

Conjugated Equine Estrogen and Risk of Benign Proliferative Breast Disease: A Randomized Controlled Trial

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- Background** Estrogens stimulate proliferation of breast epithelium and may therefore increase the risk of benign proliferative breast disease, a condition that is associated with increased risk of breast cancer. We tested the effect of conjugated equine estrogen (CEE) on risk of benign proliferative breast disease in the Women's Health Initiative (WHI) randomized controlled trial.
- Methods** In the WHI CEE trial, 10739 postmenopausal women were randomly assigned to 0.625 mg/d of CEE or to placebo. Baseline and annual breast examinations and mammograms were required. We identified women in the trial who reported breast biopsies that were free of cancer, obtained the associated histological sections, and subjected them to standardized central review. Cox proportional hazards models were used to estimate hazard ratios (HRs) and 95% confidence intervals (CIs). All statistical tests were two-sided.
- Results** A total of 232 incident cases of benign proliferative breast disease were ascertained during follow-up (average duration, 6.9 years), with 155 in the CEE group and 77 in the placebo group. Use of CEE was associated with a more than two-fold increase in the risk of benign proliferative breast disease (HR = 2.11, 95% CI = 1.58 to 2.81). For benign proliferative breast disease without atypia, the HR was 2.34 (95% CI = 1.71 to 3.20), whereas for atypical hyperplasia, it was 1.12 (95% CI = 0.53 to 2.40). Risk varied little by levels of baseline characteristics.
- Conclusion** Use of 0.625 mg/d of CEE was associated with a statistically significant increased risk of benign proliferative breast disease.

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Benign breast disease is a heterogeneous condition that includes many histological entities (1). It has been detected more frequently since the advent of mammographic screening (2). Currently in the United States, approximately 20% of women undergo a breast biopsy within a decade of starting annual screening, and most of these biopsies show no evidence of malignancy (3). Benign breast disease is important not only because it is relatively common but also because of the attendant economic and psychological consequences of a diagnosis of this condition (4) and because it may be associated with increased risk of subsequent breast cancer (2). Hence, efforts to understand its etiology are warranted, given that they might both lead to opportunities for prevention of the morbidity associated with the condition and provide insight into the etiology of breast cancer.

A substantial amount of experimental and epidemiological evidence supports a role for estrogens in the etiology of breast cancer (5-8). Estrogens stimulate the proliferation of breast epithelial cells, and they may also have direct genotoxic effects and induce aneuploidy (9). Epidemiological studies of endogenous estrogens have generally shown that they are associated with increased risk of subsequent breast cancer in postmenopausal women (10,11). Many epidemiological studies of exogenous estrogens have also

shown positive associations with risk of breast cancer, particularly among current or recent users (12,13). Furthermore, the selective estrogen receptor modulator tamoxifen has been shown to substantially reduce the risk of breast cancer in women at high risk (14). However, other epidemiological studies of exogenous estrogens have not shown an association with risk (15), and the Women's Health Initiative (WHI) randomized controlled trial of conjugated equine estrogen (CEE) did not show an increase in breast cancer risk after an average follow-up period of almost 7 years (16).

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CONTEXT AND CAVEATS

Prior knowledge

There is mixed evidence that associates estrogens with risk of benign proliferative breast disease, a condition that may lead to breast cancer. After an average follow-up of 7 years, the Women's Health Initiative (WHI) detected no increase in breast cancer risk among women taking 0.625 mg/d conjugated equine estrogen (CEE) in a randomized controlled trial of 10739 hysterectomized postmenopausal women.

Study design

Histological sections were studied from women from the WHI CEE trial who reported breast biopsies free of cancer. Incidence rates of benign proliferative breast disease in the CEE and placebo groups were compared.

Contribution

After nearly 7 years of follow-up, women who took CEE had approximately twice as much risk of benign proliferative breast disease as women who took a placebo.

Implications

Use of unopposed estrogens appears to increase risk of benign proliferative breast disease, but whether this would translate into an increase in breast cancer is not yet known.

Limitations

Trial participants were treated with only one estrogen regime and dose, and the trial was stopped early. Symptoms and signs induced by CEE might have increased the likelihood of suspected breast lesions and biopsy in the treatment arm.

There is also some support for a role for estrogens in the etiology of benign proliferative breast disease, a condition that is considered to possibly have malignant potential and to represent a lesion on the pathway to invasive breast cancer (17–19). The Breast Cancer Prevention Trial, a randomized controlled trial that showed that tamoxifen reduced the risk of invasive breast cancer by 50% in women with high breast cancer risk (20), also showed that in women aged 50 and older, tamoxifen was associated with an approximately 20% reduction in the risk of benign breast disease overall and a 40% reduction in the risk of hyperplasia, with or without atypia (21). However, that study was limited by the fact that the histological sections were not reviewed centrally. In addition to that trial, a cohort study observed an increase in risk of benign proliferative breast disease in association with relatively long-term use of hormone replacement therapy (primarily conjugated estrogens) (relative risk_{all years vs never} = 1.70, 95% confidence interval [CI] = 1.06 to 2.72) (22). Of two case-control studies, one yielded no evidence of an association between estrogen replacement therapy and risk of benign proliferative breast disease overall (23), whereas the other showed increased risk overall but no variation in risk by degree of epithelial atypia (24); however, the former was limited by its small sample size (23) and the latter by its non-population-based study design (24).

Organ response to the proliferative effects of a hormone may entail progression from normal growth to hyperplasia and ultimately to neoplasia (5). Given the proliferative effect of estrogens on breast epithelial cells, it is conceivable that exogenous

estrogens might increase breast cancer risk by first increasing the risk of putative breast cancer precursor lesions. Therefore, according to this model, in the early years of hormone use, it would be possible to observe an effect on the risk of the precursor lesions in the absence of an effect on the risk of breast cancer. We investigated this possibility by using the WHI trial of CEE to test the effect of CEE on risk of benign proliferative breast disease.

Methods

Study Population

The WHI CEE trial has been described in detail elsewhere (16). In brief, 10739 postmenopausal women who were aged 50–79 years at initial screening, had a prior hysterectomy, were likely to reside in the area for 3 years, and provided written informed consent were enrolled between 1993 and 1998 at 40 clinical centers. Exclusions were based on competing risk, safety (eg, prior breast cancer), and adherence and retention considerations. Before their baseline evaluation and random assignment, women using postmenopausal hormones at initial screening were required to have a 3-month washout period during which they did not use postmenopausal hormones. All participants had a baseline mammogram and clinical breast examination; abnormal findings suggestive of breast cancer required that breast cancer be ruled out before the participant entered the study. The study population for this study consisted of all women in the original CEE trial. The WHI and the ancillary study reported here (in which all 40 WHI clinical centers participated) were approved by institutional review boards at all participating institutions.

Study Regimens, Random Assignment, and Blinding

Participants were assigned randomly to receive either one daily tablet containing 0.625 mg of CEE (Premarin, Wyeth [formerly Wyeth Ayerst], St Davids, PA) or a matching placebo. Random assignment involved use of a randomized permuted block algorithm, stratified by clinic and age. All medication bottles had unique bar codes and computer-based selection to ensure blinded dispensing.

Data Collection

In the WHI CEE trial, comprehensive information on breast cancer risk factors was obtained at baseline by interview (for lifetime hormone use) and by self-report (for other covariates) using standardized questionnaires (25). The relevant variables included age, ethnicity, family history of breast cancer, body mass index, prior breast disease, age at menarche, age at first full-term pregnancy, parity, age at menopause, oral contraceptive use, postmenopausal hormone use, and mammographic screening history.

Follow-up

Participants were contacted by telephone 6 weeks after random assignment to assess symptoms and reinforce adherence. Follow-up contacts by telephone or clinic visit occurred every 6 months, with clinic visits required annually. Annual mammograms and clinical breast examinations were required; study medications were withheld if a mammogram was not performed or results could not be

verified, but participants continued to be followed. Study medication was discontinued for several reasons, including the development of breast cancer, deep vein thrombosis, or pulmonary embolism, and when non-study hormones were prescribed by personal health-care practitioners (16).

Ascertainment of Outcome

The outcome of interest for this study was histologically confirmed incident benign proliferative breast disease with or without atypia (see "Histology"). Clinical events, including breast cancers and breast biopsies for noncancerous lesions, were initially identified from self-administered questionnaires completed every 6 months. Breast cancers were confirmed by local and central adjudicators, who reviewed medical records and pathology reports and who were blinded both to treatment assignment and to symptoms due to study medications. For this study, women who reported breast biopsies that were free of cancer were identified and clinical centers were sent lists of potentially eligible subjects quarterly. Clinic staff contacted participants to obtain written informed consent to solicit the histological sections resulting from the biopsies. To investigate the possibility that breast biopsies were missed by using this approach, the charts of 100 randomly selected participants who did not report breast biopsy were reviewed at one center, and none was found to have unreported biopsies.

Histology

Hematoxylin- and eosin-stained histological sections were reviewed by the study pathologist (DLP), who was blinded to the randomization assignment. The benign lesions were classified using well-established criteria as nonproliferative lesions, proliferative lesions without atypia (further classified according to whether they were mild, moderate, or florid in extent), or atypical (ductal and/or lobular) hyperplasia (2,26,27).

Statistical Analysis

Incidence rates of benign proliferative breast disease in the CEE and placebo groups were compared based on the intention-to-treat principle using time-to-event analyses. The time to benign proliferative breast disease was defined as the number of days from the date of random assignment to the date of the first diagnostic biopsy after random assignment that showed benign proliferative breast disease. Follow-up terminated either when a subject was biopsied for benign proliferative breast disease or on February 29, 2004 (just before study participants were instructed to stop taking their study pills), whichever came first. Follow-up time was censored at the date of last documented contact, diagnosis of breast cancer, mastectomy, or death. Women who developed a nonproliferative benign breast lesion continued to be followed up because they remained at risk of developing a subsequent proliferative lesion. Event rates over time were summarized using cumulative hazard plots. The intervention effect was summarized using hazard ratios (HRs) and 95% confidence intervals estimated from Cox proportional hazards models (28), with stratification by age, prior breast biopsies, and random assignment to the WHI Dietary Modification and Calcium plus Vitamin D supplementation trials. Stratification was time dependent in the case of the Calcium Plus Vitamin D supplementation trial (25). The primary test of the association between CEE and risk

of benign proliferative breast disease entailed estimation of the CEE effect in the total study population. The association was then further evaluated in various sensitivity analyses that were designed to assess the robustness of the main result. Specifically, we assessed the impact on the hazard ratio for the CEE effect of 1) an "as treated" analysis (in which events were attributed to actual hormone use during the trial); 2) separate analyses that adjusted for cumulative duration of hormone therapy before trial commencement, frequency of protocol-mandated annual mammograms, and frequency of clinical breast examinations; 3) an analysis that stratified by mammogram results; and 4) separate analyses that excluded women who had used hormone therapy before trial commencement, excluded cases that arose during the first year of follow-up, and excluded women who had had a breast biopsy before enrollment. Interaction was investigated by including product terms between treatment assignment and indicator variables for the subsets of interest in Cox proportional hazards models stratified by age, prior breast biopsies, and randomization to the Dietary Modification and Calcium Plus Vitamin D trials and was assessed by testing the equality of the product term coefficients. The proportional hazards assumption, which was tested by fitting models containing a product term between the intervention and follow-up time and assessing the coefficient of the product term for statistical significance, was shown to not be violated. Annualized event rates were calculated for comparisons of absolute disease rates. All statistical tests were two-sided, and results were considered to be statistically significant when two-sided *P* values were .05 or less.

Results

At baseline, there was little difference between the CEE and placebo groups with respect to age, ethnicity, menstrual and reproductive history, use of postmenopausal hormones, family history of breast cancer, prior breast disease, and other factors (Table 1).

Data on follow-up, adherence, and unblinding have been reported elsewhere (16). In brief, at the termination of the trial, vital status was known for 10 176 subjects (94.8%). The remaining 563 subjects had withdrawn, had been lost to follow-up, or had not provided outcome information for more than 18 months; 5773 women (53.8%) had already stopped taking study medication, but dropout rates did not differ between randomization groups; by year 6, of those women still being followed, 264 of 4637 (5.7%) in the CEE group and 431 of 4754 (9.1%) in the placebo group had initiated hormone use through their own health-care clinician; and clinic gynecologists had been unblinded to treatment assignment for 100 women in the intervention group and 83 in the placebo group.

During follow-up (average duration, 6.9 years), we identified 656 potentially eligible biopsies. The eligibility of 15 biopsies could not be determined due to lack of consent, hospital refusal, or other reasons. Of the 641 biopsies confirmed to be eligible, consent for review was obtained for 639, and histological sections were obtained for 630 of these. Of the sections reviewed, seven were from biopsies that occurred before random assignment and 44 had no breast tissue; these 51 sections were excluded from further consideration. The 579 eligible sections that were reviewed were from 533 women. Of these women, 58 were excluded due to censoring

Table 1. Baseline characteristics of participants in Women's Health Initiative Conjugated Equine Estrogen trial*

Patient characteristic	CEE (n = 5310)	Placebo (n = 5429)
Age, yearst	63.60 (7.30)	63.60 (7.30)
Race/ethnicity, No. (%)		
White	4009 (76.50)	4075 (75.06)
Black	781 (14.71)	835 (15.38)
Hispanic	319 (6.01)	332 (6.12)
American Indian	41 (0.77)	34 (0.63)
Asian/Pacific Islander	86 (1.62)	78 (1.44)
Other	62 (1.17)	67 (1.23)
Unknown	12 (0.23)	8 (0.15)
Family history of breast cancer, No. (%)†	893 (16.82)	870 (16.03)
Gail model 5-y risk > 1.75, No. (%)	1561 (29.40)	1592 (29.32)
Body mass index‡§	30.10 (6.70)	30.20 (7.00)
Prior breast disease, No. (%)		
No	3894 (73.33)	3787 (69.76)
1 biopsy	680 (12.81)	748 (13.78)
2 biopsies	250 (4.71)	296 (5.46)
Unknown	486 (9.15)	598 (11.01)
Age (y) at menarche, No. (%)		
≤10	410 (7.72)	442 (8.14)
11–14	4336 (81.66)	4380 (80.68)
≥15	533 (10.04)	585 (10.78)
Unknown	31 (0.58)	22 (0.41)
Age (y) at first full-term pregnancy, No. (%)		
Never had term pregnancy	124 (2.34)	113 (2.08)
<20	1188 (22.37)	1229 (22.64)
20–29	2831 (53.31)	2906 (53.53)
≥30	209 (3.94)	260 (4.79)
Unknown	958 (18.04)	921 (16.96)
Parity, No. (%)		
Never	489 (9.21)	461 (8.49)
1	381 (7.18)	445 (8.20)
2	1072 (20.19)	1118 (20.59)
3	1161 (21.86)	1277 (23.52)
≥4	2165 (40.77)	2092 (38.53)
Unknown	42 (0.79)	36 (0.66)
Age (y) at natural menopause†	44.50 (7.70)	44.40 (7.70)
Oral contraceptive use		
Ever used, No. (%)	2053 (38.66)	2052 (37.80)
Duration of use, y†	4.80 (4.90)	4.90 (5.00)
Postmenopausal hormone use		
Estrogen alone		
Ever used, No. (%)	2438 (45.91)	2538 (46.75)
Duration of use, y†	6.80 (7.40)	6.60 (7.30)
Estrogen plus progestin		
Ever used, No. (%)	217 (4.09)	251 (4.62)
Duration of use, y†	4.50 (5.40)	4.60 (4.90)
Mammography screening within 2 y, No. (%)	3529 (66.46)	3691 (67.99)
Enrollment in WHI Dietary Modification trial, No. (%)		
All groups	3656 (68.85)	3691 (67.99)
Dietary modification group	615 (11.58)	670 (12.34)
Placebo group	1039 (19.57)	1068 (19.67)
Enrollment in WHI Calcium Plus Vitamin D supplementation trial, No. (%)		
All groups	2236 (42.11)	2327 (42.86)
Calcium plus vitamin D group	1531 (28.83)	1540 (28.37)
Control group	1543 (29.06)	1562 (28.77)

(Table continues)

Table 1 (continued).

Patient characteristic	CEE (n = 5310)	Placebo (n = 5429)
Total daily energy intake, kcal†	1624.6 (796.10)	1642.9 (837.20)
Total daily fat intake, g†	63.50 (39.90)	64.30 (42.00)
Total daily calcium intake (supplements plus diet), mg†	997.71 (669.29)	1012.36 (684.40)
Total daily vitamin D intake (supplements plus diet), IU†	7.94 (6.42)	8.00 (6.45)

* CEE = conjugated equine estrogen; WHI = Women's Health Initiative. Percentages may not add up to 100% because of rounding error.

† Mean (SD).

‡ First-degree female relative.

§ Calculated as weight in kilograms divided by the square of height in meters.

(so that the corresponding section was excluded from consideration), three had no pathological diagnosis, 240 had a nonproliferative lesion, and 232 had an incident benign proliferative lesion.

Of the 232 incident cases of benign proliferative breast disease that were ascertained overall, 155 (66.8%) occurred in the intervention group and 77 (33.2%) occurred in the placebo group. Use of CEE was associated with a statistically significant twofold increase in risk of benign proliferative breast disease (HR = 2.11, 95% CI = 1.58 to 2.81) (Table 2). The increase in risk was most pronounced for those women who had benign proliferative breast disease without atypia (HR = 2.34, 95% CI = 1.71 to 3.20) but was also evident for those who had either atypical hyperplasia or moderately extensive or florid proliferative disease without atypia (HR = 2.22, 95% CI = 1.59 to 3.10). Risk of atypical hyperplasia alone was not altered by the intervention, but analysis of this effect was based on only 18 cases in the intervention group and 15 in the placebo group (HR = 1.12, 95% CI = 0.53 to 2.40).

As shown in Figure 1, the Kaplan-Meier estimate of the cumulative hazard of benign proliferative breast disease (all types combined) indicated that the difference between the intervention and control groups began to appear about 1 year after random assignment and persisted through the end of follow-up. There was no statistically significant variation in the risk of benign proliferative breast disease by baseline level of demographic or dietary variables or of breast cancer risk factors (Table 3).

We examined the robustness of the CEE effect on risk of benign proliferative breast disease in several sensitivity analyses. In an "as treated" analysis, in which events were attributed to actual hormone use during the trial (allowing for a 6-month lag, so that if a woman on active CEE treatment stopped medication or a woman on placebo started taking CEE, events were attributed to that group only if they occurred more than 6 months after the switch), the HR was 1.90 (95% CI = 1.40 to 2.56). Adjustment for cumulative duration of use of hormone therapy before the commencement of the trial did not alter the estimate of effect (HR = 2.11, 95% CI = 1.58 to 2.81), whereas exclusion of those women who had used hormone therapy before random assignment reduced the point estimate somewhat, although the estimate of effect was still statistically significant (HR = 1.68, 95% CI = 1.14 to 2.48). The effect of CEE on risk of benign proliferative breast

Table 2. Risk of benign proliferative breast disease in association with use of conjugated equine estrogen, overall and by the presence or absence of atypia*

Type of benign proliferative breast disease	No. of cases (annualized %)		HR (95% CI)	P value†
	CEE (n = 5310)	Placebo (n = 5429)		
All types	155 (0.42)	77 (0.20)	2.11 (1.58 to 2.81)	<.001
Benign proliferative breast disease without atypia	137 (0.37)	62 (0.16)	2.34 (1.71 to 3.20)	<.001
Benign proliferative breast disease without atypia (moderately extensive or florid) or with atypia	120 (0.33)	56 (0.16)	2.22 (1.59 to 3.10)	<.001
Atypical hyperplasia	18 (0.05)	15 (0.04)	1.12 (0.53 to 2.40)	.763

* CEE = conjugated equine estrogen; HR = hazard ratio; CI = confidence interval.

† From proportional hazards model stratified by age, prior breast disease, and treatment assignment in the Dietary Modification and Calcium Plus Vitamin D supplement trials.

disease was changed little by exclusion of cases that arose during the first year of follow-up (HR = 2.19, 95% CI = 1.61 to 2.97) or by exclusion of women with previous breast biopsies (HR = 1.99, 95% CI = 1.41 to 2.82).

The frequency with which the protocol-mandated annual mammograms and clinical breast examinations were performed differed little between the CEE and control groups (29), and the effect of CEE was not changed by adjustment for the frequency of mammograms (HR = 2.08, 95% CI = 1.56 to 2.76) or clinical breast examinations (HR = 2.11, 95% CI = 1.58 to 2.81). Furthermore, the effect of the intervention did not differ between strata defined by those who had at least one abnormal mammogram result (either “suspicious” or “highly suggestive of malignancy”) during follow-up (HR = 2.27, 95% CI = 1.20 to 4.30) and those who did not have an abnormal mammogram (HR = 1.89, 95% CI = 1.31 to 2.72; $P_{\text{heterogeneity}} = .63$).

Discussion

The results of the randomized, double-blind, placebo-controlled trial reported here showed that during an average follow-up period of 6.9 years, use of 0.625 mg of CEE per day was associated with a statistically significant twofold increase in the risk of benign proliferative breast disease. This estimate was robust to various sensitivity analyses. Analyses that focused on subtypes of benign proliferative breast disease (defined by the presence or absence of atypia) indicated that the effect was observed primarily in relation to proliferative disease without atypia, but this conclusion is tempered by the fact that relatively few cases of atypical hyperplasia developed during the follow-up period. Risk varied little by baseline levels of covariates, including age, family history of breast cancer, body mass index, prior breast biopsies, and use of postmenopausal hormones before trial entry. These results, from a trial that employed serial mammography and clinical breast examinations, contemporary biopsy procedures, and centralized pathology review, provide the most reliable estimate to date of the effect of estrogen on the development of benign proliferative breast disease.

The prevailing hypothesis concerning the natural history of breast cancer is that benign proliferative breast disease without atypia, proliferative disease with atypia, and in situ cancer represent successive steps preceding the development of invasive breast carcinoma (17). In keeping with this hypothesis, women with benign proliferative breast disease have an increased risk of subse-

quent breast cancer (2). Although it is difficult to pinpoint the time of onset of putative precursor lesions, the limited evidence that is available suggests that a decade or so might be required for progression from benign proliferative breast disease to invasive breast cancer (30). Factoring in the time required for development of benign proliferative breast disease following the initiation of exposure to exogenous estrogens, it is conceivable that if there is a relationship between exogenous estrogen use and breast cancer risk that involves progression through benign proliferative breast disease, it would require more than the approximately 7 years of follow-up (on average) that was available at the termination of the WHI CEE trial, which therefore might account for the null finding of the latter with respect to risk of invasive breast cancer (16). Follow-up of the CEE trial participants is continuing, and it will be of interest to observe whether the effect estimate for CEE in relation to breast cancer risk changes in subsequent analyses based on longer durations of follow-up.

In contrast to the CEE trial, the WHI trial of estrogen plus progestin (for women with an intact uterus) showed that use of estrogen plus progestin was associated with both an increase in the risk of breast cancer (31) and of benign proliferative breast disease after an average of 5.2 years of follow-up (T. E. Rohan, MBBS, PhD, A. Negassa, PhD, R. T. Chlebowski, MD, PhD, N. Lasser, MD, PhD,

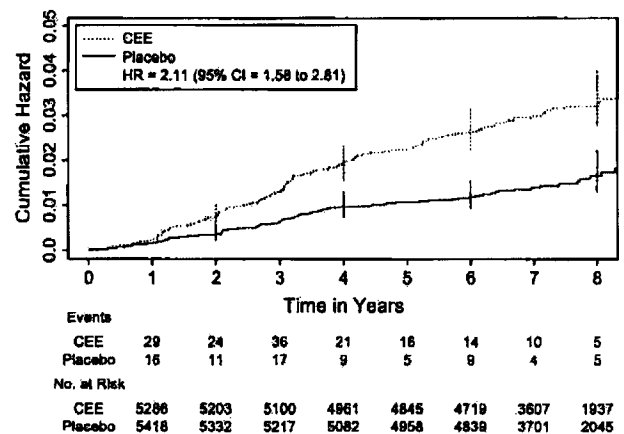


Figure 1. Kaplan-Meier estimates of the cumulative hazard of benign proliferative breast disease in association with conjugated equine estrogen (CEE), as compared with placebo (vertical lines are 95% confidence intervals [CI] at selected time points). HR = hazard ratio.

Table 3. Risk of benign proliferative breast disease in association with conjugated equine estrogen, by selected baseline characteristics*

Patient characteristic	No. cases of benign proliferative breast disease (annualized %) [‡]		HR (95% CI)	P value for interaction [†]
	CEE (n = 5310)	Placebo (n = 5429)		
Age, y				.14
50–59	49 (0.41)	31 (0.25)	1.48 (0.92 to 2.37)	
60–69	78 (0.48)	37 (0.22)	2.31 (1.52 to 3.50)	
70–79	28 (0.33)	9 (0.10)	3.60 (1.61 to 7.61)	
Race/ethnicity				.08
White	124 (0.45)	55 (0.19)	2.42 (1.73 to 3.40)	
Black	17 (0.32)	14 (0.24)	1.19 (0.57 to 2.48)	
Hispanic	4 (0.18)	5 (0.22)	0.68 (0.16 to 2.84)	
Other	9 (0.73)	2 (0.16)	6.50 (0.82 to 50.90)	
Family history of breast cancer in first-degree relative				.16
No	119 (0.42)	53 (0.18)	2.50 (1.77 to 3.53)	
Yes	30 (0.49)	18 (0.30)	1.51 (0.82 to 2.78)	
Gail model 5-y risk, %				.36
<1.25	51 (0.34)	33 (0.22)	1.63 (1.00 to 2.63)	
1.25–1.74	47 (0.42)	19 (0.16)	2.77 (1.59 to 4.83)	
≥1.75	57 (0.54)	25 (0.23)	2.16 (1.33 to 3.50)	
Body mass index[§]				.17
<25	35 (0.45)	9 (0.11)	4.71 (1.98 to 11.19)	
25–29	51 (0.41)	22 (0.16)	2.20 (1.32 to 3.64)	
30–34	39 (0.42)	28 (0.30)	1.54 (0.93 to 2.55)	
≥35	30 (0.43)	18 (0.25)	1.86 (0.97 to 3.57)	
Prior breast biopsy, No.				.84
None	96 (0.36)	47 (0.18)	1.99 (1.41 to 2.82)	
1	28 (0.62)	13 (0.26)	2.34 (1.23 to 4.45)	
≥2	15 (0.92)	7 (0.35)	2.49 (1.08 to 5.72)	
Age at menarche, y				.61
≤10	13 (0.46)	8 (0.26)	1.33 (0.52 to 3.39)	
11–14	126 (0.42)	61 (0.20)	2.19 (1.59 to 3.04)	
≥15	15 (0.41)	8 (0.20)	2.25 (0.91 to 5.53)	
Age at first full-term pregnancy, y				.51
Never had term	2 (0.24)	2 (0.25)	0.61 (0.06 to 6.33)	
<20	31 (0.38)	17 (0.20)	1.84 (0.98 to 3.44)	
≥20	100 (0.48)	50 (0.23)	2.23 (1.55 to 3.19)	
Parity				.80
0	12 (0.36)	6 (0.19)	1.68 (0.61 to 4.63)	
1	12 (0.45)	5 (0.16)	2.67 (1.00 to 8.20)	
2	33 (0.44)	19 (0.24)	1.65 (0.91 to 3.00)	
3	37 (0.47)	24 (0.27)	2.03 (1.15 to 3.59)	
≥4	61 (0.41)	23 (0.16)	2.52 (1.52 to 4.16)	
Age at natural menopause, y				.49
<48	85 (0.43)	38 (0.18)	2.32 (1.56 to 3.46)	
48 to <50	9 (0.55)	6 (0.32)	1.38 (0.48 to 3.96)	
50 to <53	19 (0.34)	13 (0.22)	1.64 (0.76 to 3.51)	
≥53	21 (0.57)	6 (0.17)	3.61 (1.37 to 9.50)	
Age at bilateral oophorectomy, y				.20
Never had	96 (0.47)	49 (0.24)	1.80 (1.33 to 2.45)	
<50	14 (0.40)	7 (0.18)	1.31 (0.49 to 3.50)	
≥50	14 (0.44)	4 (0.11)	5.23 (1.51 to 18.08)	
Oral contraceptive use, y				.59
None	85 (0.38)	39 (0.17)	2.18 (1.47 to 3.24)	
<5	41 (0.48)	15 (0.17)	2.50 (1.36 to 4.61)	
≥5	29 (0.50)	23 (0.38)	1.61 (0.88 to 2.94)	
Baseline postmenopausal hormone use, y				.19
Estrogen alone				
None	74 (0.37)	46 (0.23)	1.69 (1.15 to 2.48)	
<5	50 (0.55)	20 (0.21)	2.35 (1.37 to 4.03)	
≥5	31 (0.41)	11 (0.13)	3.64 (1.67 to 8.00)	

(Table continues)

Table 3 (continued).

Patient characteristic	No. cases of benign proliferative breast disease (annualized %) [‡]		HR (95% CI)	P value for interaction [†]
	CEE (n = 5310)	Placebo (n = 5429)		
Estrogen plus progestin				.73
None	149 (0.42)	71 (0.20)	2.14 (1.69 to 2.87)	
<5	4 (0.38)	3 (0.27)	3.22 (0.33 to 31.10)	
≥5	2 (0.38)	3 (0.44)	1.10 (0.17 to 7.01)	
Enrollment in WHI Calcium Plus Vitamin D trial				.46
No	59 (0.39)	26 (0.16)	2.47 (1.57 to 3.88)	
Active	53 (0.50)	25 (0.23)	2.20 (1.37 to 3.53)	
Placebo	43 (0.39)	26 (0.24)	1.65 (1.01 to 2.70)	
Enrollment in WHI Dietary Modification trial				.24
No	104 (0.42)	47 (0.18)	2.42 (1.67 to 3.50)	
Dietary modification group	19 (0.43)	13 (0.28)	1.19 (0.57 to 2.48)	
Control group	32 (0.44)	17 (0.22)	2.04 (1.11 to 3.74)	
Total daily energy intake, kcal				.40
<1106	42 (0.46)	16 (0.17)	3.01 (1.59 to 5.67)	
1106 to <1495	40 (0.46)	17 (0.18)	2.54 (1.39 to 4.63)	
1495 to <1988	34 (0.36)	23 (0.25)	1.66 (0.94 to 2.92)	
≥1988	39 (0.43)	21 (0.22)	1.64 (0.94 to 2.88)	
Total daily fat intake, g				.75
<37.0	39 (0.43)	19 (0.20)	2.50 (1.37 to 4.56)	
37.0 to <55.1	42 (0.46)	16 (0.17)	2.41 (1.38 to 4.32)	
55.1 to <80.3	36 (0.39)	19 (0.21)	2.02 (1.10 to 3.72)	
≥80.3	38 (0.43)	23 (0.24)	1.67 (0.97 to 2.87)	
Total daily calcium intake (supplements plus diet), mg				.98
<535.6	43 (0.47)	22 (0.24)	2.25 (1.27 to 3.97)	
535.6 to <848.4	36 (0.40)	19 (0.20)	2.20 (1.21 to 4.00)	
848.4 to <1304.1	40 (0.44)	20 (0.21)	1.93 (1.12 to 3.34)	
≥1304.1	36 (0.39)	16 (0.17)	2.09 (1.13 to 3.87)	
Total daily vitamin D intake (supplements plus diet), µg				.31
<2.8	35 (0.37)	22 (0.23)	1.77 (1.00 to 3.14)	
2.8 to <5.6	51 (0.57)	19 (0.21)	2.76 (1.57 to 4.86)	
5.6 to <12.6	39 (0.43)	16 (0.16)	2.76 (1.47 to 5.18)	
≥12.6	30 (0.34)	21 (0.23)	1.43 (0.79 to 2.58)	

* CEE = conjugated equine estrogen; HR = hazard ratio; CI = confidence interval; WHI = Women's Health Initiative.

† From proportional hazards model stratified by age, prior disease, and randomization group.

‡ For some variables, the number of events does not equal the total number shown in Table 2 due to missing values.

§ Calculated as weight in kilograms divided by the square of height in meters.

A. McTiernan, MD, PhD, R. Schenken, MD, M. Ginsberg, BA, S. Wassertheil-Smoller, PhD, D. L. Page, MD, unpublished data, 2008). Although the magnitude of the latter effect (HR = 1.74; 95% CI = 1.35 to 2.25) was slightly less than that observed here for CEE alone, the confidence intervals for the two estimates overlap substantially. Furthermore, the differences between the two study populations—in terms of their baseline characteristics, event rates, length of intervention, and follow-up time—dictate that caution be used in making direct comparison of these results (16). Nevertheless, the observation that both interventions increased the risk of benign proliferative breast disease whereas only estrogen plus progestin increased the risk of breast cancer is consistent with a possible role of progestins in breast cancer development. In addition, this observation raises the possibility that if the effect of CEE on the risk of benign proliferative breast disease is translated ultimately into an effect on breast cancer risk, it might result from the additional action of other predisposing factors.

Limitations of this trial have been discussed in detail elsewhere (16). In brief, the trial tested only one estrogen regimen and dose; the trial was stopped early, which may have decreased the precision of the effect estimates; a substantial proportion of study subjects discontinued their study medications; and there was a somewhat higher than expected drop-in rate in the placebo group. In relation to the latter two points, an "as treated" analysis, in which events were attributed to actual hormone use, showed that the effect was essentially unchanged.

With respect to this study, additional potential limitations include both differential ascertainment and misclassification of the outcome. Differential ascertainment of benign proliferative breast disease may have occurred because the symptoms and signs induced by CEE use may have increased the likelihood of detection of breast lesions and subsequent biopsy in the intervention group in particular (in both groups there may have been some underascertainment of outcome because biopsies were not

performed on all subjects). However, annual clinical breast examinations and mammograms were mandated by the study protocol, so that solid breast lumps and mammographically-detected nonpalpable lesions would have been equally likely to be detected and biopsied in the CEE and placebo groups. In this regard, compliance with the annual examinations was high and essentially the same in the two randomization groups, and adjustment for the frequency of mammograms and clinical breast examinations did not change the corresponding estimates of effect. Any misclassification of the outcome is likely to have been nondifferential and to have biased the effect estimates toward the null (32).

In conclusion, the results of the present randomized trial suggest that, during an average follow-up period of almost 7 years, use of CEE was associated with a doubling of the risk of benign proliferative breast disease. Longer follow-up of the trial participants may help to resolve the apparent contradiction between this finding and the initially null result for the effect of CEE on risk of invasive breast cancer.

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