A Longitudinal Investigation of Mammographic Density: The Multiethnic Cohort

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Abstract

Mammographic densities are hypothesized to reflect the cumulative exposure to risk factors that influence breast cancer incidence. This report analyzed percent densities over time and explored predictors of density change in relation to age. The study population consisted of 607 breast cancer cases and 667 frequency matched controls with 1,956 and 1,619 mammographic readings, respectively. Mammograms done over >20 years and before a diagnosis of breast cancer were assessed for densities using a computer-assisted method. Using multilevel modeling to allow for repeated measurements, we estimated the effect of ethnicity, case status, reproductive characteristics, hormonal therapy, body mass index, and soy intake on initial status and longitudinal change. After integrating the area under the percent density curve, cumulative percent density was compared with age-specific breast cancer rates in Hawaii. Percent densities decreased ~5.6% per 10 years but a nonlinear effect indicated a faster decline earlier in life. Cumulative percent densities and age-specific breast cancer rates increased at very similar rates; both standardized regression coefficients were >0.9. Japanese ancestry, overweight, estrogen/progestin treatment, and, to a lesser degree, estrogen-only therapy predicted a slower decline in densities with age. Case status and adult soy intake were related to higher densities whereas overweight and having any child were associated with lower densities at initial status. Risk factors that influence the decline in mammographic densities over time may be important for breast cancer prevention because cumulative percent densities may reflect the age-related increase in breast cancer risk. (Cancer Epidemiol Biomarkers Prev 2006;15(4):732–9)

Introduction

In understanding the association between risk factors and breast cancer development (1, 2), mammographic density is considered an intermediate end point because of its strong association with breast cancer (3, 4) and its relation to established risk factors shown in numerous cross-sectional studies (5-7). Although breast cancer risk increases with age, breast density declines with age and menopause (3). As the cyclic proliferative process in the breast comes to an end, mammographic images become increasingly radiolucent due to a higher proportion of fatty tissue (3). Boyd et al. (8) proposed the hypothesis that mammographic densities reflect the cumulative exposure to factors that stimulate growth of breast cells since puberty and influence breast cancer incidence. This idea is based on Pike's model, which proposes a slowing of breast tissue ageing with time and certain reproductive events and provides a good fit with the age-specific incidence curve (9). Among the few longitudinal studies of mammographic densities, a Canadian investigation estimated the average annual reduction in percent density at 1% (10). Percent density decreased by an additional 3% when women who went through menopause were compared with age-matched women who remained premenopausal. The controls in several clinical trials experienced mean annual decreases between 1.9% (11), 1% (12), or less (13, 14). In a Dutch study that used density categories, 28% of women shifted to a lower category after 8 years whereas only 15% of women moved to a higher category (15). In a longitudinal analysis of mammograms done over a 20-year period, we investigated the hypothesis that the area under the curve for age-specific densities may account for the age-related increase in breast cancer incidence (8, 10). In addition, we explored how ethnicity, case status, reproductive characteristics, hormone replacement therapy (HRT), body mass index (BMI), and soy intake influence breast density over time.

Materials and Methods

Study Population. The study subjects for this longitudinal analysis participated in a nested case-control study of mammographic densities and breast cancer risk. As described in detail elsewhere (16), all women were part of the Hawaii component of the Multiethnic Cohort that was formed between 1993 and 1996 and includes 118,441 women in Hawaii and Los Angeles (17). For the nested case-control study, we identified 1,587 potential cases and 1,584 controls that were frequency matched by age and ethnicity (16). After excluding 275 women because of death before 2002, a history of breast cancer or surgery before entry into the cohort, and never having had a mammogram, 2,896 women were eligible to participate. Of these, 1,465 subjects (734 cases and 731 controls) agreed to participate and returned all study forms, but for 191 women, no appropriate mammogram could be obtained. Therefore, the final study population included 607 cases and 667 controls. Despite the relatively low participation rate of 51%, the included women were very similar to the eligible subjects (16). The original cohort and the nested case-control study were approved by the Committee on Human Studies at the University of Hawaii. All subjects provided informed consent to participate in both studies.

Data Collection. Information on demographics, medical history, reproductive behavior, HRT use, and BMI were collected with an extensive questionnaire at entry into the cohort during 1993-1996 (17). A follow-up questionnaire mailed during 1999-2003 provided updated body weight information

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Table 1. Characteristics of the study participants in the nested case-control study

	Case	Control	All
	n (%)	*n* (%)	*n* (%)
No. women	607 (47.7)	667 (52.4)	1,274 (100.0)
Ethnicity			
Hawaiian	79 (13.0)	162 (24.3)	241 (18.9)
Japanese	292 (48.1)	292 (43.8)	584 (45.8)
Caucasian	195 (32.1)	187 (28.0)	382 (30.0)
Other	41 (6.8)	26 (3.9)	67 (5.3)
Born in the United States			
No	51 (8.4)	50 (7.5)	101 (7.9)
Yes	556 (91.6)	617 (92.5)	1,173 (92.1)
Family history of breast cancer			
No	503 (82.9)	587 (88.0)	1,090 (85.6)
Yes	104 (17.1)	80 (12.0)	184 (14.4)
Parous			
No	94 (15.6)	76 (11.5)	170 (13.5)
Yes	508 (84.4)	584 (88.5)	1,092 (86.5)
Mammograms per woman			
1	119 (19.6)	107 (16.0)	226 (17.7)
2	96 (15.8)	332 (49.8)	428 (33.6)
3	159 (26.2)	111 (16.6)	270 (21.2)
4	114 (18.8)	79 (11.8)	193 (15.1)
5+	119 (19.6)	38 (5.7)	157 (12.3)
	Mean (SD)	Mean (SD)	Mean (SD)
Age at recruitment (y)	59.9 (8.4)	57.7 (8.7)	58.7 (8.6)
Age at earliest mammogram (y)	57.0 (9.1)	57.5 (9.5)	57.3 (9.3)
Age at menarche (y)	13.0 (1.5)	13.1 (1.5)	13.1 (1.5)
Age at first live birth (y)	25.0 (4.6)	24.7 (4.5)	24.8 (4.5)
No. children	2.3 (1.6)	2.6 (1.6)	2.5 (1.6)
BMI (kg/m^2)	24.6 (5.0)	25.2 (5.4)	24.9 (5.2)
No. mammograms	3.2 (1.8)	2.4 (1.1)	2.8 (1.5)

for all cohort members. As part of the nested case-control study, a one-page breast health questionnaire elicited information on menopausal status, previous breast surgery, mammograms, and HRT use including the name of the medications, which were classified into estrogen only and combined estrogen/progestin therapy. To estimate lifetime soy intake, information on soy intake during infancy, childhood (1-9 years), adolescence (10-19 years), early adulthood (20-29), and late adulthood (30+ years) was collected (11). Participants marked the annual frequency of usual serving sizes for four categories (tofu, soy beans and sprouts, soy milk and drinks, and other soy products). For infancy, soy-based formula was the only choice. To obtain a summary score for early life and adulthood, we added the frequencies of intake, computed the mean annual intake, and created a binary variable for each period.

Mammograms. The mammographic films were retrieved from clinics located throughout the State of Hawaii using the authorization forms signed by the study subjects. The original cohort study had no records on mammography use except for one item in the baseline questionnaire. At that time, 90% of Caucasian and Japanese women and 75% of Native Hawaiian women reported previous mammography use. Only craniocaudal views were digitized using a Kodak LS 85 Film Digitizer with a pixel size of 260 μm. If available, mammograms for every second or third year were scanned with the goal to cover as wide a time period as possible for each woman. For cases, only mammograms taken before treatment for breast cancer were selected. The scanned images for both breasts were assessed for densities using the Cumulus108 software (18) by one reader (G.M.) who was blinded to case status and time sequence of the mammograms. The films were randomized by subjects: the films within subjects were viewed one after the other, but the temporal order was unknown. This procedure has been shown to be the best method for randomization and viewing of multiple mammograms (19). After the reader determined a threshold for the edge of the breast and for the edge of the dense tissue (18), the computer calculated the total number of pixels in the digitized image that constituted the total area and the dense area and computed the ratio between the two values. Given the high correlation between readings from the right and the left breast, the mean measures were used for analysis when both sides were available, but 689 (19.3%) measures were based on one side only. The reproducibility of the density assessment based on duplicate readings was high; the intraclass correlation coefficient for percent density was 0.974 (95% confidence interval, 0.968-0.978).

Statistical Analysis. Based on the ethnic background reported for both parents, persons with more than one ancestry were classified into a single category, giving first priority to Native Hawaiian, followed by Japanese, then Caucasian, and finally Other (17). Based on the responses from the questionnaire at cohort entry and from the breast health questionnaire at enrollment into the breast density study, we created an HRT use and a menopausal status variable for each year of mammogram. A comparison of the HRT information from the two questionnaires showed good agreement for overlapping years when both questionnaires reported HRT use. To classify the type of HRT, we first used the information from the breast health questionnaire. If a woman indicated that she had used HRT at any time, but the write-in field was empty, we assigned the type of HRT from the cohort questionnaire completed at baseline. For the women with missing HRT type information, we imputed the type based on hysterectomy status (20): estrogen only for women with a hysterectomy and combined therapy otherwise. A similar approach was used for menopausal status. In case we were uncertain about menopausal status in a particular year but had information that a woman had undergone menopause, we assigned perimenopausal status.

We used a multilevel regression model with percent density as the outcome variable to incorporate the repeated readings of

mammographic densities at different points in time. Multilevel modeling allows for an analysis of repeated measures with unbalanced times of measurement (21-24) because this method addresses the dependence of observations in a repeated measurement design by modeling the within-person and between-person variances simultaneously. Subjects contribute to the overall model even if they have only one mammogram. The analyses were done using the Proc Mixed procedure in SAS 9.1 (25, 26). Predictor variables in the final model included age (expressed per 10 years and centered at 32 years to make the regression coefficients easily interpretable), the square of age (nonlinear effect of age), case status, ethnicity (Caucasian, Hawaiian, Japanese, or other), BMI (<22.5, 22.5-24, 25-29, or 30+ kg/m^2), soy intake early in life (none versus any), soy intake as an adult (<36 versus 36+ servings/y), parity (0-1, 2-3, or 4+), age at menarche (<13, 13-14, or 15+ years), age at first live birth (<21, 21-30, >30 years, or no children), menopausal status (premenopausal, 0; perimenopausal, 0.5; or postmenopausal, 1), and HRT use in the year of the mammogram (none, estrogen only, or estrogen plus progestin). The covariates were chosen because they have previously been found to be related to breast cancer (27) and mammographic density (3). Family history of breast cancer and smoking status were not included in our model because they were not significantly associated with mammographic density in this data set. The variables percent density, age, square of age, menopausal status, and HRT use (level 1) were assessed for each year of mammogram and, hence, varied across time. BMI was also treated as time dependent because two measures were available: one at cohort entry and one at follow-up 5 years later. For mammograms done before entry into the cohort or after follow-up contact, the BMI measures from the baseline questionnaire or from the follow-up questionnaire were used. For mammograms done between the two points in time, we interpolated the BMI in a linear fashion. The baseline BMI was used for all mammograms of the 36 women who did not respond to the follow-up questionnaire. All other variables were independent of time (level 2) because they were measured only once at entry into the cohort or were fixed (e.g., ethnicity).

To assess the effects of the covariates longitudinally, interaction terms with age were entered into the model. Therefore, we obtained two regression coefficients for each predictor variable: an estimate for the association of each variable with percent density at age 32, the initial status, and an estimate of the influence of the variable over time. Interactions between the square of age and the other predictors were not significant ($P < 0.05$). As examination of the overall model fit, we calculated pseudo-R^2, the square of the correlation between the actual scores and the model predicted scores. Based on Hawaii Tumor Registry data for 1973-2001 (28, 29), we plotted age-specific breast cancer incidence rates from 30 to 85 years by 5-year age groups. We estimated the area under the percent density curve using integration and plotted percent density years as a measure of cumulative exposure to mammographic density. The slopes of both curves were computed as standardized regression coefficients using linear regression (30).

Results

This study included 607 breast cancer cases and 667 controls. Approximately half of the population was of Japanese ancestry. Of all participants, 1,048 (82%) women had more than one mammogram and close to half of all subjects had

Table 2. Characteristics of mammograms for 1,274 study subjects

	Case	Control	All
	n (%)	*n* (%)	*n* (%)
No. mammograms	1,956 (54.7)	1,619 (45.3)	3,575 (100.0)
Age at mammogram			
<40 y	7 (0.4)	14 (0.9)	21 (0.6)
40-44 y	73 (3.7)	62 (3.8)	135 (3.8)
45-49 y	227 (11.6)	196 (12.1)	423 (11.8)
50-54 y	352 (18.0)	357 (22.1)	709 (19.8)
55-59 y	378 (19.3)	251 (15.5)	629 (17.6)
60-64 y	377 (19.3)	247 (15.3)	624 (17.5)
65-69 y	311 (15.9)	259 (16.0)	570 (15.9)
70-74 y	182 (9.3)	154 (9.5)	336 (9.4)
75+ y	49 (2.5)	79 (4.9)	128 (3.5)
Menopausal status			
Premenopausal	246 (12.6)	390 (24.1)	636 (17.8)
Perimenopausal	163 (8.3)	76 (4.7)	239 (6.7)
Postmenopausal	1,547 (79.1)	1,153 (71.2)	2,700 (75.5)
HRT use			
None	867 (44.3)	754 (46.6)	1,621 (45.3)
Estrogen only	497 (25.4)	525 (32.4)	1,022 (28.6)
Estrogen plus progesterone	592 (30.3)	340 (21.0)	932 (26.1)
	Mean (SD)	Mean (SD)	Mean (SD)
Percent density			
All mammograms	37.0 (22.2)	29.4 (21.7)	33.0 (22.2)
<40 y	59.1 (26.1)	45.1 (26.5)	50.0 (26.6)
40-44 y	59.5 (19.1)	42.6 (24.7)	51.8 (23.3)
45-49 y	49.1 (22.9)	39.3 (24.4)	44.6 (24.1)
50-54 y	41.9 (23.3)	33.4 (22.5)	37.7 (23.3)
55-59 y	36.7 (21.1)	29.1 (20.1)	33.6 (21.0)
60-64 y	32.8 (19.7)	26.4 (21.8)	30.3 (20.8)
65-69 y	31.8 (21.6)	24.7 (21.7)	28.6 (21.9)
70-74 y	31.0 (21.6)	21.9 (17.7)	26.8 (20.4)
75+ y	36.1 (26.3)	24.2 (18.0)	28.9 (22.4)
Premenopausal	50.6 (23.2)	38.7 (25.0)	43.3 (25.0)
Perimenopausal	46.0 (21.7)	36.8 (19.4)	43.1 (21.4)
Postmenopausal	35.1 (22.0)	26.6 (20.8)	31.5 (21.9)

Table 3. Predictors of mammographic density in a multilevel nonlinear growth model

Variable	Initial status (age, 32 y)			Rate of change (per decade)		
	Estimate*	SE	*P*	Estimate	SE	*P*
Intercept	55.00	6.85	<0.0001	—		
Age (linear)	—			−5.63	2.50	0.02
Age^2 (nonlinear)	—			1.64	0.44	<0.0001
Case status						
Control (reference)	0.00			0.00		
Case	10.17	2.68	<0.0001	−1.42	0.88	0.11
Ethnicity						
Caucasian (reference)	0.00			0.00		
Hawaiian	1.73	4.56	0.70	0.18	1.56	0.91
Japanese	−5.62	4.54	0.22	3.79	1.53	0.01
Other	6.67	6.34	0.29	−0.04	2.26	0.99
BMI (kg/m^2)						
<22.5	2.68	2.65	0.31	1.34	0.90	0.14
22.5-24 (reference)	0.00			0.00		
25-29	−11.76	2.69	<0.0001	1.86	0.92	0.04
30+	−21.55	3.87	<0.0001	3.58	1.37	0.01
Age at menarche (y)						
<13	−2.98	4.79	0.53	−0.04	1.56	0.98
13-14	4.28	4.91	0.38	−1.33	1.60	0.41
>14 (reference)	0.00			0.00		
Age at first live birth (y)						
None (reference)	0.00			0.00		
<21	−12.17	6.18	0.05	0.78	2.13	0.71
21-30	−10.88	5.37	0.04	1.58	1.84	0.39
>30	−16.70	6.04	0.01	4.23	2.05	0.04
Parity						
0-1 (reference)	0.00			0.00		
2-3	7.12	4.37	0.10	−2.57	1.46	0.08
≥4	3.42	5.21	0.51	−2.67	1.70	0.12
Menopausal status†	−1.18	3.66	0.75	−1.25	1.65	0.45
HRT						
None (reference)	‡			0.00		
Estrogen only	‡			1.57	0.72	0.03
Estrogen and progestin	‡			3.28	0.76	<0.0001
Soy intake§						
Childhood	−7.55	4.20	0.07	1.59	1.42	0.26
Adulthood	8.94	4.25	0.04	−2.08	1.40	0.14

NOTE: Age, age^2 (the square of age), menopausal status, BMI, and HRT are time-varying variables (level 1). All other variables were measured only once at entry into the cohort (level 2).

*Estimate is the regression coefficient.

†Menopausal status was coded 0 for premenopausal, 0.5 for perimenopausal, and 1 for postmenopausal.

‡Hormone replacement use cannot be estimated before menopause.

§There is no reference group for soy intake because it is not dummy coded (''as child'' and ''as adult'' can both equal 1).

three or more mammograms (Table 1). After combining the readings for the right and left breast, the mean number of density measures was higher for cases than for controls; 3.2 versus 2.4 measures at different times. The mean age in both groups was 57 years at the time of the earliest mammogram and 62 years at the latest mammogram. For cases, the earliest mammogram was taken 6.3 ± 4.0 years before diagnosis. The earliest and the latest mammograms were on average 4.2 years (range, 1-18 years) apart for controls and 5.1 years (range, 1-21 years) for cases. Overall, the time between first and last mammogram was 0 to 1 years for 23% of women, 2 to 5 years for 41% of women, 6 to 10 years for 27% of women, and 11 to 21 years for 9% of women. The mean BMI at cohort entry was 25.4 ± 5.3 kg/m^2 and at follow-up 5.3 years later, it was 25.9 ± 5.8 kg/m^2 with a strong correlation between the two measures (r = 0.88). The mean rate of BMI change per year was 0.094 kg/m^2.

Unadjusted percent densities differed by ~20% between age 40 and 60 (Table 2). We estimated mean percent density at age 32 as 55% and the age-related decline as 5.63% per 10 years (Table 3). When we compared a multilevel model with age as the only predictor to a model that included also the square of age, the change in Deviance statistics (49.4, $P < 0.0001$) suggested that the nonlinear regression line provided a better fit than the linear one (Fig. 1). The nonlinear effect of 1.64% per 10 years in the full model described the faster decline of densities over time earlier in life than later (Table 3). The strongest decline in densities occurred between age 45 and 55; after age 65, breast density changed very little. The mean size

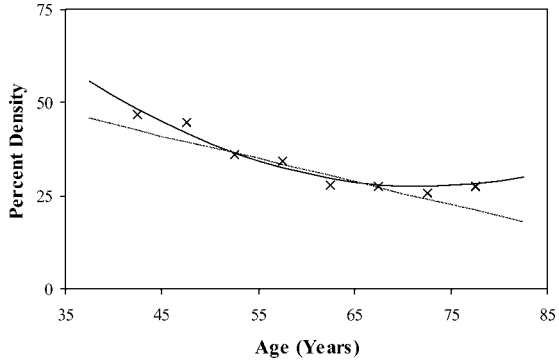

Figure 1. Linear and nonlinear growth models for percent density as a function of age (no covariates included). ×, means for five-year age groups. This nonlinear (*curved line*) growth model provides significantly better fit than the linear (*straight line*) growth model.

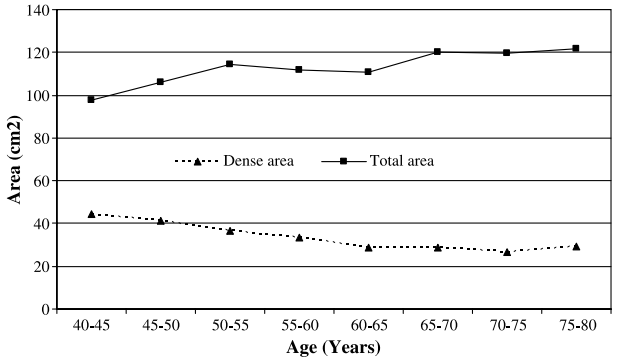

Figure 2. Change of total breast area and dense area with age.

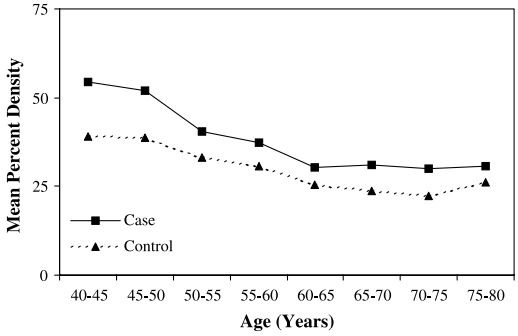

Figure 4. Unadjusted mean percent density as a function of age group and case status. The difference for the age group 45-50 years is significant.

of the total breast area was 25% larger at age 75 to 80 than at age 40 to 45, whereas the size of the dense areas decreased by 34% with age (Fig. 2). Age-specific breast cancer incidence and cumulative percent densities showed very similar increases by age (Fig. 3). The standardized regression coefficient for the breast cancer incidence model was 0.95 and for the cumulative percent density model, 0.99 ($P < 0.001$ for both).

According to the pseudo-R^2, 34% of the variance in percent density was explained by the predictor variables included in the model. The correlation between initial percent density and the rate of change was equal to -0.74 ($P < 0.0001$), suggesting that women with higher initial percent density had a faster rate of decrease in breast density over time. In all age groups, breast cancer cases had higher percent densities than controls (Fig. 4). At initial status, cases had 10.2% higher densities than controls (Table 3) but the rate of change in percent density was not significantly related to case status ($P = 0.11$). Ethnicity and BMI were significantly associated with change in percent density over time (Fig. 5). In comparison with Caucasians, Japanese women experienced a 3.8% ($P = 0.01$) smaller decline in densities for every decade of life than Caucasians but there were no significant differences at initial status. Women with BMI >25 kg/m^2 had lower initial percent density ($P < 0.0001$) than women with lower BMIs. The difference was 11.8% and 21.6% for women with BMI of 25 to 30 kg/m^2 and >30 kg/m^2, respectively. However, overweight and obese women experienced a slower decrease in density over time than women with BMI of <25 kg/m^2 (1.9%, $P = 0.04$ and 3.6%, $P = 0.01$).

Age at menarche was not related to breast density. Having any child was related to initial percent density but there was no significant difference according to age at first live birth. An age at first live birth after age 30 was related to slower rate of decrease in percent density in comparison with having no children ($P = 0.04$). Women with more than one child showed a nonsignificant 2.6% faster decline in densities than women who had 0 to 1 children. The transition of premenopausal to postmenopausal status resulted in a nonsignificant decrease in percent density. However, the estimate was based on only 112 cases and 66 controls that changed from premenopausal to postmenopausal status. HRT use could only be modeled after menopause (Fig. 5). Combined HRT use was related to a significant delay in the decline of densities (3.3%, $P < 0.0001$). Estrogen only use was also associated with a slower decrease but to a much smaller extent than combined HRT (1.6%, $P = 0.03$). Early life soy intake was weakly related to lower percent densities at initial status (-8.6%, $P = 0.07$) whereas soy consumption during adulthood predicted significantly higher densities (8.9%, $P = 0.04$). The rate of change was not significantly affected by soy intake although women who consumed soy as adults seemed to experience a faster decline than women who consumed soy as children.

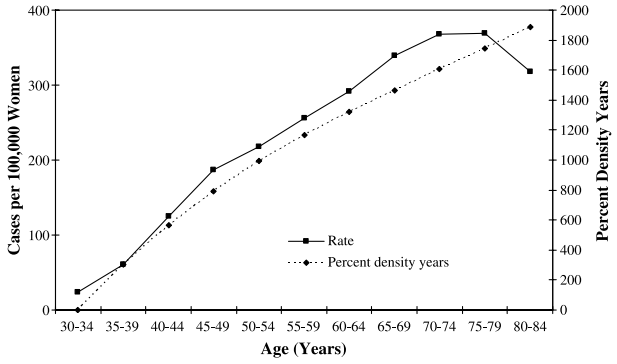

Figure 3. Age-specific breast cancer incidence and cumulative percent density. Based on data from the Hawaii Tumor Registry for 1973-2001 (28, 29).

Discussion

This longitudinal analysis of mammographic densities indicated that cumulative percent densities increase at a very similar rate between age 30 and 85 as breast cancer incidence while no causality can be inferred. Although the two curves in this report were derived from two different populations, the study participants of the case-control study represent the ethnic diversity and risk patterns of Hawaii population from which the breast cancer incidence rates were derived (27, 28). Breast cancer risk remains slightly lower in Hawaii than in the continental United States due the lower risk of some Asian Americans but the incidence rates for Japanese and Native Hawaiians are very similar to Caucasian rates (27, 31, 32). In our model, breast density decreased ~6% in 10 years and menopause led to another 1% to 2% loss in densities with a slower decline in densities with age. Combined HRT and estrogen-only use delayed the decline in densities over time but estrogen-only treatment had only half the effect of combined HRT. In addition to its strong inverse association with breast density, overweight and obesity contributed to a significantly slower decline in densities over time.

The difference of ~20% in densities between 40 and 60 years (Table 2) is very similar as in previous reports (10, 33). The magnitude of change in densities over time was similar to the 1% in the Canadian longitudinal study (10) and the rates described for controls in clinical trials with postmenopausal women: -2.1% over 2 years in a dietary modification study (12), -1.3% over 2 years in a Raloxifene versus estrogen trial

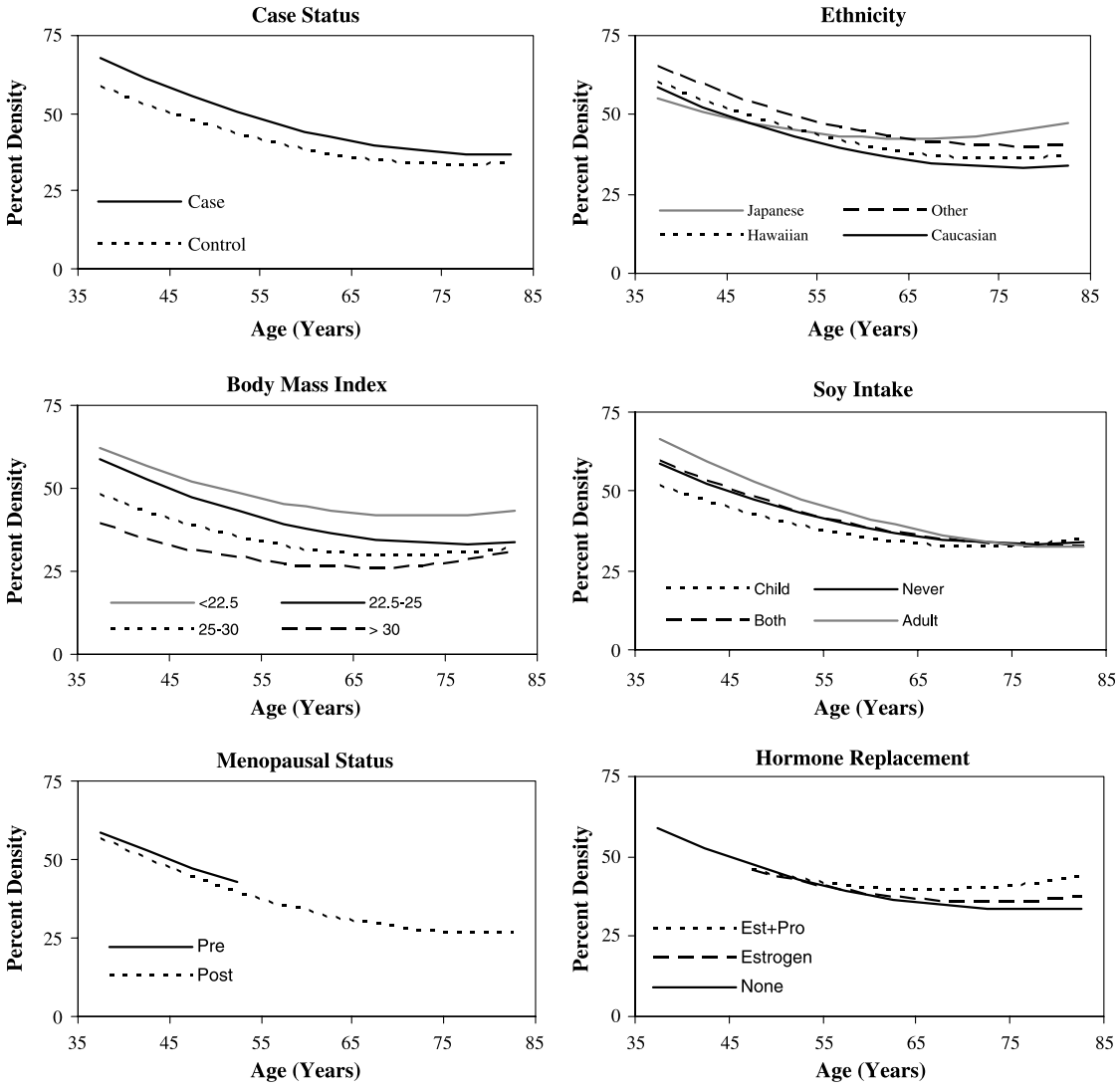

Figure 5. Nonlinear growth models for percent density as a function of age for six predictors. All models include age, square of age, ethnicity, case status, BMI, age at menarche, parity, age at first live birth, menopausal status, HRT use, soy intake in early life and adulthood as covariates.

(13), and −0.5% during 1 year in a large HRT trial (14). Not surprisingly, the decline described for premenopausal women (−1.9% per year; ref. 11) was higher than in the current study. Our findings disagree with a report that observed the greatest change in densities between age 55 and 64 (34) but the density assessment in that analysis was based on qualitative Breast Imaging Reporting and Data System categories (35). We were also not able to detect the distinct change related to menopause (3%) described by Boyd et al. (10) because we did not know the exact year of menopause. The determinants of mammographic densities observed in this report are very similar to those of other studies although we found no association with age at menarche and number of children (3, 5-7, 36).

Japanese ancestry was associated with lower densities at initial status, but a slower decline in densities with age. It is possible that the changing breast cancer risk across the different generations of Japanese migrants is responsible for the opposite direction of the two variables (37). Nevertheless, in agreement with the observation of similar breast cancer risk during recent years (27, 32), the cumulative exposure to breast density would not be very different in Japanese than in Caucasians. One possible explanation for the paradox of lower percent densities among women with high BMI despite the greater breast cancer risk associated with a higher body weight is that the effects of postmenopausal obesity are not mediated by breast density (3, 38). The novel observation from our multilevel model is that women with higher BMI experience a slower decline in mammographic densities, thereby acquiring a higher cumulative risk despite lower densities at initial status. The findings are also compatible with the epidemiologic evidence that a higher body weight before menopause is protective against breast cancer whereas overweight among postmenopausal women increases risk (2, 39).

The observation that estrogen-only therapy significantly delays the decline in breast density, although to a lesser degree than combined HRT, is an important finding. Results from two clinical trials indicated a nonsignificant increase of 1.2% and 1.3% due to estrogen-only therapy (13, 14). Our significant result is probably due to the longer observation period. The strong relation with combined HRT is consistent with the Postmenopausal Estrogen/Progestin Interventions trial that described 3% to 5% higher densities over 2 years among women taking different progestin-containing medications. The slower change to favorable density patterns with age when women are taking HRT has also been described by several studies using qualitative density categories (34, 40, 41). The fact that, in a cross-sectional investigation, HRT use showed a stronger association with breast density at an older age than

among younger women supports the idea of a cumulative effect (6).

Soy intake during childhood and adulthood had slightly different relations with breast density. Women with soy intake during adulthood only had the highest densities and the steepest decline whereas women with only childhood consumption had the lowest densities and the smallest decline (Fig. 5). Interestingly, low soy intake during early life and high soy intake during adulthood also predicted a greater reduction in percent density during a 2-year trial among premenopausal women (11), but this may have been a chance finding due to uncontrolled confounders. Although we used the lifetime soy questionnaire in this previous study, we are uncertain about its validity.

This study had a number of serious limitations, foremost the collection of information on BMI and HRT by self-report. Although the questionnaire asked for HRT use by year, this information may have been affected by recall bias. Because each woman only recorded one type of HRT in the breast health questionnaire, we may have missed a change from one type of HRT to another. BMI was available for two points in time 5 years apart (approximately 1995 and 2000) but a large proportion of mammograms was done during earlier years for which BMI information was not collected. Given the high correlation of the two BMI measures and the small rate of change over 5 years, we are confident that BMI did not change dramatically in this population. The age range of Multiethnic Cohort participants limited our ability to investigate changes in breast density among premenopausal women; only 17.3% of all mammograms were performed before menopause. Given the unfavorably risk/benefit of mammography for young women, our calculation of cumulative exposure had to omit earlier years when breast density is highest. Selection bias may also have affected our results. In addition to the relatively low participation rate in the nested case-control study, we did not locate an equal number of screening mammograms for all subjects. It is possible that women with higher mammography use experienced different changes in density patterns over time than subjects who did not receive mammograms, but, given the high self-reported mammography use at baseline, bias from this source seems unlikely.

Because the amounts of epithelial and stromal tissue as represented by the dense areas probably determine breast cancer risk (3), the size of the dense areas may provide more relevant information than percent density which is strongly influenced by body weight (38, 42-44). Nonetheless, as Fig. 2 indicates, the decrease in the size of the dense areas with age is similar to that for percent densities (Fig. 1). The somewhat larger decline in percent densities is due to the age-related softening of breast tissue, allowing greater compression during mammography that leads to an increase in total breast area on mammographic images. In a very detailed analysis of absolute versus relative density (44), it has been proposed that percent density may be the more appropriate measure when assessing breast risk because it incorporates the effects of body weight. Higher amounts of fatty tissue surrounding the dense areas may increase breast cancer risk through aromatization of androgens to estrogens (45) and lipid peroxidation (46).

On the other hand, our study had considerable strengths. The availability of multiple mammographic images over close to 20 years provided a stable exposure measure and allowed us to explore density changes prospectively. Four or more mammograms were available for 350 women covering 8.9 years and three films for another 270 women spanning 5.8 years. Although the quality of mammographic images changed during the study period due to modifications in mammographic techniques and film processing, we did not observe a significant effect of year when the mammogram was performed after including an indicator variable into the model. This suggests that the assessment method adjusts adequately for differences in the brightness and quality of films. Reading errors in density assessment are of concern, but the correlation coefficients for repeated readings in this and other studies indicate high reliability (47).

The results of this longitudinal investigation agree with the hypothesis that cumulative breast density reflects exposure to risk factors that predict age-specific breast cancer incidence (8) although a causal relation cannot be ascertained through an observational study. Because cumulative exposure to density and breast cancer incidence are both almost linear functions of age, the relations are expected to look similar. The adverse effects of overweight and HRT use on the decline in densities correspond to the higher breast cancer risk associated with these factors (48, 49). The possible mechanisms of action for risk factors to affect breast density include hormones and growth factors that may stimulate cell growth and division in the breast (8). We cannot conclude from our data whether the rate of change in densities is associated with the development of breast cancer because, in the multilevel model (Table 3), case status was not related to change in densities over time. Nevertheless, risk factors that influence the rate of decline in mammographic densities over time may be important for breast cancer prevention because cumulative percent densities reflect the age-related increase in breast cancer risk.

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