in the female reproductive tract, mammary glands, brain, and in some immune cells.[20] In addition, some progestogens can bind to other steroid receptors, including the glucocorticoid receptor (GR), as well as the androgen receptor (AR).[20] Progestogens exert their effect via interactions of steroid receptors with growth factors, oncogenes, and estrogen metabolizing enzymes.[21] Changes in the ratio of PR-A to PR-B are thought to be involved in the development of breast cancer.[20] The aberrant effects of progestogens in breast cancer pathogenesis may also be mediated by binding to steroid receptors other than PR, such as GR and/or AR. Sweeney and colleagues[19] identified a potential role for GR in breast cancer pathogenesis by studying MPAs effect on breast cancer cells as compared with that of dexamethasone and norethindrone acetate. Like dexamethasone, MPA blocked E2-induced apoptosis, allowing for continued proliferation of these cells. In addition, like dexamethasone, MPA blocked E2-induced genes related to apoptosis. Although this is not the first study to demonstrate MPA's function as a glucocorticoid,[21,22] it differs from others in implying that the increased proliferation of human breast cancer cells is mediated through MPA binding to the GR.

A role for the AR as a mediator of progestogens' carcinogenic effect was assessed in a study using an ex vivo culture system.[23] Breast explant tissue from postmenopausal women was cultured and exposed to MPA at concentrations similar to those seen in women taking an MPA-containing formulation of EPT. Although the normal physiologic role of AR signaling results in inhibition of breast cell proliferation,[24] this study found that in postmenopausal women MPA blocked the normal signaling effect of AR, preventing AR from inhibiting epithelial cell growth.[23]

Further support for the role of progestogens in breast cancer comes from studies analyzing the effects of progestogen on estrogen-metabolizing enzymes in breast cancer cells. Using T47-D and MCF-7 cells, Xu and colleagues[25] demonstrated that E2 plus MPA increased the expression of estrogen-activating enzymes—namely, aromatase, 17 β-hydroxysteroid dehydrogenase type 1, and sulfatase—but did not increase expression of the estrogen inactivating enzymes, 17 β-hydroxysteroid dehydrogenase type 2 and sulfotransferase. The increase in cellular expression of estrogen-activating enzymes with E2 plus MPA was greater than that seen when cells were treated with E2 alone. Interestingly, the increase in estrogen-activating enzymes was not associated with an increase in cell proliferation, although there was an increase in estrogen levels. It is known, however, that locally increased estrogen levels are seen in the breast cancer cell environment, and this high-estrogen environment facilitates cancer cell growth.[25] Thus, it is postulated that MPA may exert its carcinogenic effect via induction of a local hyperestrogenic state, rather than directly through cell proliferation.

PROGESTOGEN REGIMENS

The regimen of progestogen administration has also been suggested to influence breast cancer risk.[26] Analysis of the effects of combined continuous, versus a combined sequential regimen of E2 plus MPA in vitro demonstrated that estrogen-activating enzymes were stimulated by the continuous regimen, but not the sequential regimen.[25] Lyytinen and colleagues[27] found that sequential progestin use resulted in a trend toward a smaller increase in relative risk of breast cancer compared with

continuous progestin use. Within the EPT arm of the WHI, more women in the treatment group reported breast pain. Although the association between breast pain and breast cancer is uncertain, more women who were ultimately diagnosed with breast cancer reported breast pain during EPT use,[28] and breast discomfort seems to be a marker of breast gland stimulation. An ancillary study of the Kronos Early Estrogen Prevention Study (KEEPs)—an RCT designed to assess route of estrogen administration on cardiovascular effects (with cyclic progesterone administered daily for 12 days)—found that breast pain did not differ between MHT and placebo groups. Whereas MPA was given continuously in the EPT WHI trial, progesterone was given cyclically in KEEPs, suggesting differential effects of progesterone regimen on breast cancer risk. However, one must exercise caution in extrapolating information solely from this ancillary study, because sample sizes were small; future studies will facilitate the understanding of the role progestogen type, and the timing of progestogen administration, play in influencing breast cancer risk.

BREAST CANCER IN ANIMAL MODELS

Studies in animal models have contributed further to our understanding of role of progestogens in breast cancer. The characteristic response of breast cells to progesterone is ductal side branching and alveolar budding.[14] Studies in ovariectomized mice have demonstrated that estrogen plus progestogen therapy results in significantly increased breast cell proliferation when compared with estrogen treatment alone.[29–31] It is well-known that mammary gland proliferation increases during the luteal phase, when progesterone levels are higher than the estrogen levels. In progesterone receptor knockout (PRKO) mice, estrogen plus progestogen treatment did not result in the lobular–alveolar changes characteristic of EPT, suggesting that PR signaling plays an important role in breast gland tumorigenesis.[32–35]

A randomized trial in adult, ovariectomized, female macaques was used to study the effects of progestogens on risk markers for breast cancer.[36] This primate model is ideal for studying hormonal effects on breast tissue, because they have greater than a 90% average genetic coding sequence identity to humans.[37,38] In addition, the steroid receptor response to sex hormone administration, and the development of neoplastic breast tissue in this model, is similar to what occurs in humans.[36] The postmenopausal animals received 1 of 4 treatment regimens, with doses reflecting commonly prescribed doses in MHT for postmenopausal women—placebo, E2 daily, E2 plus P4 daily, or E2 plus MPA daily. After 2 months of treatment, macaques given E2 plus MPA demonstrated a significant increase in proliferation of breast lobular and ductal cells compared with placebo; this proliferative activity was not seen with E2 plus P4 treatment.[36] There was also increased expression of proliferation markers Ki67 and cyclin B1 in the E2 plus MPA–treated monkeys, but not in the E2 plus P4 treatment group. In a follow-up study using this same animal model, Wood and colleagues[15] also demonstrated differences in gene expression profiles for a given progestogen treatment. Breast biopsies were collected after 2 months of treatment, and analyzed for differences in gene expression. Breast tissue exposed to E2 plus MPA demonstrated increased expression of genes in the ErbB proliferative pathway—epidermal growth factor (EGF) and transforming growth factor-α. Genes of the Jak/Stat signal transduction pathway, including c-MYC gene expression were also differentially expressed, with a 2.5-fold change in the E2 plus MPA treatment when compared with control ($P<.01$). cMYC induces signals for cell proliferation, and is known to be involved in tumorigenesis.[15] There were no effects on genes related to apoptosis (transforming growth factor-β pathway), or genes related to

estrogen receptor activity (Trefoil 1, stanniocalcin, cyclin D) seen in any group of treated animals. Thus, rather than directly enhancing the mediated response of ER to increase breast cell proliferation, MPA may instead act via modulation of growth factor pathways. E2 plus MPA enhanced the effect of E2 on ErbB pathway–related genes, providing further support for the role of MPA in promoting breast cell proliferation through growth factor signaling mechanisms.[15]

The EGF receptor is present in normal epithelial cells, and is overexpressed in more than one-half of breast cancers.[39] It is postulated that EGF receptor contributes to tumorigenesis not only by increasing cellular proliferation, but also by increasing angiogenesis and promoting cell survival. In vitro studies on breast cancer cells have also demonstrated that PR may result in EGF receptor activation, suggesting that progestogens exert their carcinogenic effect via PR-mediated regulation of downstream growth factor pathways.[40,41]

Using a human–mouse model system, Liang and colleagues[42] analyzed the effects of progestogens on xenograft tumors. BT-474 breast cancer cells, which expressed PR and mutant p53, grown on a Matrigel substrate were used to create the tumors. These cells were then injected into nude mice, which had been pretreated with E2 before cancer cell transfer. The study found that in the presence of E2 alone, xenograft tumors underwent growth initially, followed by tumor regression. However, with administration of either progesterone or MPA, tumor regrowth ensued. The mechanism by which this occurs was felt to be related to expression of vascular endothelial growth factor (VEGF), mediated by PR. Support for the role of PR was further confirmed by addition of the PR antagonist mifepristone, which inhibited the proliferative capacity of tumor cells. VEGF has been implicated previously in tumor growth.[30,43] VEGF is proangiogenic, and promotes endothelial cell survival and proliferation.[44,45] An increased expression of VEGF in tumors after administration of progestogens suggests a progestogen-dependent regulatory mechanism. In MCF-7 cells that do not express mutant p53, there is no induction of VEGF expression and no associated growth of tumors in response to progestogens, which supports the hypothesis that acquisition of mutations predispose to breast cancer, with progestogens potentially acting on cells with existing mutations.[46] Interestingly, and in support of the ET arm of the WHI, although estrogen was needed initially to facilitate tumor growth, it did not enhance tumor growth over time but instead resulted in tumor regression, again suggesting that it is the progestogen component itself that is responsible for inducing breast tumorigenesis in vivo.[46]

Although these studies provide insight into the mechanisms involved in breast cancer acquisition, there are several differences exist in mammary development in murine models compared with humans; thus, results from such studies must be interpreted with caution. As mentioned, given the marked similarities between Macaque and human gene coding sequences, this primate serves as a more reliable model for studying the effects of MHT on breast cancer acquisition and risk.

BREAST CANCER AND THE WOMEN'S HEALTH INITIATIVE: POSTINTERVENTION CLINICAL DATA

After the initial results of the WHI Hormone Trials, the significantly increased risk of invasive breast cancer with EPT use was affirmed on long-term follow-up after the intervention was stopped.[47] Both intervention and postintervention follow-up in the EPT arm of the WHI demonstrated an increased risk of breast cancer and breast cancer mortality.[3] The intervention phase of the EPT ended in 2002, after a median of 5.6 years, owing to increased breast cancer risk and an unfavorable risk–benefit ratio.[1]

Extended follow-up continued through 2010, with a median postintervention follow-up of 8.2 years. The significantly increased risk of breast cancer incidence seen in the intervention phase remained significantly increased during the postintervention phase (HR, 1.27, 95% CI, 0.91–1.78).[1] In a sensitivity analysis adjusting for adherence, a significant difference in the HR slopes for the 2 study phases was found. There was a trend toward mitigation of breast cancer risk in EPT users in the postintervention phase (HR, 1.26; 95% CI, 0.73–2.20) compared with a hazard ratio of 1.62 (95% CI, 1.10–2.39) in the intervention phase.[48] In a sensitivity analysis adjusting for continued EPT use during the postintervention phase; however, ongoing EPT use had a higher association with breast cancer than was seen with EPT use in the intervention phase of the clinical trial ($P<.001$). Breast cancer mortality was greater in the EPT group compared with the placebo group ($P = .049$).[3] The breast tumors diagnosed in the postintervention phase were also larger than those seen in the placebo group ($P = .03$); however, there was no difference in receptors status based on EPT use.[3,48]

Chlebowski and colleagues[49] analyzed the effect of EPT on the ability of mammography and breast biopsy to detect breast cancer in the WHI EPT. There were significantly more abnormal mammograms in the EPT group compared with the placebo group. Interestingly, the number of breast cancers diagnosed by biopsy in the hormone group was less than that in the placebo group, despite the fact that breast cancers were not only increased in the EPT group, but also more cancers were diagnosed at a higher stage and were more likely to be lymph node positive. Women who began EPT within 5 years of menopause were at greater risk of breast cancer compared with women who were greater than 5 years since menopause; however, the risk did not attain significance and did not substantiate the gap hypothesis, which states there is a time frame at which administration of HRT would be most protective/beneficial, whereas administration past this optimum time frame may result in more harmful effects.[50] Subgroup analyses demonstrated no significant relationship in EPT and breast cancer incidence with respect to age, body mass index, or the Gail breast cancer risk assessment tool score.[49] Postmenopausal women aged 50 to 59 years in the hormone group were also found to have a shorter time to first biopsy and, overall, more biopsies after 5 years of EPT compared with placebo. After discontinuation of study medications, abnormal mammograms persisted for women in the EPT group for 1 year; however, postintervention data demonstrated that thereafter there were no differences.[3]

The intervention and postintervention WHI data highlight several important points. First, the increased risk in breast cancer is significant, and although it decreases over time, the risk persists even after discontinuation of hormone therapy.[51] In addition, although the absolute risk of death after breast cancer diagnosis in the EPT users was 2 per 10,000 women, caution and extensive counseling for women is important when considering long-term EPT use for relief of menopausal symptoms.[51]

An unanswered question regarding breast cancer and EPT is this: if cessation of EPT results in fewer breast cancer cases, where do the cancers go? More than 5 years are needed for a new breast cancer to be clinically detected. The time frame of the WHI is too brief to account for a new breast cancer arising de novo, and subsequently resolving within 1 year of MHT termination. A more plausible explanation is that EPT acts on a preexisting, subclinical breast cancer, spurring the initial growth of precancerous cells, which then slow or regresses after cessation of hormone therapy.[26,48] This explanation is further supported by the WHI data, demonstrating that there were no more in situ lesions in the EPT users compared with the placebo group, nor were there more new in situ lesions found upon discontinuation of EPT.[49] New cancers were not forming in response to EPT therapy.

The breast cancer data from the ET arm of the WHI trials are distinct from the EPT arm. Not only was use of estrogen alone not associated with an increased risk in breast cancer, but in contrast with the EPT data, ET data even suggested an element of risk reduction with use of estrogen alone (RR, 0.80; 95% CI, 0.62–1.04).[52] Age-specific comparisons also demonstrated fewer invasive breast cancers in the ET group compared with placebo in all age groups.[52] The intervention phase of the ET trial ended in 2004, after a median of 7.2 years of follow-up. The postintervention phase began shortly after the intervention phase was stopped, and continued through 2010. The decreased risk of breast cancer in the CEE group reached statistical significance in the postintervention phase.[53] In the postintervention phase of the WHI, women with prior hysterectomy were followed for 11.8 years. Users of CEE for a median of 5.9 years had a significantly decreased risk of developing breast cancer (HR, 0.77; 95% CI, 0.62–0.95).[53] Tumor receptor status, size, and nodal status were not different between ET users and placebo. Unlike the EPT postintervention phase, there was a significantly lesser mortality risk from breast cancer in the ET group compared with placebo (HR, 0.37; 95% CI, 0.13–0.91).[53] Despite both trials having an increased number of mammograms in participants randomized to receive hormone therapy, those in the ET group did not have a significant increase in mammographic abnormalities compared with placebo (5.4% vs 5.1%; $P = .53$) and the diagnostic utility of mammograms was not compromised significantly.[54] The specificity and negative predictive value of mammograms between the ET group and placebo group were comparable, except during the first 2 years of estrogen use, where the diagnostic performance of mammography was inferior to that seen in the placebo group. Estrogen use for 2 years did increase breast density compared with placebo to some degree, but did not affect interpretation of mammograms.[54] As such, there was no delay in breast cancer diagnosis in the CEE group. Although more biopsies were needed in the ET group to find tumors, when compared with the placebo group, a diagnosis of breast cancer was made in 8.9% and 15.8% of biopsies in the ET and placebo groups, respectively ($P = .04$).[54]

The use of ET for more than 5 years was safe, decreased breast cancer, and was associated with lower breast cancer mortality; these findings are in stark contrast with EPT use, which had an associated increased risk of breast cancer, delay in diagnosis, and increased breast cancer mortality.[3,53] As previously discussed, the progestin component itself is highly implicated, because cessation of EPT was associated with a decline in breast cancer, and the breast cancer rates for the placebo groups in both arms of the WHI were the same.[47,53]

The choice of CEE in the ET arm of the WHI may explain the favorable effects seen on the breast. CEE contains a mixture of multiple estrogens, and each estrogen type not only preferentially binds the 2 estrogen receptors, but may also exert differential actions depending on the target tissue.[55,56] Whereas E2 is the well-characterized estrogen, less is known about the many estrogenic components of CEE.[56] Unlike E2, these other estrogens differ in their B-ring saturation and in their chemical moieties at the 17 position.[55] In a study assessing the activity of an estrogenic compound with similarities to several estrogens in CEE (NCI 122–17 β-methyl-17α-dihydroequilenin), it was found that NCI 122, as well as 2 other equine estrogens, were estrogen agonists that binds both ER-α and -β, but are less potent estrogens than E2.[55] Despite their lower potency, NCI 122 and equine estrogens are able to exert transcriptional changes distinct from E2, which are postulated to account for the positive effects seen in several tissue types.[57–60] Bhavnani and colleagues[61] analyzed the effects of 11 equine estrogens (in CEE preparations) on the transcriptional activity of ER-α and -β, and found that many of the equine estrogens preferentially bind ER-β. ER-β

activation can inhibit ER-α activity on cell proliferation.[62,63] This inhibition induced by equine estrogens may in part explain the decreased risk of breast cancer observed in the WHI ET study. Further support for beneficial SERM-like properties of CEE comes from work by Song and colleagues,[64] where the effects of CEE and E2 on breast cancer cells were compared. CEE and E2 were noted to have distinct effects on gene expression. Research by Berrodin and colleagues[65] also demonstrated that several estrogenic compounds in CEE act as partial estrogen agonists; thus, like SERMs, the differences in binding and downstream cell signaling may afford CEE with specific tissue manifestations that are unlike the purely stimulatory effects of E2.[64] Additional research identifying which equine estrogens exert more SERM-like properties is needed, because they cannot only be preferentially used in MHT, but perhaps may even be of benefit in the treatment of breast cancer.

TISSUE SELECTIVE ESTROGEN RECEPTOR COMPLEX AND BREAST CANCER

Whereas MHT is the most effective pharmacologic treatment for vasomotor symptoms, the risks that MHT imposes on cancers, including breast and endometrial cancer risk, cannot be ignored. Efficacious treatment options for addressing menopausal symptoms that do not inherently increase cancer risk are needed, and research and development efforts to identify such alternative options are ongoing. Recently, a SERM and CEE have been paired to form a tissue selective estrogen complex (TSEC). TSECs pair a SERM with an estrogen, ideally blending the effects of the SERM and estrogen in a way that is more favorable than treating with 1 form of therapy alone.[66–68] The first TSEC to be approved by the US Food and Drug Administration (FDA) pairs bazedoxifene (BZA), a SERM, with CEE. In the phase 3 Selective Estrogen Menopause and Response to Therapy (SMART) trials, enrolling postmenopausal women at risk for osteoporosis with a uterus, BZA/CEE improved hot flashes and prevented bone loss.[66,69] There were no associated adverse effects on the breast, uterus, or ovary. Importantly, given the anti-estrogen effects of BZA on the uterus, combined treatment with BZA and CEE does not require the addition of a progestin, thus likely reducing the probability of breast cancer risk that has been associated with EPT treatment.[68–70] Furthermore BZA neither stimulates breast cells nor increases mammographic breast density.[68,71,72] Data from the SMART trials were reassuring, with the incidence of breast cancer being comparable in the BZA/CEE and placebo groups, although these trials were underpowered to evaluate fully this end point.[72] Additional insight comes from an in vitro study assessing the effect of BZA/CEE on MCF-7 breast cancer cells.[64] BZA was found to block the effects of CEE on breast cancer cell proliferation, and blocked antiapoptotic effects of CEE. Furthermore, like tamoxifen and raloxifene, BZA used alone did not induce estrogen agonist effects on breast cancer cells.[64] Furthermore, in tamoxifen-resistant breast tumor xenografts, BZA was able to inhibit cancer cell growth.[73] BZA seems to have an effect of breast tissue that is similar or superior to other commonly used SERMs.

For menopausal women with a uterus, a TSEC offers theoretic advantage over traditional combined MHT because the risk of breast cancer is theoretically absent.

ALTERNATIVES TO TRADITIONAL MENOPAUSAL HORMONE THERAPY AND BREAST CANCER RISK

As their name implies, SERMs are compounds that are capable of exerting both agonist and antagonist effects on estrogen receptors, depending on their target tissue.[67,74] When bound to estrogen receptors in the breast, SERMs act as estrogen antagonists, inhibiting the stimulatory action of estrogen on breast tissue.[74] Tamoxifen

and raloxifene were initially classified as anti-estrogens, given their effects on breast tissue; however, they were subsequently renamed SERMs once they were found to exert estrogenlike effects in bone and the endometrium.[75,76] Tamoxifen is used as an adjuvant treatment for breast cancer. It also reduces invasive breast cancer risk in women with ductal carcinoma in situ, and women at high risk of breast cancer.[77] Raloxifene also reduces breast cancer incidence and ductal carcinoma in situ.[78,79] Raloxifene is approved for treating and preventing osteoporosis as well as preventing breast cancer in postmenopausal women.[80] Tamoxifen and raloxifene, unlike estrogen, do not improve vasomotor symptoms, but instead can even cause worsening of vasomotor symptoms.[81–83] Although early SERMs were not developed for the purpose of treating menopausal symptoms, newer SERMs, such as ospemifene, have been developed with the intent of treating specific menopausal symptoms. Although oral ospemifene is fairly effective for the treatment of the genitourinary syndrome of menopause, it can exacerbate hot flushes.[68,84,85] There are currently no clinical data on the effects of ospemifene on breast cancer risk.

SUMMARY

Treatment with the combination of estrogen and P results in a greater risk of breast cancer than placebo. In contrast, not only does treatment with estrogen alone not increase the risk of breast cancer, but estrogen alone use by women who have had a hysterectomy may even result in a decreased risk of breast cancer. The 2 prevailing theories to explain the increased risk with combination MHT focus on estrogen and progesterone acting in concert to increase breast cancer risk, versus progesterone alone having carcinogenic properties. Given the rapid development of clinically apparent tumors and the lack of new in situ lesions seen with MHT use, it is logical to surmise the carcinogenic effect of progesterone to a mechanism that involves hormone actions on preexisting, small lesions.[86,87] Variations in the various progestogens differing in their affinity for PR, GR, and AR, variability of progestogen component of different MHT regimens, and variability in the duration of progestogen use across MHT regimens may all modulate EPT's deleterious effects on breast tissue. Continued research using breast cancer cell culture systems and animal models will hopefully allow for an improved understanding of the interplay between estrogen and progestogens that predispose to adverse effects on breast tissue. The FDA-approved TSEC (BZA/conjugated estrogen) holds promise as a treatment option that will eliminate the increased risk of breast cancer by avoiding administration of a progestogen along with estrogen in symptomatic menopausal women with intact uteri. Caution over this hypothesized benefit is warranted until it is substantiated by data on the incidence of breast cancer in TSEC users.

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