




# Chemoprevention Agents to Reduce Mammographic Breast Density in Premenopausal Women: A Systematic Review of Clinical Trials

Ana S. Salazar , MD, MPHS,<sup>1</sup> Malika Rakhmankulova, MD, MPHS,<sup>1</sup> Laura E. Simon , MLIS,<sup>2</sup> Adetunji T. Toriola , MD, PhD, MPH<sup>1,\*</sup>

<sup>1</sup>Department of Surgery, Division of Public Health Sciences, Washington University School of Medicine, St Louis, MO, USA and <sup>2</sup>Bernard Becker Medical Library, Washington University School of Medicine, St Louis, MO, USA

\*Correspondence to: A. T. Toriola, MD, PhD, MPH, Department of Surgery, Division of Public Health Sciences, Washington University in St. Louis, 660 S. Euclid Ave, Campus Box 8100, St Louis, MO 63110-0193, USA (e-mail: a.toriola@wustl.edu).

## Abstract

**Background:** Higher mammographic breast density (MBD) is associated with an increased risk of breast cancer when compared with lower MBD, especially in premenopausal women. However, little is known about the effectiveness of chemoprevention agents in reducing MBD in premenopausal women without a history of breast cancer. Findings from this review should provide insight on how to target MBD in breast cancer prevention in premenopausal women with dense breasts. **Methods:** We searched 9 electronic databases for clinical trials in English, Spanish, French, or German published until January 2020. Articles evaluating the association of pharmacological agents and MBD were included. Data were extracted on methods, type and dose of intervention, outcomes, side effects, and follow up. Quality of the studies was assessed using the US Preventive Services Task Force criteria. **Results:** We identified 7 clinical trials evaluating the associations of 6 chemoprevention agents with changes in MBD in premenopausal women without history of breast cancer. The studies evaluated selective estrogen-receptor modulators ( $n = 1$ ); gonadotropin-releasing hormone agonists ( $n = 2$ ); isoflavones ( $n = 1$ ); vitamin D ( $n = 1$ ); and Boswellia, betaine, and mayo-inositol compound ( $n = 1$ ). Hormonal interventions were associated with net reductions in percent density (tamoxifen [13.4%], leuprolide acetate [8.9%], and goserelin [2.7%]), whereas nonhormonal (vitamin D and isoflavone) interventions were not. However, MBD returned to preintervention baseline levels after cessation of gonadotropin-releasing hormone agonists. **Conclusions:** A limited number of chemoprevention agents have been shown to reduce MBD in premenopausal women. Identification of new and well-tolerated chemoprevention agents targeting MBD and larger studies to confirm agents that have been studied in small trials are urgent priorities for primary breast cancer prevention in premenopausal women with dense breasts.

In the United States, the incidence of breast cancer in younger women is increasing (1). About 15% of breast cancer cases are diagnosed in women younger than 40 years, and they tend to be larger, more aggressive, and with worse prognosis than in older women (1-4). Therefore, identifying intermediate and modifiable endpoints in breast carcinogenesis is critical for breast cancer prevention. However, interventions to reduce breast cancer risks have been predominantly studied among older women.

Mammographic breast density (MBD), which reflects the amount of epithelial and stromal tissues in relation to adipose tissue in the breast, is a strong but modifiable risk factor for breast cancer (5-7). Women with high MBD (>75%) have a 5-fold increased risk of breast cancer than women with little or no dense breast tissue (8,9). This is relevant for premenopausal

women because up to 39% of breast cancer cases are attributable to having dense breasts in this group (10-14). Studies have shown that a decrease in MBD ( $\geq 10\%$ ) in high-risk women is associated with lower breast cancer risk (63%) (15-17). Hence, MBD could serve as a surrogate marker of breast cancer development (13-16) and as a measurable endpoint of treatment response for preventive interventions (18,19), although it is worth noting that breast cancer arises from other biological pathways unrelated to, and completely independent of, MBD.

Although some pharmacological agents have been evaluated as potential breast cancer chemoprevention agents (20), only a few studies have investigated the associations of these agents with changes in MBD in premenopausal women. Therefore, the aim of this review is to summarize data from clinical trials that

Received: 23 June 2020; Revised: 5 October 2020; Accepted: 7 December 2020

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**Table 1.** Description of the PICO strategy<sup>a</sup>

PICO criteria	Description
Population	<p>Premenopausal women without a history of invasive breast cancer. We deferred to the author's definition of premenopausal status.</p> <p>Studies including both pre- and postmenopausal women were also included, as long as the effect of the intervention on MBD among premenopausal women could be extracted from the study.</p> <p>Studies that included women with history of noninvasive lesions, such as DCIS, LCIS, or AH were also considered.</p>
Intervention	Any pharmacological agent hypothesized to reduce MBD, reported by dose and duration of intervention.
Comparison	Any.
Outcome	Change in MBD reported as extractable data.

<sup>a</sup>AH = atypical hyperplasia; DCIS = ductal carcinoma in situ; LCIS = lobular carcinoma in situ; MBD = mammographic breast density; PICO = population, intervention, criteria, outcome.

have investigated the impact of chemoprevention agents on MBD in premenopausal women without a history of breast cancer. Study findings will provide evidence-based information to physicians managing premenopausal women with dense breasts and could contribute to primary breast cancer prevention.

## Methods

### Search Strategy

We followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses guidelines for conducting and reporting systematic reviews (21). An experienced medical librarian (LS) searched Ovid Medline, Embase, Scopus, Cochrane Database of Systematic Reviews, Cochrane Central Register of Controlled Trials, Health Technology Assessment Database, NHS Economic Evaluation Database, Clinicaltrials.gov, and World Health Organization International Clinical Trials Registry Platform for literature on MBD in premenopausal women published through January 22, 2020. An example of the search strategy can be found in the [Supplementary Table 1](#) (available online). We also reviewed the reference lists of all included studies for additional publications.

### Study Selection

Two reviewers (AS and MR) independently screened the titles and abstracts of interventional studies in English, Spanish, French, and German and selected eligible studies for subsequent full-text review. Studies were selected based on the population, intervention, criteria, outcome criteria, which can be found in [Table 1](#). Studies including mixed populations of pre- and postmenopausal women were also included, as long as the effect of the intervention on MBD among premenopausal women could be extracted from the study.

Interventional, as opposed to observational, studies allow for the evaluation of the direct impact of the intervention on the outcome. We excluded observational studies and conference abstracts, studies that did not report outcomes by menopausal status, and studies in which the intervention agents

were multivitamins. We also excluded studies without a comparison group (22). Disagreements were resolved by discussion. The inclusion and exclusion criteria can be found in the [Supplementary Table 2](#) (available online).

### Data Extraction and Quality Assessment

Two reviewers (AS and MR) extracted details on study title, year, design, experimental years, eligibility criteria, population characteristics (age, race, and menopause status), type and dose of chemoprevention agents, the associations between the agents and the outcome of interest, side effects, and duration of follow-up.

The quality of the studies was assessed based on the US Preventive Services Task Force (USPSTF) (23) criteria for the assessment of internal validity in clinical trials ([Supplementary Methods](#), available online). A study was considered "good" if it met all criteria, "fair" if it did not seem to meet at least 1 criterion but had no "major flaws," and "poor" if it had at least 1 major flaw ([Supplementary Table 3](#), available online).

## Results

### Study Selection

Database searches were executed on July 17, 2019, and again on January 22, 2020, resulting in a total of 3485 citations, which were identified and exported to Endnote. After the removal of 1958 duplicate records using a standard deduplication process (24), 1527 unique citations were left for the title and abstract screening. All articles were in English. Of the 34 articles that underwent full article review, 7 met the inclusion criteria ([Figure 1](#)). Review of the reference lists of selected studies generated no additional results. Because of the heterogeneity of the studies and the limited data for each chemoprevention agent, we performed a systematic review rather than a meta-analysis.

### Study Characteristics and Quality of the Evidence

Seven studies met the inclusion criteria: 5 randomized placebo-controlled trials (15,25-28) and 2 randomized no-drug controlled trials ([Table 2](#)) (29,30). The majority of the studies were performed in the late 1990s and early 2000s and took place in Canada (26), Italy (27), the United Kingdom (15,28,30), and the United States (25,29).

The study populations ranged from 19 to 400 women in their late 30s and early 40s ([Table 3](#)). Most women were non-Hispanic White. The mean body mass index (BMI) of women enrolled in the trials was less than 25 kg/m<sup>2</sup>. Three studies included women at increased risk for breast cancer (15,27,29), defined by any of the following criteria: 1) history of ductal carcinoma in situ, lobular carcinoma in situ, and atypical hyperplasia; 2) a predicted probability of invasive breast cancer of more than 1.7% based on the Gail model; and 3) known deleterious BRCA1/2 mutation carrier or a family history of hereditary breast. Only 2 studies set a predetermined baseline MBD for study enrollment (15,26). Four studies limited their study population to premenopausal women who were not at increased risk for breast cancer (25-27,29).

No studies were eliminated based on the quality assessment of bias with majority of studies falling into either the good or poor category: 3 as good (25,26,30), 2 as fair (15,28), and 2 as poor (27,29). The most common reasons for marking down a study

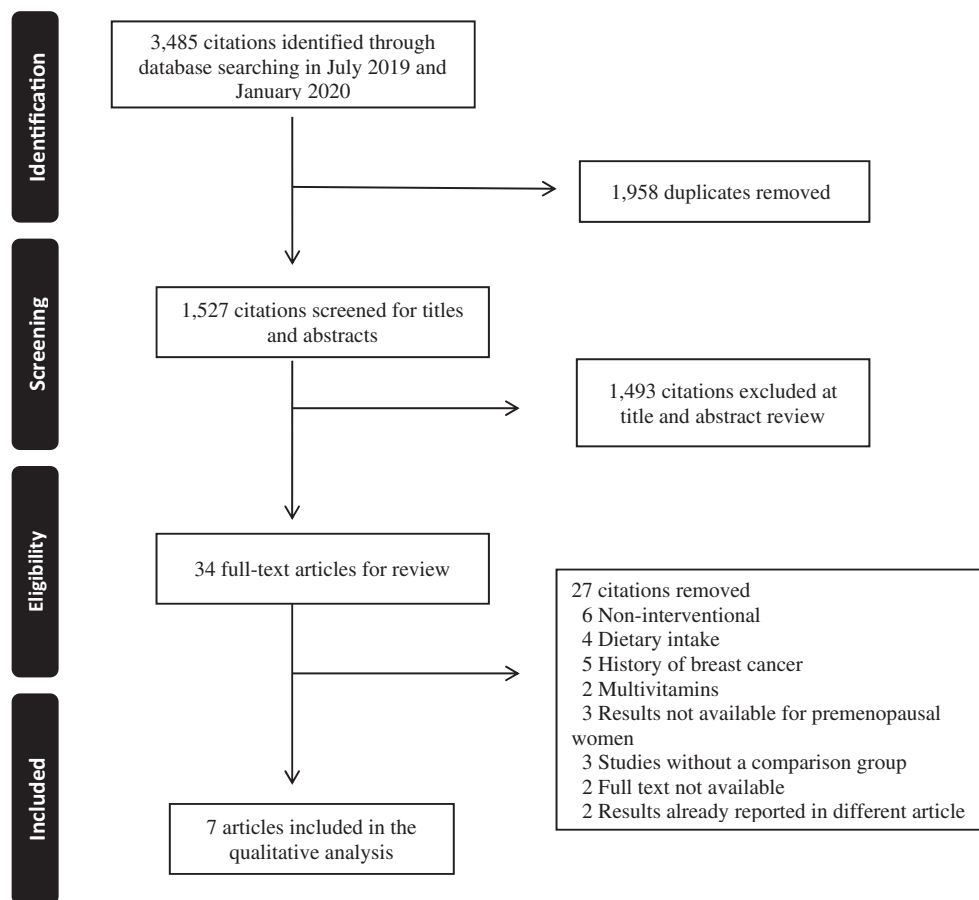


Figure 1. Flow diagram for included studies by preferred reporting items for systematic review and meta-analyses

Table 2. Characteristics of clinical trials on the associations of chemoprevention agents with mammographic breast density in premenopausal women and quality assessment<sup>a</sup>

Type of Trial	Study	Country	Experimental years	Chemoprevention agent (+ supporting agent)	Drug regimen	Treatment duration, months	Duration of follow-up, posttreatment, months	USPSTF criteria
Randomized (placebo-controlled)	Brisson et al., 2017 (26)	Canada	2012-2015	Vitamin D	PO QD 1000 IU, 2000 IU, 3000 IU	12	0	Good
	Pasta et al., 2015 (27)	Italy	2014-2015	Boswellia, betaine, and myo-inositol compound	PO QD 200 mg	6	0	Poor
	Powles et al., 2008 (28)	UK	~2000s	Isoflavone	PO QD 40 mg	36	0	Fair
	Cuzick et al., 2004 (15)	UK	1992-2001	Tamoxifen	PO QD 20 mg	54	0	Fair
Randomized (screening controlled)	Maskarinec et al., 2003 (25)	USA	~2000	Isoflavone	PO QD 100 mg	12	0	Good
	Howell et al., 2018 (30)	UK	2000-2004	Goserelin (+ raloxifene)	SQ Q28D 3.6 mg	24	84	Good
	Gram et al., 2001 (29)	USA	~1990s	Leuprolide acetate (+ conjugated estrogen + medroxyprogesterone acetate + methyltestosterone)	IM Q28D 7.5 mg	12, 24	0-12	Poor

<sup>a</sup>IU = international unit; IM = intramuscular; PO = per os/oral; QD = every day; Q28D = every 28 days; SQ = subcutaneous; USPSTF = United States Preventive Service Task Force.

**Table 3.** Baseline characteristics of premenopausal women enrolled into the clinical trials<sup>a</sup>

Type of trial	Study	Chemoprevention agent (+supporting agent)	Number of premenopausal women enrolled/analyzed	Mean or median age (SD), years	White race, %	Mean or median BMI (SD), kg/m <sup>2</sup>	Analyses adjusted for BMI and age
Randomized (placebo-controlled)	Brisson et al., 2017 (26)	Vitamin D	Total: 405/391				Yes
			1000 IU: 101/96	42.8 (5.2)	99.0	24.7 (5.1)	
			2000 IU: 104/99	42.7 (5.2)	100.0	24.3 (4.1)	
			3000 IU: 101/100	42.7 (5.5)	97.0	24.0 (3.6)	
	Placebo: 99/96	42.9 (6.2)	95.8	24.4 (4.1)			
Pasta et al., 2015 (27)	Boswellia, betaine, and myo-inositol compound	Total: 76/62				No	
		Experimental: NA/32 Placebo: NA/30	38.7 (6.1)	NA	NA		
Powles et al., 2008 (28)	Isoflavone	Total: 324/222				No	
		Experimental: 160/111 Placebo: 164/111	45 (NA) 45 (NA)	NA NA	24.8 (NA) 25.2 (NA)		
Cuzick et al., 2004 (15)	Tamoxifen	Total: 400/400				Yes	
		Experimental: 191/191 Placebo: 209/209	NA NA	NA NA	NA NA		
Maskarinec et al., 2003 (25)	Isoflavone	Total: 34/30				No	
		Experimental: 17/15 Placebo: 17/15	41.1 (3.1) 43.3 (1.7)	40.0 66.7	23.6 (3.8) 23.0 (2.9)		
Randomized (screening controlled)	Howell et al., 2018 (30)	Goserelin (+ raloxifene)	Total: 75/47				No
			Experimental: 38/20 Control: 37/27	36 (NA) 36 (NA)	NA NA	NA NA	
Gram et al., 2001 (29)	Leuprolide acetate (+ conjugated estrogen + medroxyprogesterone acetate + methyltestosterone)	Total: 21/19				No	
		Experimental: 14/12 Control: 7/7	NA NA	NA NA	NA NA		

<sup>a</sup>BMI = body mass index; IU = international unit; NA = not available

were failure to perform intention-to treat analysis and limiting the experimental group to only participants who were completely adherent to the intervention (Table 2).

### Effects of Chemoprevention Agents on Mammographic Breast Density

#### Selective Estrogen Receptor Modulators (SERM)

One randomized placebo-controlled trial investigated the effects of a SERM (tamoxifen [20 mg/day]) on MBD (15). Compared with placebo, the net reduction in percent density in the tamoxifen arm was 13.4% (95% confidence interval [CI] = 8.6% to 18.1%) after 54 months of treatment in women aged 45 years or younger at trial entry (n = 191 women), whereas in women aged 55 years or older, tamoxifen use was not associated with reduction in percent density (Table 4). The reduction in percent density was noticeable as early as the 18th month of intervention. The study controlled for important confounders, including BMI, age at entry to the trial, menopausal status, predicted familial risk of breast cancer, previous breast biopsy, and smoking status (31).

#### Gonadotropin-Releasing Hormone Agonists (GnRHA)

The 2 GnRHAs (leuprolide acetate and goserelin) were associated with net reductions in percent density in women at a high risk for developing breast cancer compared with placebo. Treatment with leuprolide acetate (n = 19 women) was associated with an 8.9% reduction in percent density (29), whereas treatment with goserelin (30) (n = 47 women) was associated with a 2.7% reduction in percent density (Table 4). The

interventions took place over 24 months in both studies. None of the studies adjusted their analyses for BMI or age.

#### Isoflavone

Isoflavone use was not associated with a reduction in MBD in the 2 studies that investigated this (25,28). There were non-statistically significant increases in the mean percent density in the isoflavone arm (2.5%) compared with the placebo arm (0.4%) after 12 months of intervention (100 mg/day of isoflavone vs placebo) (25). In the second study, the mean reduction in MBD in the isoflavone arm (40 mg/day) was lower (-3.0%) compared with the placebo arm (-6.6%,  $P = .2$ ) after 36 months of intervention (28) (Table 4). Both studies used digitized mammograms.

#### Boswellia, Betaine, and Myo-Inositol (Eumastós)

Treatment with Eumastós, which combined boswellia, betaine, and myo-inositol, was associated with a 60% reduction in MBD after 6 months of treatment ( $P < .01$ ) compared with a non-statistically significant 9.1% reduction (net difference of 50.9%) in MBD among women assigned to placebo (27). The analysis was not controlled for BMI or age.

#### Vitamin D

Vitamin D at 1000, 2000, and 3000 IU/day for 12 months did not reduce MBD when compared with placebo (26) (Table 4). Vitamin D at a dose of 3000 IU/day resulted in a slowing of the natural decrease in MBD compared with that observed in the placebo group: mean difference -1.9 (95% CI = -3.7 to -0.1;  $P = .03$ ). Dose response analyses showed a decrease in the rate of annual percent density change with an increasing dose of

Table 4. Changes in mammographic breast density (MBD) associated with chemoprevention agents among premenopausal women<sup>a</sup>

Type of agent	Study	Chemoprevention agent (+ supporting agent)	Duration of treatment, months	Mammogram time points	Mammogram type	MBD assessment (method)	Efforts to minimize bias in MBD assessment	Reduction in MBD	Difference in the mean/median change in MBD, % (95% CI) comparing intervention vs control arms	P
Selective estrogen receptor modulator (SERM)	Cuzick et al., 2004 (15)	Tamoxifen	54	Baseline, 18, 36, and 54 months	Digitized <sup>d</sup>	Boyd scale <sup>f</sup>	Control and intervention arms blinded. Readings done consecutively. Two reviewers.	Yes	13.4 (8.6 to 18.1)	<.01
GnRHa	Howell et al., 2018 (30)	Goserelin (+ raloxifene)	24	Baseline, 12, 24, and 36 months	Not specified	Boyd scale <sup>f</sup>	No blinding of treatment arms. Readings done consecutively. Two reviewers.	Yes	2.7 (NA)	<.01
	Gram et al., 2001 (29)	Leuprolide acetate (+ Conjugated estrogen + medroxy-progesterone acetate +methyltestosterone)	24	Baseline, 12, and 24 months. Additional mammogram in the intervention arm at 36 months <sup>e</sup>	Cranio-caudal mammogram Digitized <sup>d</sup>	Computer-assisted method	Control and intervention arms blinded. Readings randomized. Single reviewer.	Yes	8.9 (NA)	.01
Isoflavone	Powles et al., 2008 (28)	Isoflavone	36	Baseline, 12 and 36 months	Lateral view baseline Digitized <sup>d</sup> Digital <sup>c</sup> follow-up	Visual assessment score	Control and intervention arms blinded. Number of reviewers not specified.	No	-3.6 <sup>b</sup> (NA)	.20
	Maskarinec et al., 2003 (25)	Isoflavone	12	Baseline and 12 months	Cranio-caudal Digitized	Computer-assisted method	Control and intervention arms blinded. Readings randomized. Single reviewer.	No	-2.1 <sup>b</sup> (-6.9 to 2.6)	.37
Boswellia, beta-inositol compound	Pasta et al., 2015 (27)	Boswellia, beta-inositol compound	6	Baseline and 6 months	Not specified	Boyd scale <sup>f</sup>	Control and intervention arms blinded. Single reviewer.	Yes	50.9 (NA)	<.01
Vitamin D	Brisson et al., 2017 (26)	Vitamin D	12	Baseline and 12 months	Cranio-caudal Digital	Computer-assisted method	Control and intervention arms blinded. Films Deidentified. Single reviewer.	—	—	—
		1000 IU	—	—	—	—	—	No	-0.3 <sup>b</sup> (-2.1 to 1.5)	1.00
		2000 IU	—	—	—	—	—	No	0.1 (-1.7 to 1.9)	1.00
		3000 IU	—	—	—	—	—	No	-1.9 <sup>b</sup> (-3.7 to -0.1)	.03

<sup>a</sup>CI = confidence interval; GnRHa = gonadotropin releasing hormone agonists; IU = international unit; MBD = mammographic breast density; NA = not available.<sup>b</sup>Difference in the mean/median change in MBD with a negative sign indicates that the mean reduction in MBD in the placebo arm was greater than that in the intervention arm.<sup>c</sup>Digital format.<sup>d</sup>Mammogram digitized for measurement purposes.<sup>e</sup>Reading not available for all participants.<sup>f</sup>Boyd scale: A: 0%, B: 1%-10%, C: 11%-25%, D: 26%-50%, E: 51%-75%, and F: 76%-100%.

vitamin D ( $P = .02$ ). Analysis was controlled for age, BMI, calcium intake, initial percent breast density, timing, and technology of mammography.

### Persistent Reduction in MBD After Cessation of Intervention

Of the 4 chemoprevention agents that resulted in MBD reduction, only 2 (leuprolide acetate and goserelin) had posttreatment follow-up. Gram et al. (29) showed that MBD after 6-12 months off leuprolide acetate was not statistically significantly different from baseline. Similarly, Howell et al. (30) showed that after cessation of goserelin treatment, MBD tended to return to pretreatment baseline. Persistence of the effect of tamoxifen and Eumastós was not reported.

### Methods for Assessing Mammographic Breast Density

Methods used to assess MBD ranged from Boyd classification (15,27,30), visual assessment scores (28,30), and computer-assisted methods (Table 4) (25,26,29). Some studies used digital mammography for assessing MBD (26,28), whereas others scanned views of the mammogram and digitalized them (15,25,28,29). One study did not specify the type of mammogram used (27). For most studies, images were deidentified, order of films were randomized, and the intervention groups were blinded to reviewers to reduce bias (Table 4).

### Discussion

In this systematic review, we identified 7 clinical trials that investigated the associations of 6 chemoprevention agents with changes in MBD in premenopausal women with no history of breast cancer. To the best of our knowledge, this is the first systematic review on this subject. Only hormonal interventions (tamoxifen and GnRHAs) were associated with a reduction in MBD, and only tamoxifen was associated with a clinically relevant net reduction ( $>10\%$ ) in MBD after more than 12 months of treatment. However, MBD appeared to return to baseline levels after cessation of GnRHAs, and there is limited data on the long-term effect of tamoxifen for chemoprevention.

Hormonal interventions that modulate estrogen exposure influence breast cancer incidence in a direction predictable by their impact on estrogen levels (32,33). GnRHAs suppress the ovarian production of breast cell mitogens such as estrogen and decrease premenopausal breast epithelial cell proliferation (34,35). Interestingly, Howell et al. (30) used goserelin in conjunction with a SERM (ie, raloxifene). The latter ingredient was used as a supporting agent to mitigate adverse events (ie, preserve bone density) rather than an active ingredient for MBD reduction. Raloxifene is not indicated for use in premenopausal women because it has not shown a reduction in MBD when used on its own in this population (36). On the other hand, SERMs such as tamoxifen bind to  $\alpha$  and  $\beta$  estrogen receptors to activate estrogenic or antiestrogenic activities depending on the tissue (37,38). However, although most SERMs share a similar mechanism of action, only tamoxifen, as opposed to raloxifene or acolbifene (36,39), has shown a reduction in MBD. It is possible that this activity is due to a noncanonical pathway specific to tamoxifen, such as its pro-apoptotic activity through inhibition of cancerous inhibitor of protein phosphatase 2A and phospho-Akt (40,41). Thus, a better understanding of underlying mechanisms by which tamoxifen reduces MBD is needed.

Eumastós (boswellia, betaine, and myo-inositol) may act synergistically to exert numerous critical activities that could explain its effect on MBD. Myo-inositol reduces the expression of tumor growth factor- $\beta$  and nuclear factor- $\kappa$ B and controls inflammation in breast cancer cells (42,43). Boswellia modulates the expression of signaling molecules and cell-cycle regulators such as caspases, cytokines, and cyclin D1 (44). Betaine plays a role in breast cancer development and epigenetics specifically, serving as a methyl donor for conversion of homocysteine to methionine (45). Although Eumastós appears to be associated with a reduction in MBD after 6 months of treatment, there were several notable weaknesses in the study design, which limit how study findings can be interpreted. Thus, appropriately designed larger studies with robust adjustment for confounders and clearly defined density measures are needed to confirm the findings in the study. Finally, despite the initial promise of both vitamin D and isoflavones as chemoprevention agents from observational and experimental studies (46-50), up-to-date meta-analyses have shown no reduction in MBD with vitamin D (51,52) or isoflavones (53).

The effect of tamoxifen in premenopausal women is consistent with findings in postmenopausal women (36), and the USPSTF guidelines recommend its use as a chemoprevention agent in select populations (USPSTF) (20,54). The International Breast Cancer Intervention Study (IBIS) demonstrated that compared with women in the placebo group, women in the tamoxifen group who experienced at least 10% reduction in MBD had a 63% reduction in breast cancer risk while women who took tamoxifen but experienced less than 10% reduction in MBD did not have a reduction in breast cancer risk (55). A plausible explanation is that some women do not efficiently metabolize tamoxifen and will fail to demonstrate a treatment benefit, possibly due to genetic predisposition (56-58). Thus, for women receiving tamoxifen, assessing for MBD changes throughout the intervention period could determine who will benefit from tamoxifen and those who should receive alternative interventions (59). Furthermore, longer posttreatment cessation follow-up period is needed to evaluate the sustainability of the effect on MBD and evaluate a reduction in breast cancer incidence.

Although MBD could serve as a surrogate marker of breast cancer development (13-16), it is also worth noting that breast cancer can arise from other biological pathways unrelated to, and completely independent of, MBD. Hence, the fact that a chemoprevention agent does not reduce MBD does not imply that it has no utility in breast cancer prevention. In addition to assessing a change in MBD as an intermediate response endpoint, chemoprevention studies should evaluate the effect of other chemoprevention agents on other biomarkers that may be predictive of breast cancer incidence.

Despite tamoxifen being approved as breast chemoprevention by the Food and Drug Administration, its uptake for primary prevention in the United States is less than 3%, because of its side effects (60-63). Toxicity of the intervention can limit the acceptability and adherence to treatment. Adverse events were reported in 4 studies, although only 3 actively and systematically monitored for them. Although some studies did not assess for side effects of the interventions such as tamoxifen and leuprolide acetate, its use in other studies has been associated with vasomotor symptoms, menstrual abnormalities, uterine cancer, and thromboembolic phenomena (64,65). Goserelin was associated with vasomotor symptoms and sexual dysfunction, as well as weight gain and headaches (29). Up to 40% of the participants in the intervention arm indicated that they would not consider the option of further treatment for 5 years because of

the side effects, whereas several women opted out of the study because of treatment toxicity. As such, identification of new and well-tolerated chemoprevention agents targeting MBD is an urgent priority for primary breast cancer prevention.

A number of important limitations were found in the studies reviewed. Only 2 of the trials adjusted for important confounders such as BMI and age, which are both important determinants of MBD. Identified lifestyle factors account for 33% of the variance in MBD with current BMI accounting for the majority of this variance (66); hence, not adjusting for BMI is a major weakness when evaluating a chemoprevention agent. Methods for measuring MBD varied across studies with many of them utilizing Boyd classification and other computer-assisted measurements after digitalizing mammograms for MBD assessment. Currently, digital mammography is widely used, and new software packages are available for reporting MBD. Only 2 studies set a predetermined baseline MBD for study enrollment. Future studies should consider limiting trial enrollment to women with heterogeneously dense and extremely dense breasts because these are the groups with an elevated risk of breast cancer. Thus, there is an opportunity for future studies evaluating changes in MBD in response to chemoprevention agents to provide greater granular insight by using newer area- and volumetric-based measures, which provide more robust quantitative measures.

Studies did not always specify at which point within the menstrual cycle mammograms were taken, which is important because MBD varies with hormonal changes of the menstrual cycle (67). There was a lack of an adequate posttreatment follow-up, which is important for both determining the persistence of MBD reducing effect and long-term side effect profile. There was a lack of racial diversity in the study population, which limits generalizability of the results. Most of the studies included mostly non-Hispanic White women. This is relevant because MBD varies by race and ethnicity. Asian and African American women are more likely to have higher MBD, whereas Hispanic and Native American women have similar MBD compared with non-Hispanic White women (66,68-71). Furthermore, racial differences in response to some of the agents, such as the reduced breast cancer risk associated with isoflavone intake in Asian ethnicity vs non-Asian ethnicity women, have been previously demonstrated (47,48). Hence, studies among African American and Asian women are needed.

A potential limitation of this review is that we might have missed relevant articles. However, with our extensive literature search, we believe that all relevant publications are included in this review. An important strength of this review is that it is the first systematic review to evaluate data from clinical trials on the effect of chemoprevention agents on mammographic breast density in premenopausal women. This is especially timely given the recent USPSTF recommendations on chemoprevention use in high-risk women (54) and updated results on the IBIS-II trial in postmenopausal women (72). As such, findings could generate new ideas on breast cancer prevention in premenopausal women. Furthermore, we followed well-established guidelines on study selection.

In summary, only a limited number of chemoprevention agents have been shown to reduce MBD in premenopausal women without a history of invasive breast cancer. New and well-designed randomized controlled trials targeting new chemopreventive agents with robust MBD assessments and longer posttreatment follow-ups, as well as larger studies to confirm agents that have been studied in small trials, are needed to identify well-tolerated chemoprevention agents that can reduce MBD with minimal side effects. This has great potentials to

open up new opportunities for breast cancer prevention in premenopausal women.

## Funding

This work was supported by the National Cancer Institute at the National Institutes of Health (grants R37CA235602 and R01CA246592 to ATT).

## Notes

**Role of the funder:** The funding source did not have any role in the design and conduct of the study; interpretation of the data; preparation, review, or approval of the manuscript; and decision to submit the manuscript for publication.

**Disclosures:** All other authors declare no competing interests.

**Prior presentation:** Presented as abstract at the San Antonio Breast Cancer Symposium in San Antonio, TX, December 12, 2019.

**Disclaimer:** The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

**Author contribution:** Conception: ATT Design: ASS, MR, ATT. Collection and assembly of data: All authors. Data analysis and interpretation: All authors. Manuscript writing: All authors. Final approval of manuscript: All authors. Accountable for all aspects of the work: All authors.

## Data Availability

No new data were generated or analyzed in this research.

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