Common Breast Cancer Susceptibility Variants in LSP1 and RAD51L1 Are Associated with Mammographic Density Measures that Predict Breast Cancer Risk

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**Abstract**

**Background:** Mammographic density adjusted for age and body mass index (BMI) is a heritable marker of breast cancer susceptibility. Little is known about the biologic mechanisms underlying the association between mammographic density and breast cancer risk. We examined whether common low-penetrance breast cancer susceptibility variants contribute to interindividual differences in mammographic density measures.

**Methods:** We established an international consortium (DENSNP) of 19 studies from 10 countries, comprising 16,895 Caucasian women, to conduct a pooled cross-sectional analysis of common breast cancer susceptibility variants in 14 independent loci and mammographic density measures. Dense and nondenst areas, and percent density, were measured using interactive-thresholding techniques. Mixed linear models were used to assess the association between genetic variants and the square roots of mammographic density measures adjusted for study, age, case status, BMI, and menopausal status.

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Common Breast Cancer Susceptibility Variants and Mammographic Density

Introduction

Genetic factors play a major role in the pathogenesis of breast cancer (1–3). Recent multistage genome-wide association studies (GWAS) and candidate gene studies conducted by several groups, including the Breast Cancer Association Consortium (BCAC), have successfully identified and replicated associations between over 18 single-nucleotide polymorphisms (SNP) and risk of breast cancer in Caucasians (4–9).

Mammographic density, which reflects variations in the amounts of fat, stromal, and epithelial tissues in the breast, is one of the strongest risk factors for breast cancer with risk being 4- to 6-fold higher for women in the highest relative to lowest density categories after adjusting for age and body mass index (BMI; refs. 10, 11). The biology underlying the mammographic density and breast cancer association is essentially unknown, but twin and family studies suggest that additive genetic factors explain about 60% of variance in the density measures (12, 13). This raises the question of whether breast cancer susceptibility variants identified to date are associated with mammographic density measures. This could lead to new insights into the etiology of breast cancer by revealing the biologic reasons for these associations with breast cancer risk (14).

Five studies have examined the association of breast cancer susceptibility SNPs with age- and BMI-adjusted measures of mammographic density (14–18). The most consistent finding was an association between (lymphocyte-specific protein-1, LSP1)-rs3817198 and adjusted dense area and percent density, in the same direction as the association with breast cancer. The association was observed overall by Odefrey and colleagues (17) but only in specific subgroups by others: in premenopausal women (14), current users of postmenopausal hormones (PMH; ref. 15) or estrogen receptor (ER)+/progesterone receptor (PR)+ cases only (16). Other nominally significant reported SNP–density associations consistent with the association of these SNPs with breast cancer risk include associations of TOX3-rs12443621 (14, 15) and rs4666451 (14) with adjusted percent density, in premenopausal women only, and rs13281615 at 8q24 with both adjusted percent density and dense area (17). The largest study to date, a meta-analysis of 5 GWAS of mammographic density involving 4,877 women with and without breast cancer, identified a genome-wide significant association between ZNF365-rs10995190, a known breast cancer susceptibility SNP, and adjusted percent density as well as weak evidence of possible associations with ERα(rs2046210 (P = 0.005) and LSP1-rs3817198 (P = 0.04; ref. 18).

Only one previous study (17), however, examined the SNP associations with the components that comprise the percent density phenotype, namely, dense area and nondense area. Dense area has been hypothesized to be the more relevant density phenotype for understanding the etiology of mammographic density (19), as tumors have been shown to arise within the radiodense tissue (20). Whether these SNPs influence dense and/or nondense area could help to interpret the mechanism by which the loci influence density and possibly cancer.

Results: Consistent with their breast cancer associations, the C-allele of rs3817198 in LSP1 was positively associated with both adjusted dense area (P = 0.00005) and adjusted percent density (P = 0.001), whereas the A-allele of rs10483813 in RAD51L1 was inversely associated with adjusted percent density (P = 0.003), but not with adjusted dense area (P = 0.07).

Conclusion: We identified two common breast cancer susceptibility variants associated with mammographic measures of radiodense tissue in the breast gland.

Impact: We examined the association of 14 established breast cancer susceptibility loci with mammographic density phenotypes within a large genetic consortium and identified two breast cancer susceptibility variants, LSP1-rs3817198 and RAD51L1-rs10483813, associated with mammographic measures and in the same direction as the breast cancer association. Cancer Epidemiol Biomarkers Prev; 21(7): 1156–66. ©2012 AACR.
We established an international collaboration—the DENSNP consortium—of studies with data on established breast cancer susceptibility variants and quantitative density measures from film mammography to conduct analyses of breast cancer susceptibility SNPs in relation to the 3 density phenotypes. This article reports the findings for 15 breast cancer SNPs at 14 loci, identified through 2009 when the DENSNP consortium was established.

Materials and Methods

Study samples

The DENSNP consortium comprises 19 studies from Europe, North America, and Australia with the present analyses restricted to Caucasian women. Individual studies, their design, and sample sizes are described in Supplementary Table S1. Covariate data, including age, reproductive variables, and exogenous hormone use, were obtained through self-administered postal questionnaires (12 studies), in-person interviews (6 studies), or telephone interviews (one study; Supplementary Table S2). Participants' weights, heights, and hence BMIs were measured by trained staff (10 studies) and self-reported (9 studies). For 8 studies, there was an average of 6 months or less between mammography and collection of participant information; for 18 studies, the average was 3 years or less.

Each study obtained informed consent and relevant ethics and institutional approvals. Only anonymized data were made available to the DENSNP consortium.

Digitization and density measures

All studies obtained film mammograms—either the mediolateral oblique (MLO; 7 studies) or craniocaudal (CC; 12 studies) views—for participants, including breast cancer cases and/or noncases, except PNS which digitized copies of digital mammograms (Supplementary Table S3). For cases, the film from the unaffected contralateral breast taken at the time of cancer diagnosis was used, except for 3 nested case-control studies for which images obtained before diagnosis were used (2 studies used average measurements of both breasts; 1 study used only the right breast). For noncases, both breasts (averaged), left or right only, or the side that corresponded to the matched case was chosen.

As a requirement for entry, participating studies contributed percent density, dense area, and nondense area measures for cases and/or noncases, except PNS which digitized copies of digital mammograms (Supplementary Table S3). For cases, the film from the unaffected contralateral breast taken at the time of cancer diagnosis was used, except for 3 nested case-control studies for which images obtained before diagnosis were used (2 studies used average measurements of both breasts; 1 study used only the right breast). For noncases, both breasts (averaged), left or right only, or the side that corresponded to the matched case was chosen.

A test of the null hypothesis of no association between any of the tested SNPs and a given mammographic measure was conducted using Fisher's method (23). As individual-level data were available from all studies, primary analyses used a mixed model approach that included per-study random-effects to capture study-specific differences. When applicable, a repeated measures adjustment within families assuming a compound symmetry correlation structure was used to account for familial correlation. Models were adjusted for the fixed-effects of age (continuous), BMI (1/BMI, was used as it provided a better fit), case status, and menopausal status (pre- and
perimenopausal combined vs. postmenopausal, with the latter defined as no menstruation for ≥12 months, not due to pregnancy). A missing category was included, when applicable. Primary analyses considered SNP associations as additive genetic effects, by defining an ordinal covariate as the number of copies of the minor allele carried by the study subjects and fitted a linear association. The resulting estimate of the per-allele effect is reported as the "additive estimate" in the tables. Estimates of the adjusted mean mammographic density measures and their 95% confidence intervals (CI), corresponding to the observed genotypes of each variant, were derived by back-transformation from the square root to the original scale. Additional analyses were conducted within subsets of women defined by menopause categories (pre- and perimenopausal combined vs. postmenopausal), BMI (< vs. ≥ median of 25 kg/m²), PMH (ever vs. never use), and case status to assess whether SNP-density phenotype associations were modified by these variables.

Between-study heterogeneity was tested by fitting study-by-genotype interactions.

Figure 1. Associations of common breast cancer susceptibility variants with adjusted percent mammographic density, dense area, and nondense area.
Analyses were conducted using SAS version 9.2 (SAS Institute, Inc.). Two-sided \( P \) values were calculated. A Bonferroni adjustment to account for multiple testing was applied to define the threshold for statistical significance as \( P \leq 0.003 \) (\( 0.05 \) divided by 14 loci).

**Results**

There were 5,110 breast cancer cases and 11,785 non-cases of self-reported Caucasian race/ethnicity with available density phenotypes, risk factors, and at least 1 of the 15 SNPs considered (Table 1). The number of participants varied by SNP with the most comprehensive information for 2q35 \((n = 13,254)\), CASP8 \((n = 12,816)\), and FGFR2 \((n = 12,680)\), and least information for TGFB1 \((n = 3,099)\), RAD51L1 \((n = 7,610)\) and ESRI \((n = 8,274)\).

The majority of the participants were aged \( \geq 40 \) years (98%) and postmenopausal (77%), and approximately half of those aged \( \geq 55 \) reported ever using PMH (48%; Table 1). In all, 44% of participants had a BMI \(< 25 \text{ kg/m}^2\) (Table 1). A small proportion was nulliparous \((20\%)\).

As an example, the association between rs3817198 (LSP1) and density is shown in Fig. 1 and described in Supplementary Tables S5a–S5c. Pictured are the parameter estimates from the model, along with the 95% confidence intervals. The genetic associations above did not diminish after further adjustment for parity or view (data not shown) and, in general, did not appear to differ by case status, BMI, menopausal status, or PMH use (Supplementary Tables S6a–S6c) but the study had low power to examine interactions.

We also examined the association of these SNPs with breast cancer risk before and after adjustment for the density measures by pooling data from studies that recruited both cases and noncases (identified in Supplementary Table S1). Using 3,175 cases and 6,504 noncases from 8 studies, the per C-allele OR for rs3817198 (LSP1) was 1.04 (95% CI, 0.97–1.12) without adjustment for either density measure. When including dense area as a covariate, the OR was 1.03 (95% CI, 0.96–1.10), and after adjustment for percent density instead, the OR was 1.02 (95% CI, 0.95–1.11). Similarly, using 2,765 cases and 3,022 noncases from 4 studies, the per A-allele OR for rs10483813 (RAD51L1) was 0.92 (95% CI, 0.84–1.00) without adjustment for either density measure, 0.93 (95% CI, 0.85–1.01) after adjustment for dense area, and 0.94 (95% CI, 0.86–1.03) after adjustment for percent density.

**Discussion**

There is wide interindividual variability in mammographic density measures, but known epidemiologic risk factors account for only 20% to 30% variability in percent density (13, 24, 25). We hypothesized that common low-penetrance breast cancer susceptibility variants contribute to the remaining interindividual differences in the density phenotypes and examined this within a large international consortium (DENSNP). Here, we report the first findings from this collaborative effort and identify associations between adjusted measures of density and 2 breast cancer susceptibility SNPs, rs3817198 (LSP1) and rs10483813 (RAD51L1), which were in the same direction as the corresponding SNP associations with cancer risk.

The most marked association with density was with rs3817198 (LSP1). We also confirmed this association using the 10 studies that had not previously published on the LSP1 variant and density association, providing consistent evidence for this mammographic density locus. The mechanisms through which this SNP (or more likely the causal allele(s) it tags) may affect density and cancer risk are unclear. The LSP1 gene encodes an intracellular F-actin-binding protein, which is expressed in lymphocytes, neutrophils, and endothelium and might regulate neutrophil motility, adhesion to fibronectin matrix proteins, and transendothelial migration (26).
The SNP rs10483813 in RAD51L1, a gene on chromosome 14q24.1 involved in the double-strand DNA repair and homologous recombination pathway, may also be associated with the adjusted density measures, although the evidence is less compelling than for rs3817198 (LSP1). The biologic mechanisms underlying the possible association of this variant with density and cancer risk are unknown. RAD51L1 interacts with RAD51, and a SNP in the 5' untranslated region of RAD51 has been found to be associated with breast cancer risk for BRCA2 mutation.

Table 1. Summary characteristics of the 19 DENSNP studies

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Category</th>
<th>No. of studies</th>
<th>BC cases</th>
<th>Noncases</th>
<th>Overall</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>N (%)</td>
<td>N (%)</td>
<td>N (%)</td>
</tr>
<tr>
<td>Overall</td>
<td></td>
<td>19</td>
<td>5,110 (30)</td>
<td>11,785 (70)</td>
<td>16,895 (100)</td>
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<td>Study design</td>
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<td>16 (0.3)</td>
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<td>Cross-sectional</td>
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<td>38 (1)</td>
<td>3,064 (26)</td>
<td>3,102 (18)</td>
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<td>Case-control</td>
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<td>3,280 (64)</td>
<td>2,217 (19)</td>
<td>5,497 (33)</td>
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<td></td>
<td>Nested case-control</td>
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<td>1,599 (31)</td>
<td>2,099 (18)</td>
<td>3,698 (22)</td>
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<td></td>
<td>Family-based</td>
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<td>177 (3)</td>
<td>2,823 (24)</td>
<td>3,000 (18)</td>
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<tr>
<td>Source of demographic and reproductive data</td>
<td>Postal questionnaire</td>
<td>12</td>
<td>3,378 (66)</td>
<td>8,831 (75)</td>
<td>12,209 (72)</td>
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<td>101 (2)</td>
<td>1,678 (14)</td>
<td>1,779 (11)</td>
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<td>Age, a y</td>
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<td>221 (4)</td>
<td>145 (1)</td>
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<td>40–49</td>
<td>17</td>
<td>937 (18)</td>
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<td>2,794 (17)</td>
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<td>50–59</td>
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<td>6,486 (38)</td>
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<td>60–69</td>
<td>16</td>
<td>1,659 (32)</td>
<td>4,011 (34)</td>
<td>5,670 (34)</td>
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<td>&gt;70</td>
<td>13</td>
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<td>929 (8)</td>
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<td>Paritya</td>
<td>Nulliparous</td>
<td>19</td>
<td>614 (12)</td>
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<td>1,781 (11)</td>
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<td>10,479 (89)</td>
<td>14,808 (88)</td>
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<td>Unknown</td>
<td>8</td>
<td>167 (3)</td>
<td>139 (1)</td>
<td>306 (2)</td>
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<td>Menopausal statusa</td>
<td>Premenopausal</td>
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<td>1,185 (23)</td>
<td>2,241 (19)</td>
<td>3,426 (20)</td>
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<td>13 (0.2)</td>
<td>251 (2)</td>
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<td>143 (3)</td>
<td>98 (1)</td>
<td>241 (1)</td>
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<td>PMH use (at age ≥55)</td>
<td>Ever</td>
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<td>1,703 (53)</td>
<td>3,364 (48)</td>
<td>5,067 (48)</td>
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<tr>
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<td>16</td>
<td>1,326 (41)</td>
<td>3,474 (47)</td>
<td>4,800 (45)</td>
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<tr>
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<td>Unknown</td>
<td>8</td>
<td>178 (6)</td>
<td>537 (7)</td>
<td>715 (7)</td>
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<td>Measurements by trained staff</td>
<td>10</td>
<td>1,326 (26)</td>
<td>5,876 (50)</td>
<td>7,202 (43)</td>
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<td></td>
<td>BMI, a kg/m²</td>
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<tr>
<td></td>
<td>&lt;25</td>
<td>19</td>
<td>2,284 (45)</td>
<td>5,071 (43)</td>
<td>7,355 (44)</td>
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<tr>
<td></td>
<td>≥ 25</td>
<td>19</td>
<td>2,737 (54)</td>
<td>6,597 (56)</td>
<td>9,334 (55)</td>
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<td>10</td>
<td>89 (2)</td>
<td>117 (1)</td>
<td>206 (1)</td>
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<tr>
<td>Average time interval between mammography and data collection (months), b</td>
<td>&lt;6</td>
<td>8</td>
<td>2,129 (42)</td>
<td>4,330 (37)</td>
<td>6,459 (38)</td>
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<td></td>
<td>&gt;6</td>
<td>11</td>
<td>2,981 (58)</td>
<td>7,455 (63)</td>
<td>10,436 (62)</td>
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<td>Mammographic side, view</td>
<td>L-CC</td>
<td>8</td>
<td>831 (16)</td>
<td>2,547 (22)</td>
<td>3,378 (20)</td>
</tr>
<tr>
<td></td>
<td>R-CC</td>
<td>9</td>
<td>949 (19)</td>
<td>1,830 (16)</td>
<td>2,779 (16)</td>
</tr>
<tr>
<td></td>
<td>LR average–CC</td>
<td>3</td>
<td>2,402 (47)</td>
<td>2,285 (19)</td>
<td>4,687 (28)</td>
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<td>L–MLO</td>
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<td>465 (9)</td>
<td>1,978 (17)</td>
<td>2,443 (14)</td>
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<tr>
<td></td>
<td>R–MLO</td>
<td>1</td>
<td>447 (9)</td>
<td>418 (4)</td>
<td>865 (5)</td>
</tr>
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<td></td>
<td>LR average–MLO</td>
<td>4</td>
<td>16 (0.3)</td>
<td>2,727 (23)</td>
<td>2,743 (16)</td>
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<tr>
<td>Density reading software</td>
<td>Cumulus</td>
<td>15</td>
<td>3,814 (75)</td>
<td>10,213 (87)</td>
<td>14,027 (83)</td>
</tr>
<tr>
<td></td>
<td>Madena</td>
<td>4</td>
<td>1,296 (25)</td>
<td>1,572 (13)</td>
<td>2,868 (17)</td>
</tr>
</tbody>
</table>

Abbreviations: BC, breast cancer; CC, craniocaudal; L, left; MLO, mediolateral oblique; R, right.

aAt time of mammography and/or data collection.
bAverage time interval for each study given in Supplementary Table S2 (range, 0–5 years).
mediate the association between finding an association suggests that density is unlikely to if such a difference truly exists, and therefore the lack of between homozygote carriers and noncarriers of this SNP, an average difference in percent density of less than 1% breast cancer risk (30). Our study had 90% power to detect rs2981582 being associated with about a 26% increased receptor 2 (FGFR2), with each copy of the T-allele of specified polymorphisms in intron 2 of fibroblast growth factor receptor 2 (FGFR2), identifying breast cancer GWAS have consistently identified polymorphisms in intron 2 of fibroblast growth factor receptor 2 (FGFR2), with each copy of the T-allele of ranging from 578 (ref. 16) to 4,877 (ref. 18), which could Previous studies were based on smaller sample sizes carriers (27). However, mutations in BRCA1 and BRCA2 have not been found to be associated with the density phenotypes (28, 29). Several breast cancer GWAS have consistently identified polymorphisms in intron 2 of fibroblast growth factor receptor 2 (FGFR2), with each copy of the T-allele of rs2981582 being associated with about a 26% increased breast cancer risk (30). Our study had 90% power to detect an average difference in percent density of less than 1% between homozygote carriers and noncarriers of this SNP, if such a difference truly exists, and therefore the lack of finding an association suggests that density is unlikely to mediate the association between FGFR2 and breast cancer risk. Similar considerations apply to SNPs in several other breast cancer loci, including TOX3-rs3803662, 2q35-rs13387042 and MAP3K1-rs889312. These loci are likely to contribute independently of density to risk prediction. In fact, when we added LSP1-rs1387198 and RAD51L1-rs1048313 to a risk model with age, BMI, menopause, study, and percent density, the inclusion of these 2 SNPs did not affect the area under the curve whereas the addition of the remaining 12 SNPs increased the area under the curve from 0.62 to 0.65 (P < 0.001). Previous studies were based on smaller sample sizes ranging from 578 (ref. 16) to 4,877 (ref. 18), which could have precluded the detection of small effects. Our study is

<table>
<thead>
<tr>
<th>Risk factor Categories</th>
<th>N (%)</th>
<th>PD (%) Mean (95% CI)</th>
<th>Dense area, cm² Mean (95% CI)</th>
<th>Nondense area, cm² Mean (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;40</td>
<td>366 (2.2)</td>
<td>34.2 (30.3–38.3)</td>
<td>36.8 (31.9–42.1)</td>
<td>75.1 (66.8–83.8)</td>
</tr>
<tr>
<td>40–49</td>
<td>2,794 (16.5)</td>
<td>28.2 (25.3–31.4)</td>
<td>33.0 (29.1–37.1)</td>
<td>89.7 (82.9–96.8)</td>
</tr>
<tr>
<td>50–59</td>
<td>6,486 (38.4)</td>
<td>20.3 (17.9–22.9)</td>
<td>26.4 (23.0–30.0)</td>
<td>112.2 (104.8–119.8)</td>
</tr>
<tr>
<td>60–69</td>
<td>5,670 (33.6)</td>
<td>14.9 (12.8–17.2)</td>
<td>21.3 (18.2–24.6)</td>
<td>130.2 (122.2–138.4)</td>
</tr>
<tr>
<td>≥70</td>
<td>1,579 (9.3)</td>
<td>13.0 (11.0–15.2)</td>
<td>17.3 (14.5–20.4)</td>
<td>143.0 (134.1–152.3)</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;25</td>
<td>7,355 (44.1)</td>
<td>25.8 (23.2–28.6)</td>
<td>27.0 (23.6–30.7)</td>
<td>82.9 (77.1–89.0)</td>
</tr>
<tr>
<td>≥25</td>
<td>9,334 (55.9)</td>
<td>14.8 (12.8–16.9)</td>
<td>23.3 (20.1–26.7)</td>
<td>144.3 (136.6–152.3)</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Menopausal status</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pre- or perimenopausal</td>
<td>3,690 (22.2)</td>
<td>21.5 (19.1–24.1)</td>
<td>27.1 (23.6–30.8)</td>
<td>113.5 (106.4–120.9)</td>
</tr>
<tr>
<td>Postmenopausal</td>
<td>12,964 (77.8)</td>
<td>18.4 (16.2–20.7)</td>
<td>24.1 (20.9–27.5)</td>
<td>116.3 (109.3–123.5)</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>0.05</td>
</tr>
<tr>
<td>PMH use (at ages ≥55)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never</td>
<td>4,800 (48.6)</td>
<td>14.6 (12.5–16.9)</td>
<td>20.2 (16.7–23.9)</td>
<td>129.1 (120.4–138.2)</td>
</tr>
<tr>
<td>Ever</td>
<td>5,067 (51.4)</td>
<td>17.8 (15.5–20.4)</td>
<td>23.6 (19.9–27.7)</td>
<td>122.7 (114.2–131.6)</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Parity</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nulliparous</td>
<td>1,781 (10.7)</td>
<td>22.6 (20.1–25.2)</td>
<td>29.0 (25.4–32.9)</td>
<td>109.2 (102.2–116.4)</td>
</tr>
<tr>
<td>Parous</td>
<td>14,808 (89.3)</td>
<td>18.7 (16.5–21.0)</td>
<td>24.3 (21.1–27.7)</td>
<td>116.7 (109.8–123.8)</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mammographic view</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CC</td>
<td>6,051 (35.8)</td>
<td>17.7 (14.2–21.5)</td>
<td>25.1 (19.7–31.1)</td>
<td>122.4 (111.1–134.2)</td>
</tr>
<tr>
<td>MLO</td>
<td>10,844 (64.2)</td>
<td>20.1 (17.3–23.2)</td>
<td>24.8 (20.6–29.4)</td>
<td>111.5 (103.2–120.2)</td>
</tr>
<tr>
<td>P</td>
<td>0.3</td>
<td>0.9</td>
<td>0.1</td>
<td></td>
</tr>
<tr>
<td>Case status</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BC case</td>
<td>4,530 (37.8)</td>
<td>24.5 (20.8–28.4)</td>
<td>30.0 (24.1–36.4)</td>
<td>108.2 (95.6–121.5)</td>
</tr>
<tr>
<td>Noncase</td>
<td>7,439 (62.2)</td>
<td>19.3 (16.0–22.8)</td>
<td>24.2 (19.0–30.1)</td>
<td>117.9 (104.9–131.7)</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Abbreviations: BC, breast cancer; CC, craniocaudal; MLO, mediolateral oblique.

<ref>Adjusted for study.</ref>
<ref>Adjusted for study and age.</ref>
<ref>Adjusted for study, age, and BMI.</ref>

<ref>Restricted to 9 studies that recruited both cases and noncases and adjusted for study, age, and BMI.</ref>
the largest conducted so far with sample sizes greater than 6,000 for all but one SNP and greater than 10,000 for all but 5 SNPs. We had more than 90% power to detect per-allele differences in adjusted percent density of 1% or less for all but 3 SNPs (rs17468277, rs10483813, and rs4415084), and even for these SNPs, we were similarly powered to detect per-allele differences of less than 2%. However, limited power precluded a more detailed examination of inter-
actions with BMI (e.g., differential SNP effects in BMI-
defined quartiles) and PMH use (e.g., different SNP effects
by type of PMH, recency of use). The study also had low
power to assess the mediation of the SNP and breast
cancer associations by density.

The mammographic density readings were conducted
in different sets of films (e.g., left, right, or both breasts; CC
or MLO views), but it is unlikely that this may have
substantially affected our findings because there is a high
correlation between a woman’s density measurements
taken from the various breast view combinations (31).
For cases, both prediagnostic films and films from the
unaffected breast at the time of diagnosis, but before
treatment, were used—an approach used by others (10);
however, our findings were not modified by case
status. One small study (PNS) used digitized copies of
digital mammograms, but its exclusion did not affect the
results shown here. Although mammographic density
readings were not standardized, all studies used a similar
interactive threshold approach and had very high within-
and between-observer repeatability (typically >90%;
ref. 32). Also, all analyses were adjusted for study hence
minimizing the impact of any between-study differences
on density measurements which would have likely
reduced our power to detect real associations. Reassur-
ingly, we were able to reproduce the well-established
influences of age, BMI, parity, menopausal status, and
PMH on density phenotypes within each one of the
participating studies as well as in joint analyses.

Our findings suggest that 2 of 14 well-established breast
cancer loci may contribute to the large between-woman
differences in risk-predicting density phenotypes, consis-
tent with estimates of 5% to 10% genetic overlap between
this biomarker and breast cancer (33). The 2 common
variants in LSP1 and RAD51L1 explained 0.2% (combined,
0.1% for each) of the variance in adjusted percent density
and dense area, although the overall contribution could be
larger if the true causal variants are more strongly

![Figure 2. Study-specific associations of LSP1-rs3817198 and RAD51L1-rs10483813 with adjusted percent mammographic density and dense area.](image-url)
associated with density than the tagging SNPs we examined here. At the individual level, these SNPs were associated with a 0.6% absolute increase in percent density per allele for LSP1 and 0.8% absolute decrease in percent density per allele for RAD51L1. These magnitudes can be compared with, for example, the change in density measures of 1% decrease per year of ageing (34), 2% increase with use of PMH, and 2% decrease over the menopausal transition (35). Our findings are consistent with the hypothesis that mammographic density is likely a polygenic trait, influenced by many common low-penetrance variants, and/or rarer variants with larger effects which cannot be identified through current GWAS. Identification of such variants, and clarification of their role and function, is likely to improve our understanding of the biology of mammographic density and how this phenotype is associated with breast cancer risk.

Disclosure of Potential Conflicts of Interest

M. Pollan is the principal investigator of one of the studies included in this analysis (DDM-Spain). M.O. Leach has employment (other than primary affiliation; e.g., consulting) from Specialty Scanners PLC as Director and Ownership Interest (including patents) and is named on patents that relate to breast analysis and density measurements. If these are commercialized by the Institute of Cancer Research, then he may receive compensation under the rewards for inventors scheme. D.F. Easton is a Primary Research Fellow of Cancer Research UK. J.L. Hopper is an Australian Fellow of the NHMRC and a Victorian Breast Cancer Research Consortium (VBCRC) Group Leader. No potential conflicts of interest were disclosed by other authors.

LIFE: The ideas and opinions expressed herein are those of the authors, and no endorsement by the State of California, Department of Health Services is intended or should be inferred.

MCBC: M.C. Southey is a National Health and Medical Research Council Senior Research Fellow and a Victorian Breast Cancer Research Consortium Group Leader.

OFBCR: The content of this manuscript does not necessarily reflect the views of the National Cancer Institute or any of the collaborating centers in the BCFR, nor does mention of trade names, commercial products, or organizations imply endorsement by the US Government or the BCFR.

Authors’ Contributions


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Provided MCCS data: K. Krishnan

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Common Breast Cancer Susceptibility Variants and Mammographic Density

References


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