RESEARCH ARTICLE

# Postmenopausal Hormone Therapy is Associated with in Situ Breast Cancer Risk

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## Abstract

**Background: The relationship between postmenopausal hormone therapy (HT) and invasive breast cancer has been extensively investigated, but that with breast carcinoma in situ (BCIS) has received relatively little attention. The aim of our present study was to review and summarize the evidence provided by longitudinal studies on the association between postmenopausal HT use and BCIS risk. Methods: A comprehensive literature search for articles published up to May 2012 was performed. Prior to performing a meta-analysis, the studies were evaluated for publication bias and heterogeneity. Relative risk (RR) or odds ratio (OR) values were calculated using 14 reports (8 case–control studies and 6 cohort studies), published between 1986 and 2012. Results: There was evidence of an association between ever postmenopausal estrogen use and BCIS based on a random-effects model (RR = 1.25, 95% confidence interval (CI) = 1.01, 1.55). However, we found no strong evidence of an association between ever postmenopausal estrogen combined with progesterone use and BCIS using a random-effects model (RR = 1.55, 95% CI = 0.95, 2.51). Furthermore, our analysis showed a strong association between "> 5 years duration" of estrogen or estrogen combined with progesterone use and BCIS. Furthermore, current use of any HT is associated with increased risk of BCIS in cohort studies. Additional well-designed large studies are now required to validate this association in different populations.**

**Keywords:** Postmenopausal hormone therapy - breast carcinoma in situ

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## Introduction

Breast carcinoma in situ (BCIS) is a non-invasive form of breast cancer that has not spread from the ducts or lobules into the surrounding breast tissue, is further categorized as either lobular CIS or ductal CIS (DCIS) depending on its location (Quinn et al., 1997). In addition, DCIS can be classified into comedo (high-grade) and non-comedo (medium or low-grade) subtypes based on histopathologic characteristics such as pattern of necrosis and maximum nuclear diameter. Both biological and epidemiologic evidence suggest that some DCIS develops into invasive disease, whereas other forms of DCIS may not progress to invasive breast cancer (IBC) (Mariuzzi et al., 1994; Page et al., 1995; Gupta et al., 1997; Stoll, 2000; Renshaw, 2002; Kopans et al., 2003). Incidence rates of BCIS have increased rapidly in the past few decades, on the order of 200% or more, largely due to the widespread use of mammograms (Ernster et al., 2002), Though we know that women with these lesions are far more likely to develop invasive breast cancer than women without in situ disease (Warnberg et al., 2000), our understanding of the natural history of BCIS remains limited, making it difficult to provide optimum treatment.

Evidence is emerging that several risks may contribute to both BCIS and invasive breast lesions including older age (Kerlikowske, 2010), family history of breast cancer (Trentham-Dietz et al., 2000; Claus et al., 2001), and higher breast density (Kerlikowske, 2010). Some endogenous hormonal factors associated with invasive breast cancer have also been associated with in situ disease (e.g., fewer full-term pregnancies, older age at first birth, older age at menopause) (Claus et al., 2001), while others have not (e.g., age at menarche, breastfeeding) (Kabat et al., 2011). For invasive breast cancer, postmenopausal HT is a well-established risk. More recent studies have refined the relationship between invasive breast cancer and HT and demonstrated greater risk with combined estrogen and progesterone formulations than with estrogen alone (Collins et al., 2005). There is also some evidence that among women using combination HT, continuous use of progesterone may put women at higher risk than sequential use (Lyytinen et al., 2009). However, with regard to in situ disease, information is much more limited.

Over the last two decades, a number of studies were conducted to investigate the association between

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postmenopausal HT and BCIS. But these studies reported conflicting results. In consideration of the extensive role of postmenopausal HT in the carcinogenesis of BCIS, we carried out a meta-analysis on all eligible case–control and cohort studies to estimate the overall BCIS of postmenopausal HT as well as to quantify the between-study heterogeneity and potential bias.

## Materials and Methods

*Retrieval of published studies*

To identify the studies of interest we conducted a computerized literature search. Sources included Pubmed, Web of Science, Medline and Embase. Search terms included: postmenopausal hormone therapy combined with breast cancer in situ, or breast carcinoma in situ. The titles and abstracts of the studies identified in the computerized search were scanned to exclude any studies that were clearly irrelevant. The full texts of the remaining articles were read to determine whether they contained information on the topic of interest. The reference lists of articles with information on the topic were reviewed to identify citations to other studies of the same topic. Reference lists of review articles were also inspected to determine relevant publications for completeness of our list of publications.

*Inclusion and exclusion criteria*

A study was included if it fulfilled the following criteria: (a) was designed as a cohort study, case-control study, or clinical trial; (b) evaluated exposure of hormone; and (c) had an outcome with BCIS incidence. Studies without raw data about exposure and measurements were excluded. In the subgroup analyses, studies that did not provide more detailed information about dose-response effects were eliminated. Studies were also excluded if they included subjects that were enrolled in other, more inclusive studies. In studies with multiple publications from the same population, only data from the most recent publication were included in the meta-analysis, with reference in the text to the older publications. Inclusion was not restricted by study size.

*Date Extraction*

Data were extracted by two independent reviewers using the same standardized form. Discrepancies were settled through additional reviews until a consensus was reached. Information obtained from each study included first author, year of publication, study design, types of hormones exposure, classification of hormone use and the number of subjects in the exposure groups, and RR/OR with 95% CI.

*Statistical Analysis*

Studies were grouped by the type of hormone (estrogen or estrogen combined progesterone). Two techniques were used to estimate the pooled relative risk estimates: the Mantel–Haenszel method (Mantel et al., 1959) assuming a fixed-effects model, and the DerSimonian–Laird method (DerSimonian et al., 1986) assuming a random-effects model. The fixed-effects model leads to valid inferences about the particular studies that have been assembled, and the random-effects model assumes that the particular study samples were drawn from a larger pool of potential studies and leads to inferences about all studies in the hypothetical population of studies. If heterogeneity is not present ($P<0.05$), the fixed-effects models may be biased. When heterogeneity is found ($P\leq0.05$), the random-effects models may be biased (Mantel et al., 1959; DerSimonian et al., 1986).

To evaluate whether the results of the studies were homogeneous, we used Cochran's Q-test. We also calculated the $I^2$ quantity (Higgins et al., 2003), which describes the percentage variation across studies that is due to heterogeneity rather than chance. I2 lies between 0% and 100%. A value of 0% indicates no observed heterogeneity and larger values indicate increasing heterogeneity (Higgins et al., 2003).

Publication bias was evaluated using the funnel graph, the Begg and Mazumdar adjusted rank correlation test (Begg et al., 1994), and the Egger regression asymmetry test (Egger et al., 1997). The Begg and Mazumdar test is a statistical analogue of the visual funnel graph. It determines whether there is a significant correlation between the effect estimates and their variances. The absence of significant correlation suggests that the studies have been selected in an unbiased manner. The Egger regression asymmetry test tends to indicate the presence of a publication bias more frequently than the Begg approach. It detects funnel plot asymmetry by determining whether the intercept deviates significantly from zero in a regression of the standardized-effect estimates against their precision (Bonovas et al., 2005).

Data were stratified into subgroups based on study design to examine the consistency across varying study designs with different potential biases. Homogeneity was assessed overall and within this stratification.

To assess any association between duration of estrogen or estrogen combined progesterone use and the risk of BCIS, we used the available data from studies in which the duration is > 5 years.

All P-values are two-tailed. For all tests, a probability level $< 0.05$ was considered statistically significant. STATA 11.0 software was used for the statistical analyses (STATA Corp., College Station, TX, USA).

## Results

*Search results*

Cohort, case-control and clinical studies of HT and BCIS are described in Table 1. We identified 8 cohort studies, 8 case-control studies, and 1 clinical trial reporting on HT related to BCIS risk. Two cohort studies adopted a standardized incidence ratio (SIR) to estimate RR, were eliminated. SIR is the ratio of observed to expected cases, based on reference incidence rates for the general population (Lyytinen et al., 2006, Lyytinen et al., 2009). One randomized controlled trial study adopted a hazard ratio (HR) to estimate RR, was eliminated (Chlebowski et al., 2003). We included total 14 studies, with 9,138 cases, in the meta-analysis (Table 1, Figure 1). Of the 14 studies, 8 estimates reported statistical significance, and

**Table 1. Studies Included in the Meta-analysis**

| Study, Year | Design | Ages | Exposure assessment | In situ cases (n) | All subjects | Exposure | Classification of hormone use | RR/OR(95% CI) | Adjustment |
|---|---|---|---|---|---|---|---|---|---|
| Brinton et al, 1986 | CC | NA | A | 254 | 4218 | E | Ever use | 1.26(0.9–1.6) | 1; 2; 3; 4; 5;6 |
| | | | | | | | Duration (<5 years) | 0.90(p>0.05) | |
| | | | | | | | Duration (5–9 years) | 1.52(p<0.05) | |
| | | | | | | | Duration (10+ years) | 1.90(p<0.05) | |
| Schairer et al, 1994 | Cohort | NA | B; F | 154 | 49,017 | E | Ever use | 1.4(1.0–2.0) | 2; 25 |
| | | | | | | | Duration (<5 years) | 1.1(0.7–1.7) | |
| | | | | | | | Duration (5–9 years) | 1.5(0.8–2.6) | |
| | | | | | | | Duration (10–14 years) | 2.1(1.2–3.7) | |
| | | | | | | | Duration (15–19 years) | 1.8(0.9–3.9) | |
| | | | | | | | Duration (≥20 years) | 2.0(0.9–4.5) | |
| | | | | | | E+P | Ever use | 2.3(1.3–3.9) | |
| | | | | | | | Duration (<2 years) | 3.3(1.7–6.3) | |
| | | | | | | | Duration (2–3 years) | 3.9(1.5–9.7) | |
| | | | | | | | Duration (≥4 years) | 0.7(0.1–4.7) | |
| Stanford et al, 1995 | CC | 50–64 | E | 87 | 1029 | E | Duration (1–3 months) | 1.8(0.5–6.9) | 2; 11; 13 |
| | | | | | | | Duration (4 m-2.9 y) | 0.8(0.3-2.4) | |
| | | | | | | | Duration (3–4.9 y) | 1.3(0.4–5.0) | |
| | | | | | | | Duration (≥5 y) | 1.0(0.5–2.0) | |
| | | | | | | E+P | Duration (1–3 months) | 1.7(0.3–8.9) | |
| | | | | | | | Duration (4 m-2.9 y) | 1.7(0.8–3.6) | |
| | | | | | | | Duration (3–4.9 y) | 0.9(0.3–3.3) | |
| | | | | | | | Duration (5–7.9 y) | 2.3(0.6–8.1) | |
| | | | | | | | Duration (≥8 y) | 0.5(0.1–4.2) | |
| Longnecker et al, 1996 | CC | ≤40 or 55–64 | A | 233 | 4493 | E | Ever use-W | 1.43(0.97–2.12) | 2; 8; 10; 11; 13; 14; 20; 21 |
| | | | | | | | Ever use-K | 1.60(1.00–2.58) | |
| | | | | | | | Current | 1.65(1.10–2.46) | |
| | | | | | | | Past use | 1.45(0.92–2.28) | |
| | | | | | | | Duration (<4 years) | 1.13(0.72–1.77) | |
| | | | | | | E+P | Duration (≥4 years) | 2.00(1.34–3.00) | |
| | | | | | | | Ever use-W | 1.75(1.10–2.80) | |
| | | | | | | | Ever use-K | 1.47(0.82–2.63) | |
| Henrich et al, 1998 | CC | 45+ | C | 32 | 654 | E or E + P | Use at age 45 years or older | 1.08(0.42–2.77) | 2; 11; 14; 15 |
| Gapster et al, 1999 | Cohort | 55–69 | B | 175 | 37,105 | Any HT | Past use (≤5 years) | 0.91(0.61–1.34) | 2; 5; 8; 9; 10; 11; 12; |
| | | | | | | | (>5 years) | 0.29(0.07–1.18) | 13 |
| | | | | | | | Current (≤5 years) | 0.94(0.41–2.16) | |
| | | | | | | | (>5 years) | 1.35(0.77–2.36) | |
| Ross et al, 2000 | CC | 55–72 | A | 186 | 3534 | Any HT | 5 years of use | 1.36(1.15, 1.61) | 5; 8; 10; 11; 12; 13; 21; 24 |
| | | | | | | E | 5 years of use | 1.41(1.18, 1.69) | |
| | | | | | | E+P | 5 years of use | 1.10(0.76, 1.60) | |
| | | | | | | E+P (continuous) | 5 years of use | 1.14(0.69, 1.88) | |
| | | | | | | E+P (sequential) | 5 years of use | 1.07(0.64, 1.79) | |
| Trentham-Dietz et al, 2000 | CC | 18–74 | F | 301 | 7788 | PostM | | 1.92(1.34–2.75) | 2; 11; 13; 14; 25 |
| | | | | | | E and/or P | BCIS | 1.90(1.24–2.92) | |
| | | | | | | | | 1.75(1.00–3.05) | |
| | | | | | | | | 2.53(1.18–5.42) | |
| | | | | | | | | 2.41(1.48–3.92) | |
| | | | | | | | | 1.91(1.04–3.50) | |
| | | | | | | | | 1.66(0.85–3.25) | |
| | | | | | | | | 1.53(0.84–2.80) | |
| | | | | | | | LCIS | 1.63(0.69–3.89) | |
| | | | | | | | | 2.46(0.95–6.40) | |
| | | | | | | | DCIS/non-LCIS | 2.03(1.24–3.34) | |
| | | | | | | | | 1.83(1.05-3.20) | |
| Kerlikowske et al, 2003 | Cohort | 50–79 | C | 583 | 374,465 | E+P | Duration (≥5 years) | 1.41(1.24–1.60) | 2; 11; 17; 18; 19 |
| | | | | | | | Duration (<5 years) | 0.77(0.62-0.96) | |
| | | | | | | E | Ever use | 0.98(0.89–1.07) | |
| Reeves et al, 2006 | Cohort | 50–64 | B | 1913 | 1,031,224 | Any HT | Past use | 1.15(1.01–1.31) | 1; 2; 8; 10; 11; 13; 14; 22 |
| | | | | | | | | 1.19(1.03–1.38) | |
| | | | | | | | | 0.96(0.45–2.07) | |
| | | | | | | | | 1.02(0.77–1.37) | |
| | | | | | | | Current use | 1.55(1.40–1.72) | |
| | | | | | | | | 1.56(1.38–1.75) | |
| | | | | | | | | 2.82(1.72–4.63) | |
| | | | | | | | | 1.35(1.07–1.70) | |
| Reinier et al, 2007 | Cohort | NA | C | 176 | 61,844 | Any postM | Current use (postmenopausal) | 1.1(0.8–1.5) | 2; 8; 11; 23 |
| Phillips et al, 2009 | CC | 20–74 | E | 304 | 4276 | Any HT | Ever use | 0.94(0.66–1.32) | 1; 2; 22 |
| Calvocoressi et al, 2012 | CC | | F | 998 | 1997 | Any HT | Current use (postmenopausal) | 0.87 (0.65, 1.18) | 2; 11; 13; 14 16; 26 |
| | | | | | | | Duration(<1 year) | 0.90 (0.57, 1.43) | |
| | | | | | | | Duration (1 to <5 years) | 0.79 (0.52, 1.18) | |
| | | | | | | | Duration (5 to <10 years) | 0.77 (0.46, 1.29) | |
| | | | | | | | Duration (≥10 years) | 0.93 (0.60, 1.43) | |
| | | | | | | E | Current use (postmenopausal) | 0.97 (0.66, 1.41) | |
| | | | | | | | Duration (<1 year) | 1.55 (0.84, 2.87) | |
| | | | | | | | Duration (1 to <5 years) | 0.72 (0.41, 1.25) | |
| | | | | | | | Duration (5 to <10 years) | 0.84 (0.41, 1.74) | |
| | | | | | | | Duration (≥10 years) | 0.93 (0.56, 1.55) | |
| | | | | | | E+P | Current use (postmenopausal) | 0.78 (0.52, 1.16) | |
| | | | | | | | Duration (<1 year) | 0.51 (0.26, 1.03) | |
| | | | | | | | Duration (1 to <5 years) | 0.85 (0.49, 1.48) | |
| | | | | | | | Duration (5 to <10 years) | 0.72 (0.36, 1.46) | |
| | | | | | | | Duration (≥10 years) | 0.93 (0.44, 1.98) | |
| Reeves et al, 2012 | Cohort | 50-64 | C | 3715 | 1.1 million | Any HT | Never | 1.00 (0.92, 1.09) | 27 |
| | | | | | | | Past | 1.14 (1.02, 1.28) | |
| | | | | | | | Current | 1.51 (1.39, 1.63) | |

CI: confidence interval; CC: case control; RCT: randomized controlled trial; PR: prospective; BCDDP: breast cancer detection and prevention program; PostM: postmenopausal; DCIS: ductal carcinoma in situ; LCIS: lobular carcinoma in situ; E: estrogen; P: progesterone; HT: hormone therapy; OR: odds ratio; HR: hazard ratio; RR: relative risk; SIR: standardized incidence ratio; W: all women; K: women with known age at menopause; OC: oral contraceptive; ETOH: alcohol use; BMI: body mass index; NA: not available.A: Home interview; B: Mailed questionnaire; C: Screening mammogram questionnaire; D: Finnish medical reimbursement register; E: In-person interviews; F: Telephone interview.1, Race; 2, age; 3, center; 4, BCDDP enrollment date and time in program; 5, type of menopause; 6, time since oophorectomy; 7, randomization assignment; 8, BMI; 9, waist-to-hip ratio; 10, parity; 11, family breast cancer history; 12, ETOH; 13, age at menarche, first birth and menopause; 14, mammography screening; 15, breast symptoms; 16, history of breast biopsy or hysterectomy; 17, exam year; 18, interval between screening mammograms; 19, mammography registry; 20, socioeconomic status; 21, benign breast disease; 22, deprivation index; 23, breast density; 24, OC use; 25, education; 26, number of pregnancies; 27, self-reported screening history

**Table 2. Meta Analysis of Hormone Use and Postmenopausal Breast Carcinoma in Situ Risk**

| | No. of | Fixed-effects model | | Random-effects model | | Tests of homogeneity | | | Tests of publication bias | |
|---|---|---|---|---|---|---|---|---|---|---|
| Estrogen use | studies | RR | (95% CI) | RR | (95% CI) | Q value (d.f.) | P-value | $I^2$ | Begg's P-value | Egger's P-value |
| Ever use | | | | | | | | | | |
| All | 7 | 1.07 | (0.99, 1.15) | 1.25 | (1.01, 1.55) | 19.5(6) | 0.003 | 69.40% | 0.36 | 0.03 |
| C-C studies | 5 | 1.34 | (1.13, 1.59) | 1.34 | (1.05, 1.70) | 7.11(4) | 0.13 | 43.80% | 0.8 | 0.48 |
| Cohort studies | 2 | 1 | (0.99, 1.15) | 1.12 | (0.80, 1.58) | 3.80(1) | 0.05 | 73.70% | 1 | NA |
| Duration > 5 years | | | | | | | | | | |
| All | 6 | 1.34 | (1.17, 1.54) | 1.34 | (1.17, 1.54) | 3.30(5) | 0.65 | 0% | 1 | 0.48 |
| C-C studies | 5 | 1.33 | (1.16, 1.54) | 1.33 | (1.16, 1.54) | 3.16(4) | 0.53 | 0% | 0.8 | 0.36 |
| Cohort studies | 1 | 1.5 | (0.83, 2.70) | 1.5 | (0.83, 2.70) | 0(0) | NA | NA | NA | NA |
| Estrogen combined with progesterone use | | | | | | | | | | |
| | No. of | Fixed-effects model | | Random-effects model | | Tests of homogeneity | | | Tests of publication bias | |
| | studies | RR | (95% CI) | RR | (95% CI) | Q value (d.f.) | P-value | $I^2$ | Begg's P-value | Egger's P-value |
| Ever use | | | | | | | | | | |
| All | 4 | 1.5 | (1.21, 1.86) | 1.55 | (0.95, 2.51) | 14.7(3) | 0.002 | 79.70% | 1 | 0.06 |
| C-C studies | 3 | 1.39 | (1.10, 1.75) | 1.38 | (0.77, 2.45) | 12.0(2) | 0.002 | 83.30% | 1 | 0.15 |
| Cohort studies | 1 | 2.3 | (1.33, 3.98) | 2.3 | (1.33, 3.98) | 0(0) | NA | NA | NA | NA |
| Duration > 5 years | | | | | | | | | | |
| All | 5 | 1.4 | (1.25, 1.56) | 1.37 | (1.07, 1.75) | 8.35(4) | 0.08 | 52.10% | 0.8 | 0.7 |
| C-C studies | 4 | 1.36 | (1.07, 1.72) | 1.32 | (0.86, 2.01) | 8.28(3) | 0.04 | 63.80% | 0.73 | 0.45 |
| Cohort studies | 1 | 1.41 | (1.24, 1.60) | 1.41 | (1.24, 1.60) | 0(0) | NA | NA | NA | NA |
| Ant HT | | | | | | | | | | |
| Current use | | | | | | | | | | |
| All | 6 | 1.47 | (1.38, 1.56) | 1.33 | (1.13, 1.56) | 18.0(5) | 0.003 | 72.30% | 0.45 | 0.29 |
| C-C studies | 2 | 1.09 | (0.85, 1.38) | 1.18 | (0.63, 2.21) | 6.27(1) | 0.01 | 84.10% | NA | NA |
| Cohort studies | 4 | 1.5 | (1.41, 1.59) | 1.46 | (1.31, 1.63) | 5.37(3) | 0.14 | 44.10% | 0.73 | 0.13 |

RR, Relative risk; CI, confidence interval; df, degrees of freedom; NA, not available; E: estrogen; P: progesterone

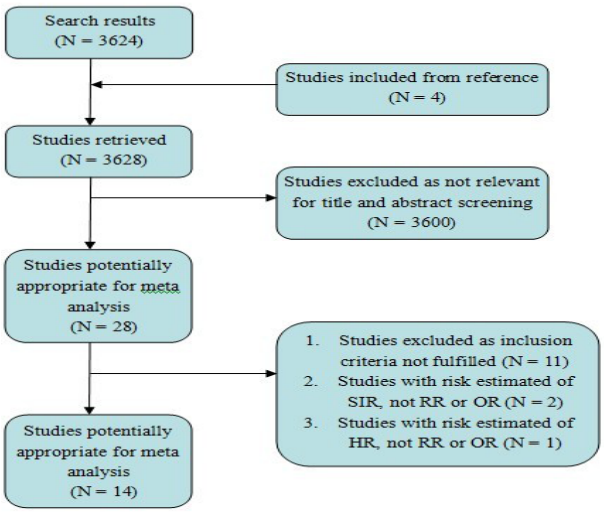


**Figure 1. Selection Process** (Abbreviations: SIR, standardized incidence ratio; RR, relative risk; OR, odds ratio; HR, hazard ratio)

5 studies reported a non-significant RR/OR that was ≥ 1. The ORs for hormone use in the 8 included case-control studies ranged from 0.5 to 2.53 (Table 1).

Thirteen studies evaluated exposure to estrogen and BCIS risk. Seven of the 14 evaluated the relationship between exposure to estrogen + progesterone and BCIS risk.

Fourteen studies (Brinton et al., 1986; Schairer et al., 1994; Stanford et al., 1995; Longnecker et al., 1996; Henrich et al., 1998; Gapstur et al., 1999; Ross et al., 2000; Trentham-Dietz et al., 2000; Kerlikowske et al., 2003; Reeves et al., 2006; Reinier et al., 2007; Phillips et al., 2009; Calvocoressi et al., 2012; Reeves et al., 2012) used newly diagnosed BCIS as a case definition and were controlled for potential confounding factors (at least for age), through matching or adjustments.

All case-control studies used non-cancer controls. Most studies were conducted in the USA (Brinton et al., 1986; Schairer et al., 1994; Stanford et al., 1995; Longnecker et al., 1996; Henrich et al., 1998; Gapstur et al., 1999; Ross et al., 2000; Trentham-Dietz et al., 2000; Kerlikowske et al., 2003; Reeves et al., 2006; Reinier et al., 2007; Phillips et al., 2009; Calvocoressi et al., 2012). Only one study was conducted in the UK (Reeves et al., 2012). The publication dates of the included studies ranged from 1986 to 2012. Study designs, along with the estimated relative risks and 95% CIs are shown in Table 1.

*Meta-analysis of exposure to estrogen*

5 case-control studies, 2 cohort studies evaluated ever use of estrogen and postmenopausal BCIS risk.

The funnel plot of ever use estrogen did not have the expected funnel shape. The underside corner of the pyramidal part of the funnel, which should contain small studies reporting negative or null results, was missing (Figure 2). The P-values for the Begg and Mazumdar test and the Egger test were P = 0.36 and P = 0.03, respectively, both suggesting a probability of publication bias. In contrast, Cochran's Q-test had a P-value of 0.003 (Q =

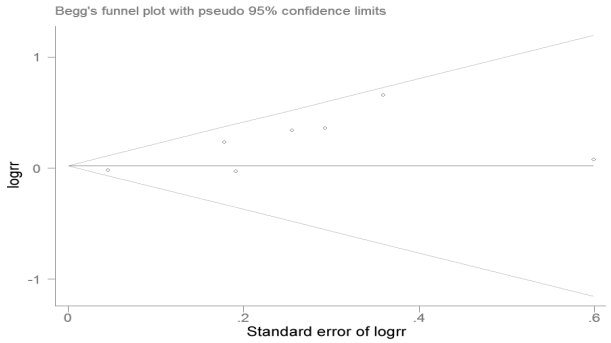


**Figure 2. Funnel Plots of the Relative Risk Between Ever Use of Estrogen and BCIS**, with the standard error, for all studies included in the meta-analysis. Relative risks are displayed on a logarithmic scale. The X axis represents standard error of logrr, and the Y axis represents logrr. For ever estrogen use: P = 0.368 for the Begg–Mazumdar test; P = 0.033 for the Egger test

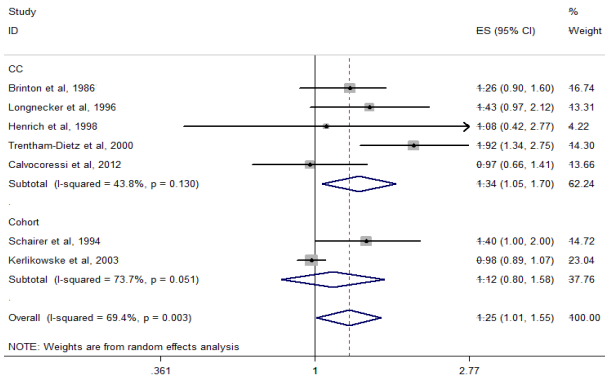


**Figure 3. Analysis of Studies, Listed by First Author and Publication Year that Examined BCIS and Its Association with Ever Estrogen Use.** The relative risk and 95% CI for each study are displayed on a logarithmic scale. Pooled estimates are from a random-effects model

19.5 on six degrees of freedom) and the quantity I2 was 69.4%, both indicating heterogeneity among the studies (Table 2).

The association of ever estrogen use with postmenopausal BCIS risk was statistically significant based on the random-effects models (RR = 1.25, 95% CI = 1.01, 1.55; n = 7) (Table 2).

After stratifying the data into subgroups based on study design, we found significant association between estrogen use and BCIS risk, in case–control studies (random-effects model, RR = 1.34, 95% CI = 1.05, 1.70; fixed-effects model, RR = 1.34, 95% CI = 1.13, 1.59; n = 5) but not in cohort studies (random-effects model, RR = 1.12, 95% CI = 0.80, 1.58; fixed-effects model, RR = 1.00, 95% CI = 0.99, 1.15; n = 2) (Table 2). Figure 3 graphs the RRs and 95% CIs from the individual studies and the pooled results.

To assess any association between duration of estrogen and postmenopausal BCIS risk, we used the available data from studies in which the duration > 5 years. The association between '> 5 years duration of estrogen use' with postmenopausal BCIS was statistically significant either based on a fixed-effects model (fixed-effects model, RR = 1.34, 95% CI = 1.17, 1.54, n = 6), or based on a random-effects model (RR = 1.34, 95% CI = 1.17, 1.54, n = 6) (Table 2).

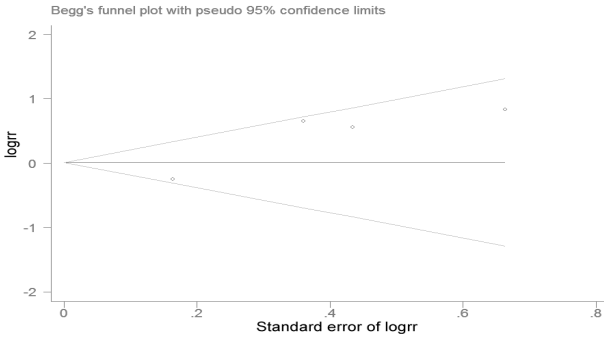


**Figure 4. Funnel Plots of the Relative Risk Between Ever Combination Use of Estrogen and Progesterone and BCIS, with the Standard Error, for All Studies Included in the Meta-analysis.** Relative risks are displayed on a logarithmic scale. The X axis represents standard error of logrr, and the Y axis represents logrr. For estrogen combined with progesterone use: P = 1.000 for the Begg–Mazumdar test; P = 0.064 for the Egger test

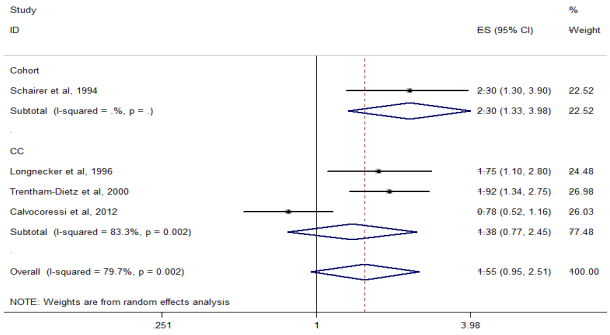


**Figure 5. Analysis of Studies, Listed by First Author and Publication Year that Examined BCIS and Its Association with Ever Estrogen Combined with Progesterone Use.** The relative risk and 95% CI for each study are displayed on a logarithmic scale. Pooled estimates are from a random-effects model

*Meta-analysis of exposure to estrogen combined with progesterone*

Three case-control studies and 1 cohort studies evaluated ever use of estrogen combined with progesterone and postmenopausal BCIS risk.

The funnel plot did not have the expected funnel shape. The underside corner of the pyramidal part of the funnel, which should contain small studies reporting negative or null results, was missing (Figure 4). The P-values for the Begg and Mazumdar test and the Egger test were P = 1.00 and P = 0.06, respectively, both suggesting a very low probability of publication bias. In contrast, Cochran's Q-test had a P-value of 0.002 (Q = 14.7 on three degrees of freedom) and the quantity $I^2$ was 79.7%, both indicating heterogeneity among the studies (Table 2).

The association between ever estrogens combined with progesterone and postmenopausal BCIS risk was statistically significant based on a fixed-effects model (RR = 1.50, 95% CI = 1.21, 1.86), but not statistically significant based on a random-effects model (RR = 1.55, 95% CI = 0.95, 2.51) (Table 2). However, the random-effects model is generally thought to be more appropriate, because it provides a more conservative estimate of the pooled effect size.

To evaluate the consistency across varying study designs with different potential biases, we stratified data

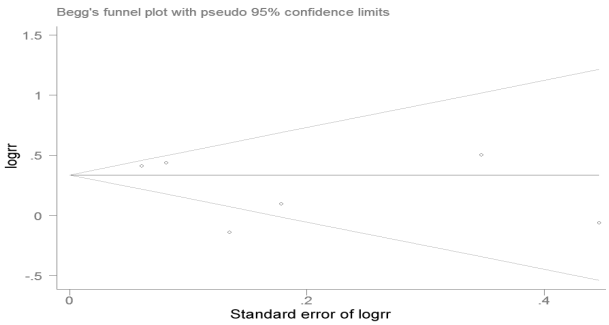


**Figure 6. Funnel Plots of the Relative Risk Between Current Use of Any HT and BCIS, with the Standard Error, for All Atudies Included in the Meta-analysis.** Relative risks are displayed on a logarithmic scale. The X axis represents standard error of logrr, and the Y axis represents logrr. For current use of any HT: P = 0.452 for the Begg–Mazumdar test; P = 0.299 for the Egger test

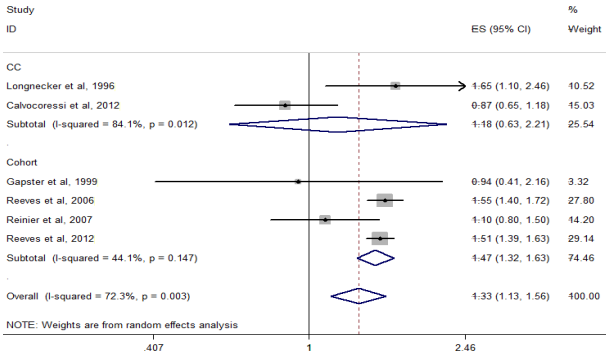


**Figure 7. Analysis of Studies, Listed by First Author and Publication Year that Examined BCIS and Its Association with Current Use of Any HT.** The relative risk and 95% CI for each study are displayed on a logarithmic scale. Pooled estimates are from a random-effects model

into subgroups based on study design. The association was not statistically significant among case-control studies (random-effects model, RR = 1.38, 95% CI = 0.77, 2.45; n= 3) (Table 2).

*Figure 5 illustrates the RRs and 95% CIs from the individual studies and the pooled results.*

To assess any association between duration of estrogen combined with progesterone and postmenopausal BCIS risk, we used the available data from studies with durations of more than 5 years. Five studies (Longnecker et al., 1996; Ross et al., 2000; Trentham-Dietz et al., 2000; Kerlikowske et al., 2003; Calvocoressi et al., 2012) were included in this analysis. The association between '> 5 years duration of estrogen combined with progesterone' with postmenopausal BCIS was statistically significant based on a fixed-effects model (fixed-effects model, RR = 1.40, 95% CI = 1.25, 1.56, n = 5), or based on a random-effects model (RR = 1.37, 95% CI = 1.07, 1.75, n = 5) (Table 2).

*Meta-analysis of exposure to any HT*

2 case-control studies, 4 cohort studies evaluated current use of any HT and postmenopausal BCIS risk.

The funnel plot of current use of any HT has the expected funnel shape (Figure 6). The P-values for the Begg and Mazumdar test and the Egger test were P = 0.45 and P = 0.22, respectively, both suggesting a probability of publication bias. In contrast, Cochran's Q-test had a P-value of 0.003 (Q = 18.0 on five degrees of freedom) and the quantity I2 was 72.3%, both indicating heterogeneity among the studies (Table 2).

The association of current use of any HT with postmenopausal BCIS risk was statistically significant based on the random-effects models (RR = 1.33, 95% CI = 1.13, 1.56; n = 6) (Table 2).

After stratifying the data into subgroups based on study design, we found significant association between current use of any HT and BCIS risk, in cohort studies (random-effects model, RR = 1.46, 95% CI = 1.31, 1.63; fixed-effects model, RR = 1.50, 95% CI = 1.41, 1.59; n = 4) but not in case–control studies (random-effects model, RR = 1.18, 95% CI = 0.63, 2.21; fixed-effects model, RR = 1.09, 95% CI = 0.85, 1.38; n = 2) (Table 2). Figure 7 graphs the RRs and 95% CIs from the individual studies and the pooled results.

## Discussion

We found that postmenopausal use of estrogen alone was associated with BCIS among ever users. In addition, "> 5 years duration" of estrogen alone, or progesterone combined with estrogen use was also associated with BCIS risk. And current use of any HT is associated with increased risk of BCIS in cohort studies.

Most other reports (Brinton et al., 1986; Schairer et al., 1994; Stanford et al., 1995; Longnecker et al., 1996; Henrich et al., 1998; Gapstur et al., 1999) have also described elevated BCIS risks associated with postmenopausal hormone use. The prevalence of postmenopausal hormone use has been increasing (Wysowski et al., 1995), and use of screening mammography has also been increasing since the 1980s (Breen et al., 1994). These two behaviors are highly correlated (Seeley, 1994). Furthermore, the effects of postmenopausal hormones on the density of breast tissue (Laya et al., 1996; Persson et al., 1997; Greendale et al., 1999) may increase the likelihood of biopsy and the serendipitous finding of BCIS, particularly of lobular BCIS. Thus, it is difficult to disentangle the independent effects of postmenopausal hormones on BCIS incidence.

Although the majority of treated BCIS cases perhaps will not subsequently develop to invasive cancer, ductal BCIS is generally recognized as the penultimate step in the progression of invasive tumors (Strah et al., 1992; Miller et al., 1993). Lobular BCIS is less likely to progress to invasive cancer, but it is considered a marker for significantly increased risk of invasive breast cancer (Strah et al., 1992).

The case–control studies that assessed the relationship between postmenopausal HT use and BCIS did adjust for multiple covariables, though they varied in which covariables were included, and in the precision of covariable measurement. Four of the seven case-control studies showed modestly elevated risk with estrogen and/or progesterone use in some analyses (Brinton et al., 1986; Longnecker et al., 1996; Ross et al., 2000; Trentham-Dietz et al., 2000), usually stronger with current use or longer duration, though one of these studies found decreasing risk

with longer use (Trentham-Dietz et al., 2000). Of these four studies, one adjusted for number of mammograms, one included a less precise measure of screening (ever use) (Trentham-Dietz et al., 2000), one did not appear to have adjusted for screening (Ross et al., 2000), and one included participants in the Breast Cancer Detection Demonstration Project and did account for time in the program (Brinton et al., 1986). Three case–control studies had negative findings, though two of these studies included very few subjects with in situ disease (Stanford et al., 1995; Henrich et al., 1998).

Across studies, there were also differences in the outcomes investigated. The majority reported on non-specific BCIS (Brinton et al., 1986; Schairer et al., 1994; Stanford et al., 1995; Henrich et al., 1998; Ross et al., 2000; Chlebowski et al., 2003; Lyytinen et al., 2006; Reinier et al., 2007; Lyytinen et al., 2009), whereas others reported on DCIS (Gapstur et al., 1999; Kerlikowske et al., 2003; Reeves et al., 2006; Phillips et al., 2009; Reeves et al., 2012) or LCIS (Trentham-Dietz et al., 2000; Reeves et al., 2006) separately. Because there is some evidence that HT may be more strongly associated with lobular than ductal lesions in invasive disease (Daling et al., 2002; Biglia et al., 2005; Phipps et al., 2010), and in situ cancers (Reeves et al., 2006), reporting on BCIS without examining specific histology may have missed important distinctions. A recent study by Phillips and colleagues (Phillips et al., 2009) is, to our knowledge, the first to examine HT use in relation to comedo and non-comedo DCIS, in addition to examining the effect on DCIS overall (shown in Table 1) and on invasive breast cancer. Among the subset of postmenopausal women in that study, the impact of HT on DCIS did not differ by DCIS subtype, but numbers of women in each group were small. Additional factors to consider in the assessment of BCIS outcomes include the expression of hormone receptors and other biomarkers. We do know that in situ tumors express receptors for estrogen and progesterone (Lari et al., 2011), but to our knowledge, the assessment of HT in relation to BCIS by hormone receptor status and other common markers has yet to be undertaken. Breast cancers may include distinct entities that can be differentiated based on specific tumor characteristics, including hormone receptor status, and HT may differentially affect the development of these tumors (Chen et al., 2004). Combining, for example, estrogen receptor (ER) positive and ER negative DCIS tumors as one outcome could, potentially, obscure a significant association if HT contributes primarily to the development of DCIS that is ER positive.

Several limitations should be considered in interpreting the results of this meta-analysis. First, most of the studies had a very small sample size and did not have adequate power to detect the possible risk for hormone use and postmenopausal BCIS risk, and the observed significant ORs in some studies of small sample size may be false association. Therefore, larger, well-designed should be performed to further confirm all these results.

Second, our search was restricted to studies published in indexed journals. We did not search for unpublished studies or for original data. However, we did not impose any exclusion criteria regarding language, place of publication or quality.

Third, the included studies were different in terms of study design and definitions of hormone exposure. We tried to explore sources of heterogeneity conducting several subgroup analyses. However, the summary effect estimates are based on sparse and heterogeneous data. Furthermore, because the HT induced BCIS risk is known to be different by the histological type, more articles are needed to conduct the subgroup analysis by histological type of BCIS.

Fourth, the methods used to elicit the exposure differ among the individual studies. Most studies used home interview (Brinton et al., 1986; Longnecker et al., 1996; Ross et al., 2000) or in-person interviews (Stanford et al., 1995; Phillips et al., 2009) or telephone interviews (Schairer et al., 1994; Trentham-Dietz et al., 2000; Calvocoressi et al., 2012) or mailed questionnaires (Schairer et al., 1994; Gapstur et al., 1999; Reeves et al., 2006) that rely on the subject's ability to recall, which has repeatedly been shown to be relatively poor for hormone use. Fewer studies (Henrich et al., 1998; Kerlikowske et al., 2003; Reinier et al., 2007) used Screening mammogram questionnaire that provide detailed information on dates of use and types of drugs used. Because the information is recorded prospectively, it is equally good for cases and controls irrespective of the event of interest.

Fifth, the dose-response relationship was evaluated based on "> 5 years duration" intake, which is not very precise and may not be indicative of the lack of dose dependency. Therefore, our results should be interpreted with caution. Besides these, all the included studies are in USA population.

Despite the limitations listed above, our present meta-analysis also had some advantages. First, substantial number of cases and controls were pooled from different studies, which greatly increased statistical power of the analysis. Second, the quality of case–control studies included in this meta-analysis was satisfactory according to our selection criteria. Our analysis shows for the first time that postmenopausal use of estrogen alone was associated with BCIS among ever users. In addition, "> 5 years duration" of estrogen alone, or progesterone combined with estrogen use was also associated with BCIS risk. More precise evaluation of postmenopausal hormone use and BCIS will depend upon larger study populations.

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