Breast Cancer Research and Treatment (2006) 97: 157–166
DOI 10.1007/s10549-005-9105-8

Epidemiology

Breast cancer risk factors and mammographic breast density in women over age 70

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Key words: breast cancer risk, epidemiology, hormones, mammographic breast density, prospective cohort study, SOF

Summary

Background. Breast density is a strong risk factor for breast cancer, but little is known about factors associated with breast density in women over 70.

Methods. Percent breast density, sex hormone levels and breast cancer risk factor data were obtained on 239 women ages 70–92 recruited from 1986 to 1988 in the United States. Multivariable linear regression was used to develop a model to describe factors associated with percent density.

Results. Median (range) percent density among women was 23.7% (0–85%). Body mass index ($\beta = -0.345$, $p < 0.001$ adjusted for age and parity) and parity ($\beta = -0.277$, $p < 0.001$ adjusted for age and BMI) were significantly and inversely associated with percent breast density. After adjusting for parity and BMI, age was not associated with breast density ($\beta = 0.05$, $p = 0.45$). Parous women had lower percent density than nulliparous women (23.7 versus 34.7%, $p = 0.005$). Women who had undergone surgical menopause had greater breast density than those who had had a natural menopause (33.4 versus 24.8%, $p = 0.048$), as did women who were not current smokers (26.0 versus 17.3% for smokers, $p = 0.02$). Breast density was not associated with age at menarche, age at menopause, age at first birth, breastfeeding, estrogen levels or androgen levels. In a multivariable model, 24% of the variance in percent breast density was explained by BMI ($\beta = -0.35$), parity ($\beta = -0.29$), surgical menopause ($\beta = 0.13$) and current smoking ($\beta = -0.12$).

Conclusion. Factors associated with breast density in older, post-menopausal women differ from traditional breast cancer risk factors and from factors associated with breast density in pre-menopausal and younger post-menopausal women.

Introduction

Breast cancer is the most common cancer in American women, with an estimated 211,240 new invasive cases to be diagnosed in 2005 [1]. Age is one of the greatest risk factors for the disease, with incidence rates increasing steadily between the ages of 20 and 80 [2]. More than 35% of newly diagnosed invasive cases are in women over the age of 70 [3].

After age and carriage of a *BRCA1*/2 mutation, mammographic breast density is the strongest risk factor for breast cancer. Both case–control and cohort studies estimate the risk associated with greater density to range from 1.4 to 6.2, with an apparent dose-dependent relationship [4]. Notably, breast cancer risk associated with dense breast tissue may be greater for older women [5], although breast density *per se* decreases with age [4], With the exception of age and BMI, most established breast cancer risk factors are positively associated with increasing breast density [4,6]. Moreover, hormone therapy, an established breast cancer risk factor [7], increases breast density [8]. In contrast, tamoxifen and raloxifene, two selective estrogen receptor modulators (SERMs) associated with reduced breast cancer risk [9,10], do not increase breast density [11,12]. These observations, together with studies suggesting that reducing breast density also decreases subsequent breast cancer risk [13,14] imply that understanding factors influencing breast density may help us understand breast cancer risk. The fact that increasing breast density reduces mammographic screening sensitivity and specificity [15,16] further underscores the need to understand

factors associated with breast density. However, most studies of breast density have been conducted in women under age 70 and very little is known about factors associated with breast density in this older, potentially ''higher risk'' age group.

The steady increase in the aging population of the United States [17] together with the increasing lifespan of women [18] in this country suggests that the number of breast cancer cases in women over age 70 will steadily rise. Hence, women over 70 will account for a large and growing proportion of the breast cancer population. Therefore, understanding factors that may be associated with mammographic breast density, which may subsequently impact both breast cancer risk and early detection, in women age 70+ can have important public health implications. We used data from an ongoing study of women age 70 and over to begin to understand factors associated with mammographic breast density in older women.

Methods

Study participants

Subjects for the current study were a subset of participants in the Study of Osteoporotic Fractures (SOF), a prospective study of 9704 white, community-dwelling women who were at least 65 years of age at study enrollment [19]. SOF participants were recruited from 1986 to 1988 using population-based lists (e.g., voter registration, health maintenance organizations, and motor vehicle tapes) at four clinical centers: the University of Maryland (Baltimore), the University of Minnesota (Minneapolis), the University of Pittsburgh, and the Kaiser Permanente Center for Health Research (Portland, OR). Women were excluded from the SOF if they reported a bilateral hip replacement or were unable to walk without assistance. African-American women were initially excluded from the SOF because their risk of hip fracture is low and thus are not included in the analyses presented here. The institutional review boards at each institution approved the study. All women provided written informed consent at study entry and at each clinical examination.

For the analysis presented here, we limited the women to those who had had baseline measurements of sex steroid hormones as part of two SOF sub-studies. The first study investigated the association between hormone levels and risk of hip or vertebral fractures [20]. The second study investigated the association between hormone levels and breast cancer. Both studies excluded women taking any form of hormone therapy at baseline and employed a case-cohort design [21] in which cases were the women with the outcome of interest (fracture or breast cancer) and controls were women free from the outcomes and randomly chosen from the remainder of the SOF cohort. A benefit of the case-cohort design is that because the controls are chosen at random from the cohort, they are representative of the entire cohort. A total of 827 women were included in those two sub-studies. After eliminating women who died or reported a history of breast cancer ($n = 563$) prior to study visit 6 (10 years post-enrollment), 264 women remained eligible for the present study on mammographic density. At study visit 6, informed consent to participate in this new ancillary study along with the date and location of the last screening mammogram were obtained from each of the 264 women who were eligible for the study. Participants also signed a release form providing permission for the study team to obtain a copy of their latest mammographic films. A letter requesting a copy of the latest films along with a copy of the signed release form was then sent to all the patient-identified mammography clinics. Among the 264 women, mammograms were not located on 25 women (9.5%); therefore, a total of 239 women from all four SOF sites were included in this analysis.

Demographic and risk factor data

At the baseline visit, age at menarche, age at menopause, reproductive history, family history of breast cancer, past use of hormone therapy (HT), current walking habits, smoking and alcohol use, and prior estrogen use were determined by self-administered questionnaires, which were reviewed by trained staff. A positive family history of breast cancer was defined as breast cancer in a mother or sister. Weight (in lightweight clothing with shoes removed) was measured using a calibrated balance beam scale. Self-reported height at 25 years of age was used to calculate the modified body mass index (BMI). Height at age 25 years rather than current height was used to calculate BMI because as they age, older women tend to experience height loss due low bone mass and subsequent vertebral fractures.

Measurement of sex steroid hormones

Estrogen and androgen levels were measured in serum obtained at baseline by the two SOF sub-studies [20]. Participants were instructed to adhere to a fat-free diet the evening before and the morning of the blood draw in order to minimize lipemia that could interfere with the hormone measurement assays. Blood was drawn between 8:00 am and 2:00 pm, separated into serum, plasma and buffy coat according to standardized protocols, and immediately frozen at −20 °C. All samples were shipped to a central repository within two weeks, where they were stored at −190 °C until analyzed.

Hormone measurements were done by Endocrine Sciences (Calabasas Hills, CA) for women from the fracture sub-study and by Corning Nichols Institute (San Juan Capistrano, CA) for women from the breast cancer sub-study. At Endocrine Sciences, total estradiol and estrone were measured by radioimmunoassay (RIA) and separated by liquid chromatography (inter-assay

variability 8–12.5%, 6.2–7%, respectively). Total testosterone was measured by RIA after extraction and aluminum oxide column chromatography (inter-assay variability 6.1–13.4%). Free testosterone was measured with an ammonium sulfate precipitation procedure (inter-assay variability 10.7–15.5%). The testosterone-binding capacity of sex hormone binding globulin (SHBG) was measured by means of a displacement technique (inter-assay variability 4.1–14.4%). At Corning Nichols Institute, estrone was measured using extraction, chromatography and RIA (inter-assay variability $<8\%$). Total estradiol was measured using liquid organic extraction, column chromatography and RIA (inter-assay variability 6–12%). SHBG was measured using RIA (inter-assay variability 4.4%). Total testosterone was measured using RIA with chromatographic purification. Equilibrium dialysis was used for free testosterone method. Calculation of free testosterone was adjusted for albumin concentration (inter-assay variability 7%). DHEAS was measured using RIA after preparation for analysis by serial dilution (inter-assay variability 10–13%). Undetectable levels of total estradiol ($n=67$) were given a value of midway between 0 and the laboratory's reported sensitivity of the assay (2.5 pg/ml for assays performed by Endocrine Sciences and 1 pg/ml for assays performed by Corning Nichols Institute because the limits of detection of total estradiol were 5 pg/ml at Endocrine Sciences and 2 pg/ml at Corning Nichols Institute).

Measurement of breast density

Mammographic density was determined by a single, expert reader (Ms. Martine Salane) [22,23] using the craniocaudal view of the right breast unless the quality of the mammogram on the right side was poor, in which case the left side was used. All films were relabeled with a study ID so that the reader remained blinded to the subject's identity. All areas of mammographic density on a craniocaudal view were outlined using a china marker. Isolated calcifications, biopsy scars, Cooper's ligaments, and breast masses were not considered in the assessment. Total area of the breast and the outlined regions of mammographic densities were measured using a compensating polar planimeter (LASICO, Los Angeles, CA). Measurements of total breast area and marked dense area(s) on the mammogram were measured twice to ensure accuracy. Percent mammographic breast density was calculated by dividing the total breast areas with density by the total breast area. Ms. Salane's planimetry method has shown high correlation with computer-assisted density measurements ($\rho=0.90$) [22].

To determine the reproducibility of the density readings, mammographic images of five women from each of the four centers (20 women total) were randomly chosen, assigned a new study ID and re-evaluated. The reviewer was blind to the original readings. Intra-observer agreement in the involved breast area and total breast area were 97 and 99%, respectively. These data are consistent with previous reports of reproducibility for Ms. Salane's readings from the Breast Cancer Detection Demonstration Project (BCDDP) (ICC $\rho=0.915$ for 193 sets of films) [23].

Statistical analyses

To assess the associations of reproductive and hormonal factors with percent breast density, we first used linear regression or analysis of variance (ANOVA), adjusting for age, BMI and parity, three breast cancer risk factors previously consistently shown to be related to breast density. Linear regression was used for continuous variables (hormone levels, age, BMI, age at first period, at last period, years since last period, age at first birth). Mean percent density across levels of categorical variables was assessed using ANOVA. All categorical variables were dichotomous (yes/no) and included: ever parous, age at first birth <20, age at first birth >35, ever breast fed, had a surgical menopause, past use of estrogen, current smoker, walks for exercise, ever had fibrocystic breast disease, and reported family history of breast cancer. Because age, BMI, walking for exercise and perhaps current smoking may affect breast density and because mammogram films used in the analyses were those identified as being taken closest to visit 6, visit 6 values were used these variables. Baseline values were used for all other variables. Finally, we used forward, stepwise multivariable linear regression to develop a model describing the factors associated with percent breast density [24]. The model building process proceeded as follows. First, we separately regressed on the outcome variable (percent breast density) each individual explanatory variable whose adjusted p-value from the linear regression or ANOVA analyses was <0.20. The variable that explained the largest proportion of the outcome variation was then selected as the first variable to be entered into the regression equation. Each remaining explanatory variable was then regressed on the outcome variable jointly with the first variable. The variable that provided the largest gain in explanatory power was then added in as the second variable in the multiple regression equation. This process was repeated for the remaining variables. At each step the maximum gain in variation explained was tested against the variation still unexplained at this stage and the process was terminated when the maximum gain at a given step was not significantly different from pure random variation [24]. All analyses were done with the SPSS statistical software package (version 11.0). Two-sided p-values <0.05 are reported as statistically significant.

Results

Table 1 shows the baseline characteristics of the study population compared to all SOF women. Participants were somewhat younger and heavier than the entire

Table 1. Comparison of study population characteristics to all the Study of Osteoporotic Fractures (SOF) participants at baseline and to SOF participants at visit 6

	Baseline		*p*-value	Visit 6		*p*-value
	Sample $n=239$ Mean (SD)	SOF $n=9704$ Mean (SD)		Sample $n=239$ Mean (SD)	SOF $n=6991$ Mean (SD)	
Age (years)	69.6 (3.9)	71.7 (5.3)	<0.001	78.6 (3.8)	80.9 (4.7)	0.15
Weight (kg)	69.0 (12.6)	67.1 (12.5)	0.01	68.3 (12.7)	65.6 (12.9)	0.12
Height at age 25 (cm)	163.1 (6.0)	162.6 (6.0)	0.20	163.1 (6.0)	157.4 (6.2)	0.46
Body mass index (kg/m^2)	27.0 (4.6)	26.5 (4.7)	0.09	27.3 (4.7)	26.5 (4.8)	0.10
Age at menarche (years)	12.9 (1.4)	13.0 (1.5)	0.37	12.9 (1.4)	13.0 (1.5)	0.24
Age at first birth (years)	25.7 (4.9)	25.4 (5.0)	0.36	25.6 (4.9)	25.4 (5.0)	0.42
Age at menopause (years)	48.1 (5.3)	47.9 (5.8)	0.63	47.2 (6.0)	47.0 (6.4)	0.69
Parity	2.43 (1.86)	2.24 (1.8)	0.09	2.43 (1.86)	2.31 (1.75)	0.26
Surgical menopause (%)	10.5	11.9	0.44	10.5	13.0	0.23
Ever pregnant (%)	84.5	83.7	0.75	84.5	84.0	0.79
Nulliparous (%)	16.7	18.9	0.38	16.7	21.2	0.27
Ever breastfed (%)	58.6	57.5	0.74	58.6	58.2	0.84
Family history of breast cancer (%)	12.6	13.3	0.72	12.6	14.2	0.58
Current smoker (%)	8.8	10.0	0.56	5.0	8.0	0.19
Past estrogen use (%)	33.1	26.7	0.09	33.9	32.0	0.59
Walks for exercise (%)	54.0	50.1	0.23	41.8	52.5	0.12

cohort. Because only women who attended visit 6 were eligible for this sub-study, we also compared the study population to those SOF women who attended study visit 6. The differences between participants in the sub-study and women attending visit 6 were similar to that of the entire cohort, although visit 6 participants were more likely to walk for exercise, smoke, have had their ovaries removed or report a family history of breast cancer. However, none of these differences between all women attending visit 6 and the subset of women included in this current study approached significance, suggesting that in general the subset of participants included in these analyses were representative of both the entire SOF cohort and those attending visit 6. At visit 6, participants in this sub-study were on average over 78 years old (range: 70–92 years) and overweight (mean BMI = 27.3 kg/m^2, range: 18.1–45.2 kg/m^2).

Percent mammographic density was not normally distributed and ranged from 0 to 85% (Figure 1). Mean (SD) percent density was 25.6% (20.2) and median percent density was 23.7%. Thirty women (12.5%) had a density measurement of 0. The median (25th, 75th percentiles) time between mammogram date and visit 6 was 7 (3, 12) months. Almost 95% of women had had a mammogram within 2 years of study visit 6. As shown in Table 2, percent density appeared to increase with age. The mean percent density ranged from 19.2% for women ages 70–74, to 33.9% for women 85 and over.

Table 3 summarizes the sex-steroid hormone serum concentrations. In general, hormone levels were not normally distributed. Total estradiol levels were undetectable in 67 (28%) the women. Table 4 summarizes the associations between demographic, reproductive and hormonal factors and percent breast density. Body mass index ($\beta=-0.345$, $p<0.001$ adjusted for age and parity) and parity ($\beta=-0.277$, $p<0.001$ adjusted for age and BMI) were significantly and inversely associated with percent breast density. Women who had given birth were more likely to have a lower mean percent breast

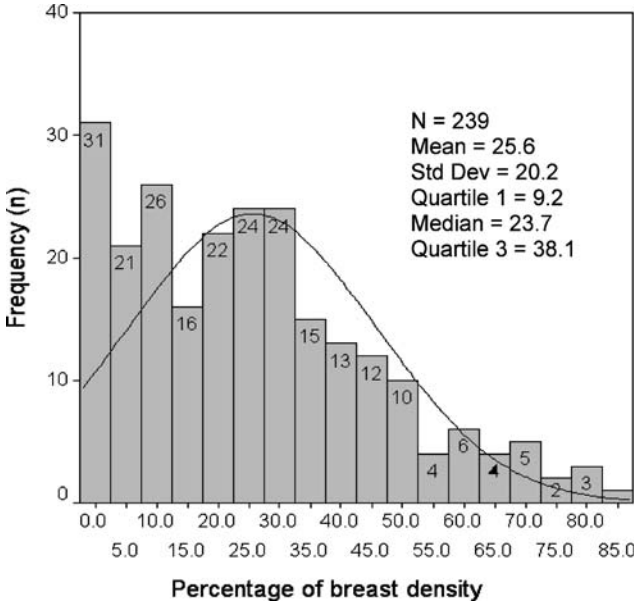

Figure 1. Percent mammographic density in 239 women ages 70–92 participating in SOF (line shown is a normal curve overlayed on the data).

Table 2. Percent breast density by age category in 239 women ages 70–92 participating in SOF

Age group	*n*	Mean (SD)	Range	25th Percentile	50th Percentile	75th Percentile
70–74	22	19.2 (18.7)	0–62.7	5.1	11.9	33.8
75–79	128	24.37 (18.9)	0–80.7	9.4	23.4	36.0
80–84	72	27.7 (20.7)	0–84.9	11.2	24.9	40.3
85+	17	33.9 (25.5)	0–74.1	9.9	28.8	59.6
All women	239	25.6 (20.2)	0–84.9	9.2	23.7	38.1

Table 3. Sex-steroid hormone serum concentrations in 239 women ages 70–92 participating in SOF

	Mean (SD)	Range	25th Percentile	50th Percentile	75th Percentile
Estrogens					
Total estradiol, pg/ml	8.2 (7.9)	1.0–56.0	4.0	7.0	10.0
Estrone, pg/ml	29.9 (28.4)	4.0–230	17.0	23.0	32.5
Androgens					
Dehydroepiandrosterone sulfate, μg/dl	52.6 (45.3)	0–333	23.0	41.0	66.0
Total testosterone, pg/ml	207.7 (137.8)	32–1000	100.0	170.0	272.5
Free testosterone, pg/ml	2.4 (1.8)	0.2–11	1.0	1.9	3.2
Percent free testosterone, %	1.1 (0.5)	0.3–3.8	0.8	1.1	1.4
Other					
SHBG, μg/dl	1.6 (1.1)	0.10–6.9	0.9	1.4	2.1

density compared to nulliparous women (23.7 versus 34.7%, $p = 0.005$). Women who had undergone surgical menopause had greater breast density than those who had had a natural menopause (33.4 versus 24.8%, $p = 0.048$), as did women who were not current smokers (26.0 versus 17.3% for smokers, $p = 0.02$). Past users of estrogen also had greater breast density, although that relationship was not significant (29.5 versus 23.5%, $p = 0.14$). Although in univariate analyses age was significantly associated with percent breast density ($\beta = 0.172$, $p = 0.008$, data not shown), this relationship did not remain after adjustment for BMI and parity, suggesting evidence of confounding between BMI and age. Indeed, as shown in Table 5, BMI significantly decreased with increasing age ($p = 0.003$).

Finally, Table 6 shows the results from the multivariable forward, stepwise linear regression for percent breast density. Included in the stepwise regression procedure were BMI, parity, surgical menopause, current smoker, and past use of estrogen. The final model included only BMI, parity, surgical menopause, and current smoker and explained 24% of the variation in percent breast density. BMI ($\beta = -0.35$), parity ($\beta = -0.29$), and current smoking ($\beta = -0.12$) were significantly and inversely associated with density in this model, whereas surgical menopause ($\beta = 0.13$) was significantly and positively associated with percent breast density.

Discussion

In this study, approximately 25% of white women between the ages of 70 and 92 years old who do not use hormone therapy have a mammographic breast density of more than 50%. This finding is consistent with the only other study to date to report percent breast density specifically in women over 70, wherein approximately 24% of women 70–79 were found to have greater than 50% density [25]. In our study, we found that almost one-quarter of the variation in breast density among these older women could be explained by BMI, parity, surgical menopause, and current smoking. To our knowledge this is the first study to look at correlates of breast density specifically in women over 70.

Mammographic breast density serves as an estimate of the proportion of fibroglandular tissue to fat in the breast. As a woman ages, the breast epithelium involutes and the percentage of breast fat increases, causing mammograms to become more radiolucent [26]. The drop in endogenous hormones during the menopausal transition further augments breast epithelium involution [27]. Consistent with this biology, mammographic breast density has been shown to decrease with age, especially after the menopause [28]. As well, later age at menopause has been associated with greater breast density [29].

In contrast to the inverse association between age and breast density reported among pre-menopausal women and younger post-menopausal women [30–33], we found that among older post-menopausal women breast density appeared to increase with age. However, this association was no longer evident after adjusting for BMI and parity. The fact that in our population BMI significantly decreased with increasing age combined with the fact that breast density increased with decreasing BMI suggests that confounding between age and BMI likely explains the apparent positive associa-

Table 4. Associations between percent breast density and reproductive and hormonal factors among 239 women ages 70–92 participating in SOF

	n	Beta	Adjusted *p*-value[a]	
Steroid hormones				
Total estradiol, pg/ml	192	0.059	0.37	
Estrone, pg/ml	237	0.028	0.64	
Free testosterone, pg/ml	237	−0.005	0.93	
Total testosterone, pg/ml	237	−0.046	0.43	
Percent free testosterone, %	237	0.018	0.77	
Dehydroepiandrosterone sulfate, μg/dl	231	0.001	0.99	
SHBG, μg/dl	237	−0.043	0.47	
Demographic and reproductive variables				
Age	238	0.046	0.45[b]	
Body mass index	238	−0.345	**< 0.001**[c]	
Age at first period	226	−0.056	0.35	
Age at last natural period	236	0.025	0.67	
Years since last natural period	236	−0.030	0.67	
Age at First birth	195	−0.033	0.62	
Parity	238	−0.277	**< 0.001**[d]	
Comparison of percent density between groups				
	No. of subjects	Mean percent density	SD	Adjusted *p*-value[e]
Parous				
No	40	34.72	22.64	
Yes	198	23.70	19.12	**0.005**[f]
Age at first birth < 20[g]				
No	177	23.87	18.94	
Yes	18	23.48	21.62	0.86
Age at first birth > 35[g]				
No	187	23.7	19.37	
Yes	8	26.94	13.17	0.91
Ever breast fed				
No	98	28.19	20.71	
Yes	140	23.71	19.58	0.37
Surgical menopause				
No	206	24.78	19.29	
Yes	24	33.43	25.6	**0.05**
Past estrogen use				
No	154	23.48	18.84	
Yes	81	29.54	21.83	0.17
Current smoker				
No	226	25.99	20.33	
Yes	12	17.29	13.92	**0.02**
Walks for exercise				
No	138	24.86	19.91	
Yes	99	26.33	20.51	0.93
Ever had fibrocystic breast disease				
No	193	24.22	19.79	
Yes	37	31.20	20.09	0.57
Family history of breast cancer				
No	185	26.09	20.77	
Yes	29	26.88	18.82	0.98

[a]Adjusted for age, BMI and parity, except as noted.
[b]Adjusted for BMI and parity.
[c]Adjusted for age and parity.
[d]Adjusted for age and BMI.
[e]Adjusted for age, BMI and parity, except as noted.
[f]Adjusted for age and BMI.
[g]Among parous women.

Table 5. Body mass index (BMI) by age category in 239 women ages 70–92 participating in SOF

Age group	*n*	Mean (SD)
70–74	22	29.7 (4.7)
75–79	128	27.7 (4.9)
80–84	72	26.4 (3.9)
85+	17	25.1 (3.8)

tion between breast density and age in this group of women. The lack of an association between age and density after adjusting for BMI and parity suggests that the age-breast density association may weaken and even disappear after a certain age.

Furthermore, in contrast to the majority of reports in the literature, which again were conducted in younger post-menopausal women [29,33], we did not find any significant association between age at menopause and breast density within our population of older women. This observation is consistent with the only other previous study to investigate the association between age at menopause and breast density according to current age. El-Bastawissi et al. [33] reported that among 14,432 women, later age at menopause was associated with greater BIRADS breast density in women under 65 but not in women over 65. Interestingly, we observed a positive association between surgical menopause and breast density, possibly reflecting an early life hormonal milieu that could affect breast structure as well as result in the need for a surgical menopause. Notably, age at menopause (whether surgical or natural) is an established breast cancer risk factor [34], although we found no association between breast cancer and age at menopause in the overall SOF cohort [35,36].

Several other reproductive factors have been reported to be associated with both increased breast cancer risk and greater breast density in pre- and post-menopausal women, including nulliparity, late age at first birth, lower parity, and possibly early age at menarche [25,37–51]. However, in the current study, we found no association between breast density and these breast cancer risk factors, except for parity. Like increasing age and later age at menopause, bearing children may also have a biological basis for altering breast density. Pregnancy is associated with a change in breast structure to more differentiated lobules with less cell proliferation [52]. The greatest effect on breast structure derives from the first pregnancy, with some continued differentiation with each subsequent pregnancy [52]. Our findings that parity but not other reproductive factors is associated with breast density in older women suggests that some factors may have a more permanent effect on breast density, whereas the effects of other factors on density may diminish with time. For example, pregnancy induces a permanent structural change in the breast. In contrast, it is possible that the hormonal effects of an early menarche or late menopause might decrease as a woman ages; that is, the "recency" of the exposure may explain the difference between our findings in older post-menopausal women and those in younger post-menopausal women reported by others.

In contrast to our general lack of findings between hormonal exposures and breast density, we observed an inverse association between smoking and percent breast density. This finding is consistent with recent data reported for pre- and peri-menopausal women in the Study of Women Across the Nation (SWAN) [53]. Notably, the association between smoking and breast cancer is inconsistent [54]. Nonetheless, the inverse relationship between smoking and breast density might reflect the anti-estrogenic effects of cigarette smoking [55], which could reduce the proliferation of breast epithelial cells and subsequently decrease breast density. However, the association between estrogens and breast density remain unclear. Epidemiologic data linking breast cancer risk factors to breast density generally support the contention that endogenous estrogen levels affect breast density just as they do breast cancer risk [56]. We, too, have reported that elevated estrogen levels are associated with an increase in breast cancer risk among women in the SOF cohort [57]. To date, only one study has examined the association between endogenous hormones and breast density. Boyd et al. [58] showed that free estradiol (negatively, $\beta = -0.28$, $p < 0.001$) and sex hormone binding globulin (SHBG) (positively, $\beta = 0.05$, $p < 0.001$) were significantly related to percent breast density among 189 post-menopausal women. These associations were attenuated (but remained significant) after adjustment for age and waist circumference (adjusted $\beta = -0.09$ and 0.02 for estradiol and SHGB, respectively). In our study, there were no associations between percent breast density and any of the

Table 6. Multivariable stepwise linear regression for percent breast density among 239 women ages 70–92 participating in SOF

Variable	Simple regression			Stepwise multiple regression[a]		
	Beta	Standard error	$Pr > \|t\|$	Beta	Standard error	$Pr > \|t\|$
Body mass index	−0.37	0.26	< 0.001	−0.35	0.25	< 0.001
Parity	−0.30	0.67	< 0.001	−0.29	0.64	< 0.001
Surgical menopause	0.13	4.31	0.046	0.13	3.83	0.03
Past estrogen use	0.14	2.73	0.028	Removed from model		
Current smoker	−0.09	5.94	0.144	−0.12	5.5	0.04

[a] $R^2 = 24\%$.

steroid hormones in univariate analyses or in multivariable models adjusting for age, BMI and parity. We must note, however, that the hormones were measured in baseline sera, whereas the breast density measurements used mammograms taken approximately 10 years after baseline. Thus, it is possible that the lack of association observed in the present study reflects the time lapse between hormone and density measurements. However, given the fact that in the overall SOF study, baseline hormone levels were predictive of subsequent breast cancer development [57] and assuming that breast density is a surrogate marker for breast cancer risk, a relationship between baseline endogenous hormones and subsequent breast density was anticipated. The observed lack of an association between hormone levels and breast density in this prospective study among older women may further support the contention that recency of a hormonal exposure might be an important factor in its relationship to breast density. That is, as a woman ages, the effect of earlier hormonal exposure on breast density decreases.

Previously, we reported that traditional Gail model risk factors for breast cancer [59] may not apply to older post-menopausal women [35,36]. This suggests that breast cancer in older post-menopausal women may be etiologically different from disease in younger post-menopausal women. Data on tumor biology support this assertion: women over age 65 tend to have tumors with less aggressive histologies and more favorable tumor profiles characterized by a higher percentage of estrogen-receptor positive (ER+) tumors, with the percent of ER positivity increasing with age [60,61]. Lower s-phase fraction and HER2/neu negativity, markers of reduced cell proliferation, are also features of tumors in elderly women [62]. Interestingly, there appears to be no difference in breast density between women with ER+ tumors and those with ER− tumors [63]. In general, the results presented here suggest that factors associated with breast density in older post-menopausal women are different from those identified in younger post-menopausal women and pre-menopausal women. Thus, if breast density is a biomarker for breast cancer risk, the data presented herein would lend support to the assertion that, epidemiologically, breast cancer in older women may be distinct from that in younger women.

There are several features of the present study that warrant discussion. First, the limited sample size precludes us from detecting modest associations. Nonetheless, this is the first study of breast density in older post-menopausal women and our sample size was larger than the only other study to examine the association between endogenous hormone levels and breast density in post-menopausal women [58]. Moreover, the use of a single, expert breast density reader helps to reduce variability in breast density measurements and subsequent measures of effect. Because risk factor data were obtained by self report, these factors may have been under- or over-reported by participants, thereby attenuating our results. However, because these data were prospectively collected, it is unlikely that reporting would systematically differ according to mammographic density; thus recall bias should be minimal. In addition, standardized methods for collecting risk factor information help to ensure the data quality. Participants in this sub-study were, on average, 2 years younger than women in the entire SOF cohort, suggesting an under-representation of older post-menopausal women, although 27% of the women in this ancillary study were over age 80. In addition, because participants were consented 10 years after baseline, women who died or were too ill to attend the visit were not given the chance to participate. Hence, the women included here likely represent healthier older women and our results could reflect characteristics unique to those women. However, at least for the demographic factors we explored, the women included in this analysis resembled women in both the overall SOF cohort and those who attended clinic visit 6 (Table 1). Finally, we cannot exclude the possibility that our findings may be the result of a differing prevalence of breast cancer and density risk factors between the SOF participants and participants in other studies of post-menopausal breast density. The results may also be due to chance or may be confounded by some unidentified factor.

In conclusion, this is the first study to investigate correlates of breast density exclusively in women age 70 years and older. Factors associated with breast density appear to be different for these older women compared to published data on younger post-menopausal women and different from the traditional Gail model breast cancer risk factors. Surprisingly, most women had more than 23% breast density and density did not decrease with increasing age as would be expected in women over 70 years of age. Because breast density is a determinant of mammographic sensitivity and breast density is believed to decrease with age, our data suggest that mammographic screening may not be as sensitive as anticipated in women over age 70. Our findings together with the aging population of the US and the increasing life span of women emphasize the public health importance of large-scale prospective studies of factors associated with mammographic breast density and breast cancer risk after menopause, especially in women over age 70.

Acknowledgments

We thank Chandra Marriott for her technical assistance with this manuscript and Dr. Jeffrey L. Eppinger for his comments on this work. Investigators in the Study of Osteoporotic Fractures Research Group: University of California, San Francisco (Coordinating Center:) SR Cummings (principal investigator), KL Stone (co-investigator), DC Bauer (co-investigator), MC Nevitt (co-investigator), W Browner, (co-investigator), DM Black (study statistician), H.K. Genant (director, central

radiology laboratory), R. Benard, T. Blackwell, M. Dockrell, S. Ewing, C. Fox, R. Fullman, D. Kimmel, S. Litwack, LY. Lui, J. Maeda, P. Mannen, L. Nusgarten, L. Palermo, M. Rahorst, C. Schambach, J. Schneider, R. Scott, D. Tanaka, C. Yeung. University of Maryland: M.C. Hochberg (principal investigator), L. Makell, (project director), R. Nichols, C. Boehm, L. Finazzo, T. Page, S. Trusty, B. Whitkop. University of Minnesota: KE Ensrud (principal investigator), K. Margolis (co-investigator), P. Schreiner (co-investigator), K. Worzala (co-investigator), S. Love (clinical research director), E. Mitson (clinic coordinator), C. Bird, D. Blanks, F. Imker-Witte, K. Jacobson, K. Knauth, N. Nelson, E. Penland-Miller, G. Saecker. University of Pittsburgh: J.A. Cauley (principal investigator), L.H. Kuller (co-principal investigator), M. Vogt (co-investigator), L. Harper (project director), L. Buck (clinic coordinator), C. Bashada, D. Cusick, G. Engleka, A. Flaugh, A. Githens, M. Gorecki, D. Medve, M. Nasim, C. Newman, S. Rudovsky, N. Watson, D. Lee. The Kaiser Permanente Center for Health Research, Portland, Oregon: T. Hillier (principal investigator, E. Harris (co-principal investigator), E. Orwoll (co-investigator), H. Nelson (co-investigator), Mikel Aiken (biostatistician), Jan Van Marter (project administrator), Mary Rix (clinic coordinator), Jane Wallace, Kathy Snider, Kathy Canova, Kathy Pedula, JoAnne Rizzo. This work was financially supported in part by the National Institutes of Health grants CA80668, CA95113, CA57703, CA103730, AG05407, AR35582, AG05394, AR35584, AR35583, and AG000181.

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